



260115

FORM 7A

THE PATENTS ACT, 1970
(39 OF 1970)

AND

THE PATENTS RULES, 2003

REPRESENTATION FOR OPPOSITION TO GRANT OF PATENT
[See rule 55]

We, Indian Pharmaceutical Alliance, hereby give representation by way of opposition to the Grant of patent in respect of Application No. **9668/DELNP/2007** Dated **13/12/2007** made by **THE REGENTS OF THE UNIVERSITY OF CALIFORNIA** And published on **20/06/2008** on the grounds of Filing of Written Submission u/s 25(1) of the Act for Pre-Grant Opposition.

Our address for service in India is:-

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nayanrawal@gmail.com

NayanRawal
Constituted Attorney for
The Opponents
IN/PA 654

Dated- 7th January, 2016

To
The Controller of patents,
The patent office,
At Delhi



General Stamp Office, Mumbai.

L. S. Bamble No. 222 महाराष्ट्र MAHARASHTRA

-7 APR 2010

PROPER OFFICER

BHRI. L. S. BAMBLE

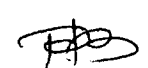
सी. काचिन हर्षद बागाळे

याद्री कोर्टगंभीर, अ. का. मार्ग

वादि (पूर्व), मुंबई - ५१.

CX 510202

श्री/श्रीमती/श्रीमान 15 APR 2010
Indian Pharmaceutical Alliance
यांना ले.....आधीचर मुद्रांक विपर विकला .


परधाना धारक मुद्रांक विक्रेता 20753

GENERAL POWER OF ATTORNEY

We, Indian Pharmaceutical Alliance, a Society registered under the Societies Registration Act having its mailing address as follows C/o VISION CONSULTING GROUP, 201 Darvesh Chambers, 743 P D Hinduja Road, Khar (W), Mumbai 400 052 hereby authorize Mr. Nayan J. Rawal, Patent Agent (Agent No.654), having its Office at L-303, Panchsheel Gardens, Mahavir Nagar, Kandivli West, Mumbai 400 067, to act as our Agents and Attorneys for various Pre-Grant and Post-Grant Opposition, rectification and any other proceedings under the Patents (Amendment) Act, 1970 in respect of various Patents filed in India.


We request that all notices, requisitions and communications may be sent to the said Agents at the following address:-

Vision Consulting Group, 201 Darvesh Chambers, 743 P.D Hinduja Road, Khar (W), Mumbai 400 052.

We hereby revoke all previous authorizations, if any; we hereby ratify all acts done by the said Agents

Dated this 19th day of April 2010

For Indian Pharmaceutical Alliance


D G Shah
Secretary General

**IN THE MATTER OF THE PATENTS ACT, 1970
(as amended by the Patents (Amendment) Act, 2005)**

and

**IN THE MATTER OF THE PATENT RULES, 2003
(as amended by the Patent (Amendment) Rules, 2006)**

and

**IN THE MATTER OF INDIAN PATENT APPLICATION NO. 9668/DELNP/2007 FILED BY
THE REGENTS OF THE UNIVERSITY OF CALIFORNIA**

.....THE APPLICANTS

and

**IN THE MATTER OF A REPRESENTATION BY WAY OF OPPOSITION UNDER SECTION
25(1) AND RULE 55 THERETO BY INDIAN PHARMACEUTICAL ALLIANCE**

.....THE OPPONENTS

REPRESENTATION BY WAY OF OPPOSITION U/S 25(1)

1.0 It is respectfully submitted on behalf of Indian Pharmaceutical Alliance, that a pre-grant Opposition under Section 25(1) of the Patents Act, 1970 and rule 55(1) of the Patents Rules, 2003 (as amended by the Patents (Amendment) Rules 2006), is hereby presented by the "Opponents" against Indian Patent Application No. **9668/DELNP/2007** (hereinafter also referred to as the "Opposed Application") in the name of **THE REGENTS OF THE UNIVERSITY OF CALIFORNIA** (hereinafter referred to as the "Applicants").

It is respectfully submitted:

2.0 The Opponents are an associations of person registered under the SOCIETIES REGISTRATION ACT, XXI OF 1860 in the name and style of "INDIAN PHARMACEUTICAL ALLIANCE" having its registered office 115/11, GROUND FLOOR, WORLD TRADE CENTRE, BABAR ROAD, CONNAUGHT PLACE, NEW DELHI – 110001, the main object of as follows:-

- (a) To support the development of international and regional policies, which seek to ensure, access to medical care for all customers.
- (b) To promote balanced and generic friendly intellectual property rights in the pharmaceuticals sector to ensure that timely access to markets is guaranteed for new and generic pharmaceutical products.
- (c) To promote the global harmonization relating to generic products.
- (d) To support the right of all governments to regulate their own pricing, substitution, prescribing and reimbursement policies.
- (e) To suggest measures for enhancing pharmaceutical research in India, both in the areas of basic as well as applied research.
- (f) To interact with the environmental protection agencies to evolve uniform standards of environmental protection measures across the country and ensure implementation of the same.

2.4 It is respectfully submitted that the Opposed Application entitled "*Diaryl thiohydantoin compounds*" has been filed on December 13, 2007 and published in the Official Journal of the Indian Patent office on June 20, 2008. The specification of Opposed Application is attached herewith as **Document 12 (D12)**.

2.5 The Opponents are filing this Representation by way of Opposition against Indian Patent Application No. 9668/DELNP/2007 (the Opposed Application), along with documentary evidence and facts in support thereof.

2.6 In this representation by way of opposition, the following grounds enumerated in Section 25 (1) of The Patents Act, 1970 are relied upon (hereinafter referred to as the "Act"):

(a) that the applicant for the patent or the person under or through whom he claims, wrongfully obtained the invention or any part thereof from him or from a person under or through whom he claims;

(b) that the invention so far as claimed in any claim of complete specification has been published before the priority date of the claim –

i) in any specification filed in pursuance of an application for a patent made in India on or after the 1st day of January, 1912; or

ii) in India or elsewhere, in any other document

Provided that the ground specified in sub-clause (ii) shall not be available where such publication does not constitute an anticipation of the invention by virtue of sub-section (2) or sub-section (3) of section 29;

(c) that the invention so far as claimed in any claim of the complete specification is claimed in a claim of a complete specification published on or after the priority date of the applicant's claim and filed in pursuance of an

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application for a patent in India, being a claim of which the priority date is earlier than that of the applicant's claim;

(d) that the invention so far as claimed in any claim of the complete specification was publicly known or publicly used in India before the priority date of the claim.

Explanation:- For the purpose of this clause, an invention relating to a process for which a patent is claimed shall be deemed to have been publicly known or publicly used in India before the priority date of the claim if a product made by that process had already been imported into India before that date except where such importation has been for the purpose of reasonable trial or experiment only;

(e) That the invention so far as claimed in any claim of the complete specification is obvious and clearly does not involve any inventive step having regard to the matter published as mentioned in clause (b) or having regard to what was used in India before the priority date of the applicant's claim;

(f) that the subject matter of any claim of the complete specification is not invention within the meaning of this Act, or is not patentable under this Act;

(g) that the complete specification does not sufficiently and clearly describe the invention or the method by which it is to be performed;

(h) that the applicant has failed to disclose to the Controller the information required by section 8 or has furnished the information which in any material particular was false to his knowledge;

(i) that in the case of convention application, the application was not made within twelve months from the date of the first application for protection for the invention made in a convention country by the applicant or a person from whom he derives title;

(j) that the complete specification does not disclose or wrongly mentions the source or geographical origin of biological material used for the invention;

(k) That the invention so far as claimed in any claim of the complete specification is anticipated having regard to the knowledge, oral or otherwise, available within any local or indigenous community in India or elsewhere;

The present Representation By Way of Opposition U/S 25(1) takes into consideration the following documents:

Document 1 [D1] – US 5411981, Date of Patent-May 02, 1995.

Document 2 [D2] – US 6087509, Date of Patent-July 11, 2000.

Document 3 [D3] – US 5434176, Date of Patent-July 18, 1995.

Document 4 [D4] – US 5750553, Date of Patent-May 12, 1998.

Document 5 [D5] - J Med Chem, 2000, 43, 3344-3347.

Document 6 [D6] - J Med Chem, 2005, 48, 917-925

Document 7 [D7] - J Steroid Biochem Mol Biol, 1994, 48, 111-119

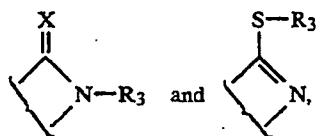
Document 8 [D8] - J Steroid Biochem Mol Biol, 1994, 51, 47-56

Document 9 [D9] - WO 97/00071, Date of publication-January 03, 1997.

Document 10 [D10] - US 2005/159468 (corresponding US publication of WO 2004/099188, Date of publication-November 18, 2004).

Document 11 [D11] – US 2006/135583 (corresponding US publication of WO 2004/111012, Date of publication-December 23, 2004).

Document 12 [D12] - Specification of Opposed Application.

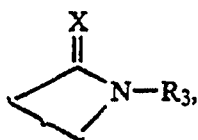


X is -O- or -S-

R3 is selected from the group consisting of hydrogen, alkyl, alkenyl and alkynyl of up to 12 carbon atoms, aryl and aralkyl of up to 12 carbon atoms, all optionally substituted with at least one member of the group consisting of -OH, halogen, -SH, -CN, acyl and acyloxy of up to 7 carbon atoms, -aryl, -O-aryl, -O-aralkyl -S-aryl of up to 12 carbon atoms, the aryl and aralkyl being optionally substituted with a member of the group consisting of halogen, -CF₃, alkyl, alkoxy, alkenyl, alkenyloxy, alkynyl and alkynyloxy with the sulfur being optionally oxidized to sulfone or sulfoxide, free, esterified, amidified or salified carboxy, -NH₂, mono and dialkylamino and heterocyclic of 3 to 6 ring members and containing at least one heteroatom selected from the group consisting of oxygen, sulfur and nitrogen, the alkyl, alkenyl and alkynyl being optionally interrupted with at least one member of the group consisting of oxygen, nitrogen and sulfur optionally oxidized to sulfoxide or sulfone, trialkylsilyl with the alkyl having 1 to 6 carbon atoms and acyl and acyloxy of an organic carboxylic acid of 1 to 7 carbon atoms and,

Y is -O-, -S- or -NH-, except the compounds,

wherein -A-B- is



X is oxygen,

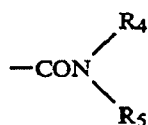
R3 is hydrogen and,

Y is oxygen or -NH-,

R2 is -CF₃ or halogen and,

R1 is -NO₂ or halogen and their non-toxic, pharmaceutically acceptable acid addition salts (cf column 1, lines 30-68-column 2, lines 1-15).

D1 further teaches that the amidified carboxy are of the type,



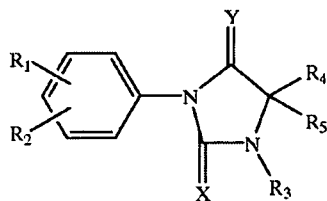
wherein R4 and R5 are individually selected from the group consisting of hydrogen and alkyl of 1 to 4 carbon atoms such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec.-butyl and tert.-butyl (cf column 3, lines 6-16).

Thus from the disclosure and teachings of D1 it is apparent that D1 explicitly discloses the compound of the impugned application. Therefore it is respectfully submitted that the alleged invention of the opposed application lacks novelty over the disclosure and teachings of D1.

Disclosure of D2

The prior art document D2 discloses and teaches 1-imidazolidinyl phenyl. D2 also discloses and teaches process for the preparation of the compounds disclosed and taught in D2 as well as pharmaceutical compositions, doses and pharmacological activity of the compounds disclosed and taught in D2.

D2 discloses and teaches 1-imidazolidinyl phenyls having following structural formula,



in which: R1 and R2, identical or different, are chosen from cyano, nitro, trifluoromethyl radicals and halogen atoms,

R3 represents a linear or branched aryl, arylalkyl, alkyl, alkenyl or alkynyl radical containing at most 10 carbon atoms and optionally substituted by one or more radicals chosen from halogen atoms and cyano, hydroxyl, alkoxy, carboxy, acyl and acyloxy radicals, in which, if appropriate, the alkyl, alkoxy and acyl radicals are linear or branched, containing at most 10 carbon atoms, the carboxy radical is free,

salified, esterified or amidified and the hydroxy radical is free, esterified, etherified or protected,

R4 and R5 identical or different, represent a linear or branched alkyl radical containing at most 4 carbon atoms and optionally substituted by a halogen atom, or form with the carbon atom to which they are linked a cyclic radical constituted by 3 to 7 members and optionally containing one or more identical or different heteroatoms, chosen from oxygen, sulphur or nitrogen atoms,

X and Y, identical or different, represent an oxygen or sulphur atom,

said products of formula (I) being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with mineral and organic acids or with mineral and organic bases of said products of formula (I) (cf column 1, lines 10-50).

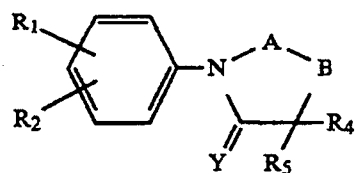
D2 further teaches that by amidified carboxy is meant the groups of -CON(R6)(R7) type in which the R6 and R7 radical identical or different represent a hydrogen atom or an alkyl radical having 1 to 4 carbon atoms such as the following radicals: methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl or tert-butyl. Among the -CON(R6)(R7) groups defined above, those in which the -N(R6)(R7) represents the amino, mono- or dimethylamino radical are preferred (cf column 3, lines 28-33).

Thus from the disclosure and teachings of D2 it is apparent that D2 explicitly discloses the compound of the impugned application. Therefore it is respectfully submitted that the alleged invention of the opposed application lacks novelty over the disclosure and teachings of D2.

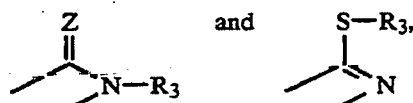
Disclosure of D3

The prior art document D3 discloses and teaches phenylimidazolidines. D3 also discloses and teaches process for the preparation of the compounds disclosed and taught in D3 as well as pharmaceutical compositions, doses and pharmacological activity of the compounds disclosed and taught in D3.

D3 discloses and teaches phenylimidazolidines having following structural formula,



wherein R1 and R2 are individually selected from the group consisting of -CN, -NO₂, halogen, -CF₃, free carboxy, salified carboxy and carboxy esterified with lower alkyl, -A-B- is selected from the group consisting of,



Z, is -O- or -S-,

R3 is selected from the group consisting of hydrogen, alkyl, alkenyl and alkynyl all of up to 12 carbon atoms, aryl and aralkyl of up to 12 carbon atoms, all optionally substituted with at least one member of the group consisting of -OH, halogen, -SH, -CN, acyl of up to 7 carbon atoms, acyloxy of up to 7 carbon atoms, -S- aryl of up to 12 carbon atoms optionally substituted with a member of the group consisting of -CF₃, alkyl, alkoxy, alkenyl, alkenyloxy, alkynyl and alkynyloxy, with the sulfur being optionally oxidized to sulfone or sulfoxide, free, esterified, amidified or salified carboxy, -NH₂, mono and dialkylamino and heterocyclic of 3 to 6 ring members and containing at least one heteroatom selected from the group consisting of oxygen, sulfur, and nitrogen, the alkyl, alkenyl, and alkynyl being optionally interrupted with at least one member of the group consisting of oxygen, nitrogen, and sulfur optionally oxidized to sulfoxide or sulfone,

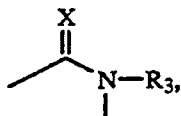
Y is -O-, -S- or =NH,

R4 and R5 are individually selected from the group consisting of hydrogen and alkyl of up to 12 carbon atoms optionally substituted with at least one halogen or, taken together with the carbon atom to which they are attached, form cycloalkyl of 3 to 7 carbon atoms except the compounds,

wherein R4 and R5 are both methyl or one is hydroxymethyl,

Y is -O- or =NH,

-A-B- is



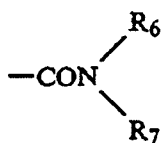
X is oxygen,

R3 is hydrogen,

R1 is 4-NO₂ and

R2 is 3-CF₃; and their non-toxic, pharmaceutically acceptable acid addition salts (cf column 1, lines 28-68-column 2, lines 1-17).

D3 further teaches that the amidified carboxy are of the type,



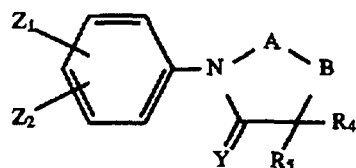
wherein R6 and R7 are individually selected from the group consisting of hydrogen and alkyl of 1 to 4 carbon atoms such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec.-butyl and tert-butyl (cf column 3, lines 9-20).

Thus from the disclosure and teachings of D3 it is apparent that D3 explicitly discloses the compound of the impugned application. Therefore it is respectfully submitted that the alleged invention of the opposed application lacks novelty over the disclosure and teachings of D3.

Disclosure of D4

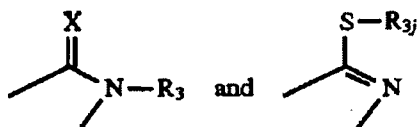
The prior art document D4 discloses and teaches phenylimidazolidines. D4 also discloses and teaches process for the preparation of the compounds disclosed and taught in D4 as well as pharmaceutical compositions, doses and pharmacological activity of the compounds disclosed and taught in D4.

D4 discloses and teaches phenylimidazolidines having following structural formula,



in which: Z1 and Z2, identical or different, represent a cyano, nitro radical, a halogen atom, a trifluoromethyl radical or an esterified, amidified or salified free carboxy radical,

the group -A-B- is chosen from the radicals



in which X represents an oxygen or sulphur atom,

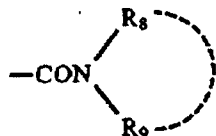
R3j represents R3 with the exception of the hydrogen value and R3 is chosen from the following radicals:

a hydrogen atom,

alkyl, alkenyl, alkynyl, aryl or arylalkyl radicals having at most 12 carbon atoms, these radicals being optionally substituted by one or more substituents chosen from halogen atoms and the following radicals: optionally esterified, etherified or protected hydroxy, alkoxy, hydroxyalkyl, alkenyloxy, alkynyloxy, trifluoromethyl, mercapto, cyano, acyl, acyloxy, aryl, optionally substituted S-alkyl, S-aryl, in which the sulphur atom is optionally oxidized in the form of the sulphoxide or sulphone, free, esterified, amidified or salified carboxy, amino, mono- or dialkylamino, a cyclic radical containing 3 to 6 members and optionally containing one or more heteroatoms chosen from sulphur, oxygen or nitrogen atoms

Y represents an oxygen or sulphur atom or an NH radical (cf column 1, lines 23-61).

D4 further teaches that by amidified carboxy is meant the groups of



type in which the R8 and R9 radicals, identical or different, represent a hydrogen atom or an alkyl radical having 1 to 4 carbon atoms such as the methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl or tert-butyl radicals (cf column 4, lines 30-40).

Thus from the disclosure and teachings of D4 it is apparent that D4 explicitly discloses the compound of the impugned application. Therefore it is respectfully submitted that the alleged invention of the opposed application lacks novelty over the disclosure and teachings of D4.

Therefore it is respectfully submitted that the invention allegedly claimed in 1-48 claims of the opposed application lacks novelty in view of the single prior art document D2, D3, D4 and D5.

4.0 LACK OF INVENTIVE STEP-Section 25(1)(e)

It is respectfully submitted that the opponent has already established that the impugned invention is not patentable and lacks novelty under Section 25(1)(b). Even though if the Ld. Controller does not agree with the opponent for the reasons and statements made above, the opponent respectfully requests the Ld. Controller to separately look into objections raised under Section 25(1)(e) and consider them too.

In connection with the above mentioned ground of opposition, the opponent would rely upon the following documents, which were available to the public prior to the priority date of the opposed application:

Document 1 [D1] – US 5411981, Date of Patent-May 02, 1995.

Document 2 [D2] – US 6087509, Date of Patent-July 11, 2000.

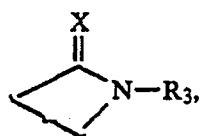
Document 3 [D3] – US 5434176, Date of Patent-July 18, 1995.

Document 4 [D4] – US 5750553, Date of Patent-May 12, 1998.

consisting of oxygen, nitrogen and sulfur optionally oxidized to sulfoxide or sulfone, trialkylsilyl with the alkyl having 1 to 6 carbon atoms and acyl and acyloxy of an organic carboxylic acid of 1 to 7 carbon atoms and,

Y is -O-, -S- or -NH-, except the compounds,

wherein -A-B- is



X is oxygen,

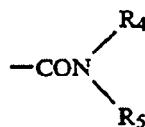
R₃ is hydrogen and,

Y is oxygen or -NH-,

R₂ is -CF₃ or halogen and,

R₁ is -NO₂ or halogen and their non-toxic, pharmaceutically acceptable acid addition salts (cf column 1, lines 30-68-column 2, lines 1-15).

D1 further teaches that the amidified carboxy are of the type,



wherein R₄ and R₅ are individually selected from the group consisting of hydrogen and alkyl of 1 to 4 carbon atoms such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec.-butyl and tert.-butyl (cf column 3, lines 6-16).

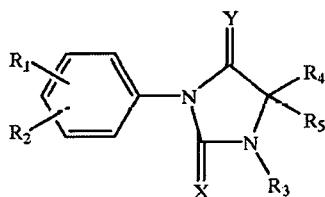
Thus from the disclosure and teachings of D1 it is apparent that D1 explicitly teaches the compound of the impugned application. Therefore it is respectfully submitted that the alleged invention of the opposed application lacks inventive step over the disclosure and teachings of D1.

Teachings of D2

The prior art document D2 discloses and teaches 1-imidazolidinyl phenyl. D2 also discloses and teaches process for the preparation of the compounds disclosed and

taught in D2 as well as pharmaceutical compositions, doses and pharmacological activity of the compounds disclosed and taught in D2.

D2 discloses and teaches 1-imidazolidinyl phenyls having following structural formula,



in which: R1 and R2, identical or different, are chosen from cyano, nitro, trifluoromethyl radicals and halogen atoms,

R3 represents a linear or branched aryl, arylalkyl, alkyl, alkenyl or alkynyl radical containing at most 10 carbon atoms and optionally substituted by one or more radicals chosen from halogen atoms and cyano, hydroxyl, alkoxy, carboxy, acyl and acyloxy radicals, in which, if appropriate, the alkyl, alkoxy and acyl radicals are linear or branched, containing at most 10 carbon atoms, the carboxy radical is free, salified, esterified or amidified and the hydroxy radical is free, esterified, etherified or protected,

R4 and R5 identical or different, represent a linear or branched alkyl radical containing at most 4 carbon atoms and optionally substituted by a halogen atom, or form with the carbon atom to which they are linked a cyclic radical constituted by 3 to 7 members and optionally containing one or more identical or different heteroatoms, chosen from oxygen, sulphur or nitrogen atoms,

X and Y, identical or different, represent an oxygen or sulphur atom,

said products of formula (I) being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with mineral and organic acids or with mineral and organic bases of said products of formula (I) (cf column 1, lines 10-50).

D2 further teaches that by amidified carboxy is meant the groups of -CON(R6)(R7) type in which the R6 and R7 radical identical or different represent a hydrogen atom

alkyl, alkoxy, alkenyl, alkenyloxy, alkynyl and alkynyloxy, with the sulfur being optionally oxidized to sulfone or sulfoxide, free, esterified, amidified or salified carboxy, -NH₂, mono and dialkylamino and heterocyclic of 3 to 6 ring members and containing at least one heteroatom selected from the group consisting of oxygen, sulfur, and nitrogen, the alkyl, alkenyl, and alkynyl being optionally interrupted with at least one member of the group consisting of oxygen, nitrogen, and sulfur optionally oxidized to sulfoxide or sulfone,

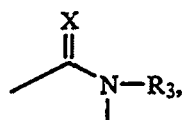
Y is -O-, -S- or =NH,

R4 and R5 are individually selected from the group consisting of hydrogen and alkyl of up to 12 carbon atoms optionally substituted with at least one halogen or, taken together with the carbon atom to which they are attached, form cycloalkyl of 3 to 7 carbon atoms except the compounds,

wherein R4 and R5 are both methyl or one is hydroxymethyl,

Y is -O- or =NH,

-A-B- is



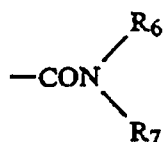
X is oxygen,

R3 is hydrogen,

R1 is 4-NO₂ and

R2 is 3-CF₃; and their non-toxic, pharmaceutically acceptable acid addition salts (cf column 1, lines 28-68-column 2, lines 1-17).

D3 further teaches that the amidified carboxy are of the type,



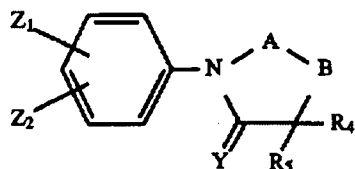
wherein R6 and R7 are individually selected from the group consisting of hydrogen and alkyl of 1 to 4 carbon atoms such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec.-butyl and tert-butyl (cf column 3, lines 9-20).

Thus from the disclosure and teachings of D3 it is apparent that D3 explicitly teaches the compound of the impugned application. Therefore it is respectfully submitted that the alleged invention of the opposed application lacks inventive step over the disclosure and teachings of D3.

Teachings of D4

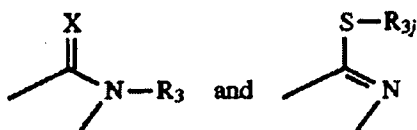
The prior art document D4 discloses and teaches phenylimidazolidines. D4 also discloses and teaches process for the preparation of the compounds disclosed and taught in D4 as well as pharmaceutical compositions, doses and pharmacological activity of the compounds disclosed and taught in D4.

D4 discloses and teaches phenylimidazolidines having following structural formula,



in which: Z1 and Z2, identical or **different**, represent a **cyano**, nitro radical, a halogen atom, a **trifluoromethyl radical** or an esterified, amidified or salified free carboxy radical,

the group -A-B- is chosen from the radicals



in which X represents an oxygen or **sulphur atom**,

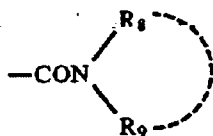
R3j represents R3 with the exception of the hydrogen value and R3 is chosen from the following radicals:

a hydrogen atom, alkyl, alkenyl, alkynyl, **aryl** or arylalkyl radicals having at most 12 carbon atoms, these radicals being optionally **substituted** by one or more substituents chosen from **halogen atoms** and the following radicals: optionally esterified, etherified or protected hydroxy, alkoxy, hydroxyalkyl, alkenyloxy, alkynyloxy, trifluoromethyl, mercapto, cyano, acyl, acyloxy, aryl, optionally

substituted S-alkyl, S-aryl, in which the sulphur atom is optionally oxidized in the form of the sulphoxide or sulphone, free, esterified, **amidified** or salified **carboxy**, amino, mono- or dialkylamino, a cyclic radical containing 3 to 6 members and optionally containing one or more heteroatoms chosen from sulphur, oxygen or nitrogen atoms

Y represents an **oxygen** or sulphur atom or an NH radical (cf column 1, lines 23-61).

D4 further teaches that by amidified carboxy is meant the groups of



type in which the R8 and R9 radicals, identical or **different**, represent a **hydrogen** atom or an **alkyl radical having 1 to 4 carbon atoms such as the methyl**, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl or tert-butyl radicals (cf column 4, lines 30-40).

Thus from the disclosure and teachings of D4 it is apparent that D4 explicitly teaches the compound of the impugned application. Therefore it is respectfully submitted that the alleged invention of the opposed application lacks inventive step over the disclosure and teachings of D4.

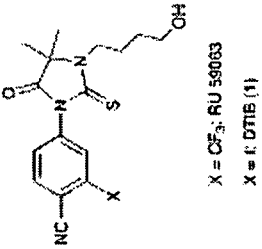
Several other Prior art documents

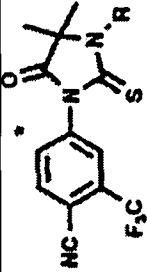
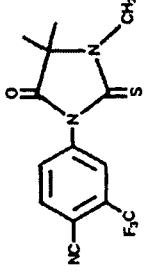
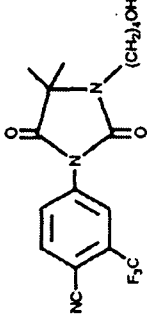
In connection with the submissions made above, the opponent would also like to rely upon several other prior art documents which teach substantially the same subject matter allegedly claimed in the opposed application.

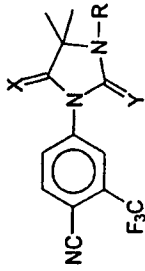
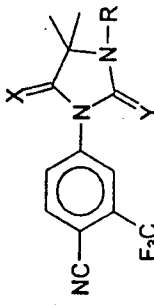
The opponent would like to invite kind attention of the Ld. Controller towards following documents which were available to the public before the priority date of the impugned application.

1. J Med Chem, 2000, 43, 3344-3347 (D5)
2. J Med Chem, 2005, 48, 917-925 (D6)
3. J Steroid Biochem Mol Biol, 1994, 48, 111-119 (D7)

Tabular representation of the disclosure and teachings of above mentioned prior art documents is provided below.

Prior art document	Disclosure and teachings contained therein	Conclusion
<p>J Med Chem, 2000, 43, 3344-3347 (D5)</p>	<p>D5 teaches compounds of following general formula as high-affinity nonsteroidal androgen receptor ligand,</p>  <p>X = CF₃, RU 59063 X = 4-OTIB (1)</p>	<p>The applicant at the priority date of the impugned application was very well aware that which scaffold possess ant androgenic activity. Thus he will use the scaffold disclosed and taught in D5 for further modification in order to identify other potent candidates.</p>
<p>J Med Chem, 2005, 48, 917-925 (D6)</p>	<p>D6 teaches Three-Dimensional Structure-Activity Relationships of Nonsteroidal Ligands in Complex with Androgen Receptor Ligand-Binding Domain. Thus D6 teaches what kind of modifications can result into search of other potent candidates as compared to earlier known compounds showing similar activity.</p>	<p>From D6 the applicant is well aware which kind of compounds can show potent antiandrogenic activity and what kind of modification can be done in order to identify other potent candidates.</p>
<p>J Steroid Biochem Mol Biol, 1994, 48, 111-119 (D7)</p>	<p>D7 teaches following compounds,</p>	<p>From D7 the applicant was well aware</p>

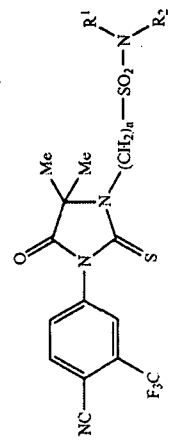
<p>J Steroid Biochem Mol Biol, 1994, 51, 47-56 (D8)</p>	 <p>a) R = CH₃ b) R = CH₂CH₂OH c) R = (CH₂)₄OH</p>	<p>about the modification site in the scaffold taught in D5 as well as D7. Therefore he will reasonably be motivated to modify that site only in order to find out other potent candidates showing antiandrogenic activity.</p>
<p>D8 teaches preliminary pharmacokinetics and metabolism of some non-steroidal antiandrogens having following structural formula,</p>  <p>RU 56187</p>  <p>RU 58841</p>	<p>D8 teaches preliminary pharmacokinetics and metabolism of some non-steroidal antiandrogens having following structural formula,</p>	<p>Thus at the priority date of the impugned application the applicant was very well aware regarding the pharmacokinetics and metabolism of the known antiandrogen compounds and their biological profile. Therefore he will be motivated to modify the known scaffold accordingly to identify other potent antiandrogens.</p>

<p>WO 97/00071 (D9)</p>	<p>D9 teaches compounds of following formula where X, Y and R are defined in the description.</p> 	<p>From D9 the applicant was aware that if he wants to modify the known scaffold of D5-D9 where he has to apply modifications. Further, from D1-D4 he was also well aware what kind of modifications can be advantageous. Thus he will do those modifications only in order to identify other potent antiandrogen compounds.</p>
<p>US 2005/159468 (D10)</p>	<p>D10 teaches compounds of following general formula where substitutions are defined therein,</p> 	<p>From D10 the applicant was aware that if he wants to modify the known scaffold of D5-D10 where he has to apply modifications. Further, from D1-D4 he was also well aware what kind of modifications can be advantageous. Thus he will do those modifications only in order to identify other potent antiandrogen compounds.</p>

11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41 42 43 44 45 46 47 48 49 50

US 2006/135583 (D11)

D11 teaches compounds of the following general formula where the substitutions are defined in the description part of D11.



From D11 the applicant was aware that if he wants to modify the known scaffold of D5-D11 where he has to apply modifications. Further, from D1-D4 he was also well aware what kind of modifications can be advantageous. Thus he will do those modifications only in order to identify other potent antiandrogen compounds.

5.0 NON-PATENTABLE SUBJECT MATTER-Section 25(1)(f)

Section 3(d)

According to Section 3(d) of the Act, "the mere discovery of a new form of a known substance which does not result in the enhancement of the known efficacy of that substance or the mere discovery of any new property or new use for a known substance or of the mere use of a known process, machine or apparatus unless such known process results in a new product or employs at least one new reactant."

Explanation.—For the purposes of this clause, salts, esters, ethers, polymorphs, metabolites, pure form, particle size, isomers, mixtures of isomers, complexes, combinations and other derivatives of known substance shall be considered to be the same substance, unless they differ significantly in properties with regard to efficacy;

Claimed compound is not patentable under Section 3(d) of the Act

The compound allegedly claimed in the opposed application is not patentable under Section 3(d) of the Patents Act, 1970 as amended by the Patents (Amendment) Act, 2005 as it is a new form of known substances. The compound claimed in the opposed application is derivative of the earlier known compounds and the applicant of the opposed application has not provided enhancement in the known therapeutic efficacy. In absence of such data the compound claimed in the opposed application does not qualify for patentable invention under Section 3(d) of the Act.

Section 3(e)

According to Section 3(e) of the Indian Patent Act, "a substance obtained by a mere admixture resulting only in the aggregation of the properties of the components thereof or a process for producing such substance." is not patentable.

Claim 3 of the opposed application is not patentable under Section 3(e) of the Act

The pharmaceutical composition allegedly claimed in Claim 3 of the opposed application is not patentable under Section 3(e) of the Patents Act, 1970 as amended by the Patents (Amendment) Act, 2005 as it is a mere admixture of two or more substances resulting only in the aggregation of the properties of the components. The applicant of the opposed application has not provided any surprising or superior effect of the claimed composition.

Claim 4 of the opposed application is not patentable under Section 3(e) of the Act

The pharmaceutical composition allegedly claimed in Claim 4 of the opposed application is not patentable under Section 3(e) of the Patents Act, 1970 as amended by the Patents (Amendment) Act, 2005 as it is a mere admixture of two or more substances resulting only in the aggregation of the properties of the components. The applicant of the opposed application has not provided any surprising or superior effect of the claimed composition.

Claim 5 of the opposed application is not patentable under Section 3(e) of the Act

The pharmaceutical composition allegedly claimed in Claim 5 of the opposed application is not patentable under Section 3(e) of the Patents Act, 1970 as amended by the Patents (Amendment) Act, 2005 as it is a mere admixture of two or more substances resulting only in the aggregation of the properties of the components. The applicant of the opposed application has not provided any surprising or superior effect of the claimed composition.

Claim 6 of the opposed application is not patentable under Section 3(e) of the Act

The pharmaceutical composition allegedly claimed in Claim 6 of the opposed application is not patentable under Section 3(e) of the Patents Act, 1970 as amended by the Patents (Amendment) Act, 2005 as it is a mere admixture of two or more substances resulting only in the aggregation of the properties of the components. The applicant of the opposed application has not provided any surprising or superior effect of the claimed composition.

Section 3(i)

According to Section 3(i) of the Indian Patent Act, "any process for the medicinal, surgical, curative, prophylactic diagnostic, therapeutic or other treatment of human beings or any process for a similar treatment of animals to render them free of disease or to increase their economic value or that of their products." is not patentable.

Claim 2 of the opposed application is not patentable under Section 3(i) of the Act

It is respectfully submitted that Claim 2 of the opposed application is not patentable under Section 3(i) of the Patents Act, 1970 as amended by the Patents (Amendment) Act, 2005 as Claim 2 of the opposed application essentially claims method of treatment in guise of the compound claim which is not allowable under Section 3(i) of the Act.

7.0 LACK OF CLAIRTY AND SUFFICIENCY-Section 25(1)(g)

It is respectfully submitted that the applicant of the opposed application has not provided any example illustrating method of treatment allegedly claimed in Claim 2 of the opposed application. Thus the specification of the opposed application does not clearly and sufficiently describe the present invention and does not enable the reader to practice the invention to its fullest.

It is respectfully submitted that the applicant of the opposed application has not provided any example illustrating pharmaceutical composition allegedly claimed in Claims 3-14 of the opposed application. Thus the specification of the opposed application does not clearly and sufficiently describe the present invention and does not enable the reader to practice the invention to its fullest.

CONCLUSION

12.0 In view of the submissions presented above, we humbly pray that:

- i) the Indian Patent Application No. 9668/DELNP/2007 be dismissed *in toto*;
- ii) the opponent be granted leave to file further evidence;
- iii) the copy of reply statement and evidence filed by the applicant in the response to this opposition be made available to the opponent;
- iv) the opponent be permitted to file further response and evidence to the reply or evidence produced by the applicant;
- v) the opponent be granted leave to make further submissions in case the applicant makes any amendments in the claims;
- vi) any other relief as the Learned Controller may deem fit be awarded in favor of the opponents.

As a matter of precaution we request the Learned Controller to grant us an oral hearing before disposing of this representation.

Dated this the 7th day of Jan, 2016



Nayan J. Rawal

Constituted Attorney for
the Opponent

IPA no. 654

D1

9668/DELNP/2007



US005411981A

United States Patent [19]
Gaillard-Kelly et al.

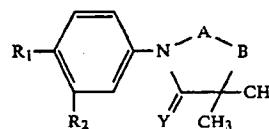
[11] **Patent Number:** 5,411,981
[45] **Date of Patent:** May 2, 1995

- [54] **PHENYLIMIDAZOLIDINES HAVING ANTIANDROGENIC ACTIVITY**
- [75] **Inventors:** Martine Gaillard-Kelly; Francois Goubet, both of Paris; Daniel Philibert, La Verenne Saint Hilaire; Jean-Georges Teutsch, Pantin, all of France
- [73] **Assignee:** Roussel Uclaf, France
- [21] **Appl. No.:** 64,257
- [22] **Filed:** May 18, 1993

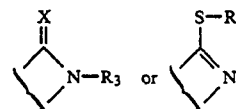
Primary Examiner—Floyd D. Higel
Attorney, Agent, or Firm—Bierman and Muserlian

[57] **ABSTRACT**

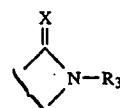
A compound of the formula



wherein R₁ is —CN, —NO₂ or halogen, R₂ is —CF₃ or halogen, —A—B— is of



X is —O— or —S—, R₃ is hydrogen, alkyl, alkenyl or alkynyl of up to 12 carbon atoms, aryl and aralkyl of up to 12 carbon atoms, all optionally substituted by —OH, halogen, —SH, —CN, acyl and acyloxy of up to 7 carbon atoms, —aryl, —O—aryl, —O—aralkyl —S— aryl of up to 12 carbon atoms the aryl and aralkyl being optionally substituted by halogen, —CF₃, alkyl, alkoxy, alkenyl, alkenyloxy, alkynyl or alkynyloxy with the sulfur being optionally oxidized to sulfone or sulfoxide, free, esterified, amidified or salified carboxy, —NH₂, mono and dialkylamino and heterocyclic of 3 to 6 ring members and containing at least one heteroatom selected from the group consisting of oxygen, sulfur and nitrogen, the alkyl, alkenyl and alkynyl being optionally interrupted by at least one oxygen, nitrogen or sulfur optionally oxidized to sulfoxide or sulfone, trialkylsilyl with the alkyl having 1 to 6 carbon atoms and acyl and acyloxy of an organic carboxylic acid of 1 to 7 carbon atoms and Y is —O—, —S— or —NH—, except the compounds wherein —A—B— is



X is oxygen, R₃ is hydrogen and Y is oxygen or —NH—, R₂ is —CF₃ or halogen and R₁ is —NO₂ or halogen and their non-toxic, pharmaceutically acceptable acid addition salts.

20 Claims, No Drawings

Related U.S. Application Data

- [63] Continuation-in-part of Ser. No. 819,910, Jan. 9, 1992, abandoned.

[30] **Foreign Application Priority Data**

Jan. 9, 1991 [FR]	France	91 00185
Jul. 8, 1992 [FR]	France	92 08431

- [51] **Int. Cl.⁶** C07D 233/72; A61K 31/415
- [52] **U.S. Cl.** 514/386; 514/391; 514/342; 548/311.1; 548/317.1; 548/318.5; 548/320.1; 548/320.5
- [58] **Field of Search** 514/391, 386, 342; 548/317.1, 318.5, 320.1, 320.5, 311.1

[56] **References Cited**

U.S. PATENT DOCUMENTS

3,823,240	7/1974	Sauli et al.	514/391
4,097,578	6/1978	Perronet et al.	548/317.1
4,234,736	11/1980	Bernauer et al.	548/317.1
4,304,782	12/1981	Dymont et al.	548/321.1 X
4,407,814	10/1983	Bernauer et al.	548/317.1
4,427,438	1/1984	Nagano et al.	548/317.1
4,473,393	10/1984	Nagpal	548/321.1 X
4,482,739	11/1984	Bernauer et al.	564/54
4,749,403	6/1988	Liebl et al.	548/321.1 X
4,859,228	8/1989	Prisbylla	548/321.1 X
4,873,256	10/1989	Coussediere et al.	548/317.1
4,944,791	7/1990	Schroder et al.	548/321.1 X
5,069,711	12/1991	Fischer et al.	548/317.1 X

FOREIGN PATENT DOCUMENTS

0217893	6/1958	Australia	548/321.1
0017976	10/1980	European Pat. Off.	548/317.1
2102605	7/1971	Germany	548/321.1
48-87030	11/1973	Japan	548/317.1

OTHER PUBLICATIONS

- Raynaud et al., *J. Steroid Biochem.*, vol. 11, pp. 93-99 (1979).
- Rao et al., *J. Steroid Biochem.*, vol. 31, pp. 731-737 (1988).

PHENYLIMIDAZOLIDINES HAVING ANTIANDROGENIC ACTIVITY

PRIOR APPLICATION

This application is a continuation-in-part of U.S. Patent application Ser. No. 819,910, filed Jan. 9, 1992, now abandoned.

Japanese application No J 48087030 describes 3-phenyl-2-thiohydantoin useful for inhibiting the germination of certain plants. U.S. Pat. No. 4,097,578 describes imidazolidines different from formula I having antiandrogenic activity. Other pertinent art includes U.S. Pat. Nos. 3,823,240; No. 4,873,256; No. 4,407,814; No. 4,482,739 and No. 4,234,736.

OBJECTS OF THE INVENTION

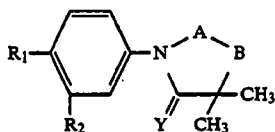
It is an object of the invention to provide the novel compounds of formula I and a novel process and novel intermediates for their preparation.

It is another object of the invention to provide novel anti-androgenic compositions and a novel method of inducing anti-androgenic activity in warm-blooded animals.

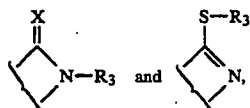
These and other objects and advantages of the invention will become obvious from the following detailed description.

THE INVENTION

The novel phenylimidazolidines of the invention have the formula

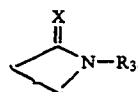


wherein R_1 is selected from the group consisting of $-\text{CN}$, $-\text{NO}_2$ and halogen, R_2 is $-\text{CF}_3$ or halogen, $-\text{A}-\text{B}-$ is selected from the group consisting of



X is $-\text{O}-$ or $-\text{S}-$, R_3 is selected from the group consisting of hydrogen, alkyl, alkenyl and alkynyl of up to 12 carbon atoms, aryl and aralkyl of up to 12 carbon atoms, all optionally substituted with at least one member of the group consisting of $-\text{OH}$, halogen, $-\text{SH}$, $-\text{CN}$, acyl and acyloxy of up to 7 carbon atoms, $-\text{aryl}$, $-\text{O}-\text{aryl}$, $-\text{O}-\text{aralkyl}$ $-\text{S}-\text{aryl}$ of up to 12 carbon atoms, the aryl and aralkyl being optionally substituted with a member of the group consisting of halogen, $-\text{CF}_3$, alkyl, alkoxy, alkenyl, alkenyloxy, alkynyl and alkynyloxy with the sulfur being optionally oxidized to sulfone or sulfoxide, free, esterified, amidified or sulfated carboxy, $-\text{NH}_2$, mono and dialkylamino and heterocyclic of 3 to 6 ring members and containing at least one heteroatom selected from the group consisting of oxygen, sulfur and nitrogen, the alkyl, alkenyl and alkynyl being optionally interrupted with at least one member of the group consisting of oxygen, nitrogen and sulfur optionally oxidized to sulfoxide or sulfone, trialkylsilyl

with the alkyl having 1 to 6 carbon atoms and acyl and acyloxy of an organic carboxylic acid of 1 to 7 carbon atoms and Y is $-\text{O}-$, $-\text{S}-$ or $-\text{NH}-$, except the compounds wherein $-\text{A}-\text{B}-$ is



X is oxygen, R_3 is hydrogen and Y is oxygen or $-\text{NH}-$, R_2 is $-\text{CF}_3$ or halogen and R_1 is $-\text{NO}_2$ or halogen and their non-toxic, pharmaceutically acceptable acid addition salts.

The following examples are given for the values of R_3 . Alkyl of up to 12 carbon atoms includes methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec.-butyl, tert.-butyl, pentyl, isopentyl, sec.-pentyl, tert.-pentyl, neopentyl, hexyl, isohexyl, sec.-hexyl, tert.-hexyl, heptyl, octyl, decyl, undecyl and dodecyl, branched or linear. Preferred are alkyl of 1 to 6 carbon atoms, especially methyl, ethyl, propyl and isopropyl, n-butyl, isobutyl, tert-butyl and branched or linear pentyl and hexyl.

Examples of alkenyl of up to 12 carbon atoms are vinyl, allyl, 1-propenyl, butenyl, pentenyl and hexenyl and preferably alkenyl of 2 to 4 carbon atoms and especially vinyl, allyl or butenyl. Examples of alkynyl of up to 12 carbon atoms are ethynyl, propargyl, butynyl, pentynyl and hexynyl and preferably 2 to 4 carbon atoms such as ethynyl and propargyl.

Examples of aryl are carbocyclic aryl such as phenyl and naphthyl, heterocyclic aryl of 5 to 6 ring members containing at least one heteroatom selected from the group consisting of oxygen, sulfur and nitrogen. Examples of 5 ring heteroaryls are furyl, thienyl, pyrrolyl, thiazolyl, oxazolyl, imidazolyl, thiadiazolyl, pyrazolyl and isoxazolyl. Examples of 6 ring heteroaryl are pyridyl, pyrimidinyl, pyridazinyl and pyrazinyl. Examples of condensed aryls are indolyl, benzofuranyl, benzothienyl and quinoleinyl. The preferred aryl is phenyl.

Examples of aralkyl include the alkyl recited above substituted with the aryl cited above. The preferred aralkyl are triphenylmethyl, phenethyl and benzyl. Examples of halogen are fluorine, chlorine, bromine and iodine but preferred are fluorine, chlorine and bromine. Examples of alkyl substituted with at least one halogen are fluoromethyl, chloromethyl, bromomethyl, iodomethyl, difluoromethyl, dichloromethyl, dibromomethyl and trifluoromethyl.

Examples of substituents for aryl and aralkyl are phenyl substituted by fluorine, $-\text{OCH}_3$ or $-\text{CF}_3$ in the p-position.

Examples of acyl are preferably those of up to 7 carbon atoms such as acetyl, propionyl, butyryl and benzoyl as well as valeryl, hexanoyl, acryloyl, crotonoyl, carbamoyl or formyl. The acyloxy may be derived for the same acids, especially acetyloxy and propionyloxy.

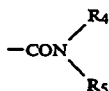
The esterified carboxy may be alkoxycarbonyl such as methoxycarbonyl, ethoxycarbonyl, propoxycarbonyl, butoxycarbonyl, tert.-butoxycarbonyl, cyclobutyloxy carbonyl, cyclopentylloxy carbonyl and cyclohexyloxy carbonyl.

Examples of easily cleavable esters includes methoxymethyl, ethoxymethyl; acyloxyalkyl such as pivaloyloxymethyl, pivaloyloxyethyl, acetoxymethyl and acetoxyethyl; alkoxycarbonyloxyalkyl such as methoxycar-

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bonyloxymethyl, methoxycarbonyloxyethyl, isopropoxycarbonyloxymethyl and isopropoxycarbonyloxyethyl. Other esters are described in European Patent No. 0.034.536.

The amidified carboxy are of the type



wherein R₄ and R₅ are individually selected from the group consisting of hydrogen and alkyl of 1 to 4 carbon atoms such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec.-butyl and tert.-butyl.

Examples of the mono and dialkylamino are methylamino, ethylamino, dimethylamino, diethylamino and methylethylamino. The hetero-cyclic of 5 to 6 ring members optionally containing another heteroatom may be pyrrolyl, imidazolyl, pyridyl, pyrazinyl, pyrimidyl, indolyl, piperidino, morpholino and piperazinyl, preferably piperidino or morpholino.

Examples of salts of salified carboxy are sodium, potassium, lithium, calcium, magnesium, ammonium and organic bases such as methylamine, propylamine, trimethylamine, diethylamine and triethylamine. Sodium salt is preferred.

The alkylamino and dialkylamino are preferably alkyl of 1 to 4 carbon atoms such as methylamino, ethylamino, propylamino, isopropylamino, dimethylamino, diethylamino and ethylmethylamino.

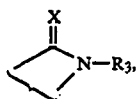
Examples of the heterocyclics containing at least one heteroatom are the saturated monocyclics such as oxiranyl, oxolannyl, dioxolannyl, imidazolidinyl, pyrazolidinyl, piperidyl, piperazinyl and morpholinyl.

The alkyl, alkenyl and alkynyl may be optionally interrupted by one or more sulfur, oxygen or nitrogen heteroatoms. Examples are alkoxyalkyl such as methoxymethyl, methoxyethyl, methoxypropyl or methoxybutyl or alkoxy alkoxyalkyl such as methoxyethoxymethyl.

Examples of trialkylsilyl groups are trimethylsilyl, triethylsilyl and (1,1-dimethylethyl) dimethylsilyl.

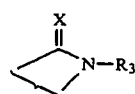
When the products of formula I contain a salifiable amino group, the acid addition salts of non-toxic, pharmaceutically acceptable acids may be formed. Examples of said acids are inorganic acids such as nitric acid, hydrochloric acid, sulfuric acid and phosphoric acid and organic acids such as formic acid, acetic acid, propionic acid, benzoic acid and methane sulfonic acid.

Among the preferred compounds of formula I are those wherein Y is oxygen except for the compounds wherein —A—B— is



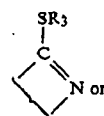
X is oxygen, R₃ is hydrogen, R₂ is —CF₃ or halogen and R₁ is —NO₂ or halogen. Other preferred compounds of formula I are those wherein —A—B— is

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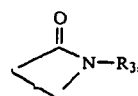


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X is sulfur and R₃ has the above definition, those wherein R₃ is hydrogen or alkyl of 1 to 4 carbon atoms optionally substituted with —OH or methoxy, those wherein R₁ is cyano or halogen, preferably chlorine and those wherein —A—B— is



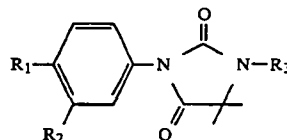
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and R₃ is optionally substituted alkyl or alkenyl of up to 6 carbon atoms and optionally interrupted by oxygen or optionally oxidized sulfur or optionally substituted aralkyl, acyl or trialkylsilyl.

Other preferred examples of the invention are those in which R₃ is alkyl of up to 6 carbon atoms optionally substituted by at least one member of the group consisting of halogen, free or esterified hydroxy or carboxy, heterocyl, O-aralkyl or S-aryl in which the aryl radical is optionally substituted by at least one halogen or alkoxy and the sulfur atom is optionally oxidized in the form of the sulfoxide or sulfone and quite particularly those in which R₃ is alkyl of 2 to 4 carbon atoms substituted by a member of the group consisting of chlorine, ethoxycarbonyl, tertbutoxycarbonyl, cyclopentyl-oxycarbonyl, 4-fluorophenylthio optionally oxidized in the form of the sulfoxide or sulfone, morpholino, phenylmethoxy, triphenylmethoxy and methylsulfonyloxy.

Other preferred compounds of formula I are those wherein R₃ is acetyl or benzoyl or (1,1-dimethylethyl) dimethylsilyl, those wherein R₁ is nitro and R₃ is alkyl or alkenyl of up to 4 carbon atoms optionally substituted with esterified or salified or free carboxy and those of the formula



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in which R₁, R₂ and R₃ have the above meaning with the exception of the products in which R₁ is nitro, R₂ is trifluoromethyl and R₃ is hydrogen.

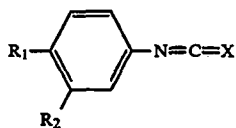
60 Examples of specific preferred compounds of formula I are 4-(5-oxo-2-thioxo-3,4,4-trimethyl-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile, 4-(4,4-dimethyl-5-oxo-2-thioxo-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile, 4-[4,4-dimethyl 3-(2-hydroxyethyl)-5-oxo-2-thioxo-1-imidazolidinyl]-2-(trifluoromethyl) benzonitrile, 3-(3,4-dichlorophenyl)-2-thioxo-1,5,5-trimethyl-4-imidazolidinone, 1-(4-nitro-3-(trifluoromethyl)-phenyl)-3,4,4-trimethyl-2,5-imidazol-

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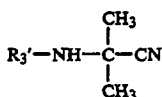
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dinedione, 4-[[4,5-dihydro-4,4-dimethyl-5-oxo-2-benzyl-thio]-1H-imidazo-1-yl]-2-(trifluoromethyl) benzonitrile, 4-[4,4-dimethyl 3-(2-hydroxyethyl) 5-oxo 2-thioxo 1-imidazolidinyl] 2-(trifluoromethyl) benzonitrile, 4-(4,4-dimethyl 2,5-dioxo 3-(4-hydroxybutyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile, 4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile and 3-(4-cyano 3-(trifluoromethyl) phenyl) 5,5-dimethyl 2,4-dioxo 1-imidazolidinebutanoic acid.

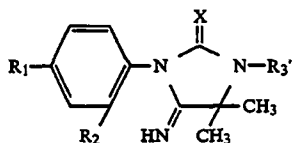
The process of the invention for the preparation of a compound of formula I comprises either reacting a compound of the formula



wherein R_1 , R_2 and X have the above definitions with a compound of the formula

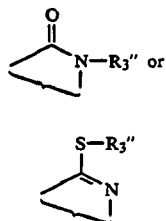


in the presence of a tertiary base wherein R'_3 has the definition of R_3 with reactive group optionally protected and if R_1 is $-\text{NO}_2$ or halogen, R_2 is halogen or $-\text{CF}_3$ and X is oxygen, R'_3 is not hydrogen to obtain a compound of the formula



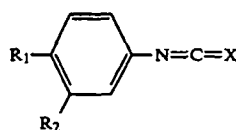
wherein R_1 , R_2 , X and R'_3 have the above definitions and optionally subjecting the latter to one or more of the following reactions in any order:

- reaction to eliminate the optional protective groups of R'_3
- reaction of hydrolysis of $\text{C}=\text{NH}$ to a ketone function or transformation of $\text{C}=\text{S}$ to $\text{C}=\text{O}$
- transformation reaction of $\text{C}=\text{O}$ to $\text{C}=\text{S}$
- and reacting the products of formula IV wherein R'_3 is hydrogen and after hydrolysis of $\text{C}=\text{NH}$ to a ketone with a compound of the formula $\text{R}''_3\text{-Hal}$ where Hal is a halogen and R''_3 is R'_3 except hydrogen to obtain a compound of formula I wherein $-\text{A}-\text{B}-$ is

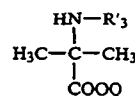


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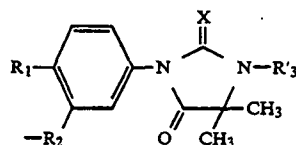
and optionally reacting the latter to eliminate the protective group of R''_3 or reacting the same with an esterification, salification or amidification agent or reacting a compound of the formula



in which R_1 , R_2 and X have the above meaning in the presence of a tertiary base with a product of the formula

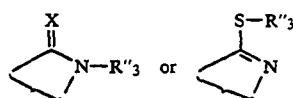


in which R'_3 has the above meaning and Q is either an alkali metal for example sodium or alkyl of 1 to 6 carbon atoms to obtain a product of the formula

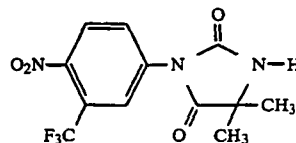


in which X , R_1 , R_2 and R'_3 have the above meaning which if desired is subjected to any one or more of the following reactions in any order:

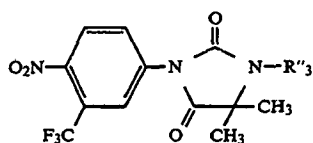
- elimination reaction of the optional protective groups that can be carried by R'_3 ;
- conversion reaction of the $>\text{C}=\text{O}$ group or groups into the $>\text{C}=\text{S}$ or if appropriate of $>\text{C}=\text{S}$ into $>\text{C}=\text{O}$;
- the action on the products of formula IVa in which R'_3 is hydrogen of a reagent of formula $\text{Hal}-\text{R}''_3$ in which R''_3 has the values of R'_3 with the exception of hydrogen and Hal is halogen to obtain the products of formula I in which $-\text{A}-\text{B}-$ is



in which R''_3 has the above meaning, then, if desired, the action of these products of an elimination agent of the optional protective groups that can be carried by R''_3 or if appropriate, the action of an esterification, amidification or salification agent, or reacting a reagent of the formula $\text{R}''_3\text{-Hal}$ as defined above with a compound of the formula



to obtain a compound of the formula



and optionally subjecting the latter to one or more of the following reactions:

- elimination reaction of optional protective groups of R''_3 and then to reaction with an esterification, salification or amidification reagent
- reaction of transformation of $C=O$ to $C=S$.

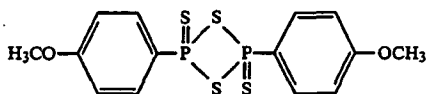
The reaction of the products of formula II with the products of formula III is preferably effected in an organic solvent such as tetrahydrofuran or dichloroethane or ethyl ether or isopropyl ether in the presence of a tertiary base such as pyridine or methylethyl pyridine.

The optional reactive functional groups of R_3 which are optionally protected in compounds of formula III, IVa or IV'' are $-OH$ or amino which are protected by the usual protective groups. Examples of such protective groups for $-NH_2$ are tert.-butyl, tert.-amyl, trichloroacetyl, chloroacetyl, benzhydryl, trityl, formyl and benzyloxycarbonyl. Examples of hydroxy protective groups are formyl, chloroacetyl, tetrahydropyran-nyl, trimethylsilyl and tert.-butyldimethylsilyl.

The above list of protective groups is not intended to be exhaustive and any protective group known, for example, in peptide chemistry may be used. Other known protective groups are described in French Patent No. 2,499,995 which is incorporated herein by reference. The optional reactions to eliminate groups are indicated in the said patent and the preferred method of elimination is acid hydrolysis with hydrochloric acid, benzene sulfonic acid, p-toluene sulfonic acid, formic acid or trifluoroacetic acid, preferably hydrochloric acid.

The optional reaction of hydrolysis of $C=NH$ to $C=O$ is preferably effected with an acid such as refluxing aqueous hydrochloric acid. When the hydrolysis of $C=NH$ into a $C=O$ is effected with a molecule also containing $C=S$, the latter may be transformed in $C=O$ group. The free hydroxy optionally contained in R_3 may also be transformed into $-SH$.

The transformation of the group $C=O$ into $C=S$ is effected with a Lawesson reagent of the formula



which is a commercial product sold by Fluka for example and is described in Bull. Soc. Chim. Belg., Vol 87 No. 3 (1987), p. 229. When two $C=O$ groups are to be changed to $C=S$, the reaction is effected in an excess of the Lawesson reagent. The same is used also when the molecule contains both $C=S$ and $C=O$ and it is desired to change the $C=O$ to $C=S$.

On the contrary, when part of the molecule contain two $C=O$ and it is desired to obtain a product with only one $C=S$, a deficiency of the Lawesson reagent is used to obtain a mixture of 3 products, each of two products with a $C=O$ and $C=S$ and one containing two $C=S$.

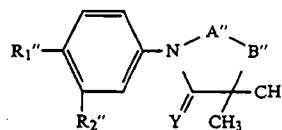
The said products can be separated by known methods such as chromatography.

The reaction of the compounds of formulae IV, IVa or IV' with a compound of the formula R''_3-Hal is effected in the presence of a strong base such as sodium hydride or potassium hydride in a phase transfer reaction in the presence of quaternary ammonium salts such as tert.-butyl ammonium. The protective groups of R''_3 may be those discussed above for R_3 . The reaction to eliminate the protective groups are as discussed above. For example, a tert-butyl dimethylsilyl group may be removed by hydrochloric acid as described in the examples infra.

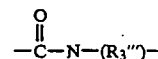
The optional esterification of the compounds of formula I wherein R''_3 is free $-OH$ is effected under the classical conditions using for example an acid or a functional derivative thereof such as its anhydride like acetic acid anhydride in the presence of a base such as pyridine. The optional esterification or salification of the compounds of formula I wherein R''_3 is $-COOH$ may be effected by known methods.

The optional amidification of the compounds of formula I wherein R''_3 is $-COOH$ is effected also under classical conditions with primary or secondary amine with a functional derivative of $-COOH$ such as a symmetrical or mixed anhydride thereof.

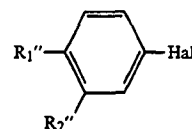
The process of the invention to prepare compounds of the formula



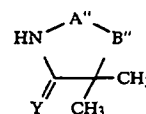
wherein R''_1 , R''_2 and $-A''-B''-$ have the definitions of R_1 , R_2 and $-A-B-$ except when $-A''-B''-$ is



and R''_3 , is hydrogen or alkyl of 1 to 7 carbon atoms and Y is oxygen, R''_1 is $-CN$ comprises reacting a compound of the formula



wherein R''_1 and R''_2 have the above definitions and Hal is halogen with a compound of the formula



wherein $-A''-B''-$ and Y have the above definitions in the presence of a catalyst and optionally a solvent. In the compounds of formula V, the halogen is preferably chlorine but may be iodine or bromine.

The role of the catalyst is obviously to trap the hydrogen halide as it forms and to facilitate the condensation reaction of the compounds of formulae V and VI to form the desired product. The catalyst is preferably a metal in its native form or its oxide or salt form or it may be a base. When the catalyst is a metal, it is preferably copper or nickel and the metallic salts are preferably the chloride or acetate. When the catalyst is a base, it is preferably sodium hydroxide or potassium hydroxide and dimethylsulfoxide may be added to the reaction medium.

The catalyst of the process may be selected from cuprous oxide, cupric oxide, metallic copper or a base such as sodium hydroxide or potassium hydroxide, preferably cuprous oxide in powdered form. The solvent used preferably is a high boiling point ether such as phenyl oxide, diglyme, triglyme and dimethylsulfoxide but also useful are high boiling point oils such as paraffin or vaseline. Preferably, the process is effected in another solvent such as phenyl oxide, diglyme, triglyme or dimethylsulfoxide, most preferably in phenyl oxide or triglyme.

The process may be effected at atmospheric pressure or under pressure at temperatures above 100° C., preferably above 150° C. for more than two hours. The reaction is preferably effected with cuprous oxide in triglyme at temperatures of 200° C. or higher for more than three hours.

The novel anti-androgenic compositions of the invention are comprised of an anti-androgenically effective amount of at least one compound of formula I and its non-toxic, pharmaceutically acceptable acid addition salts and an inert pharmaceutical carrier. The compositions may be in the form of tablets, dragees, capsules, syrups, suppositories, creams, pomades, lotions or injectable solutions prepared in the usual manner.

Examples of suitable excipients are aqueous or non-aqueous vehicles, arabic gum, lactose, starch, magnesium stearate, cocoa butter, fatty bodies of animal or vegetable origin, paraffinic derivatives, glycols, diverse wetting agents, dispersants or emulsifiers and preservatives.

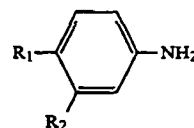
The compositions inhibit the effect of androgens on peripheral receptors and have an anti-androgenic activity useful for therapy in adults without the certain effects of a chemical castration. The compositions are useful for the treatment of adenomas and neoplasies of the prostate as well as benign hypertrophy of the prostate as well as the treatment of benign or malignant tumors of cells containing androgen receptors. They are particularly useful for the treatment of breast, brain, skin and ovarian cancer and bladder, lymphatic system, liver and kidney cancers. They are equally useful for the treatment of hirsutism, acne, seborrhea, androgenic alopecia and hyperpilosity and in the veterinary field.

The compositions of the invention are useful in dermatology and can contain another ingredient such as an antibiotic such as derivatives of retinoids for the treatment of acne, or with a 5 α -reductase inhibitor such as (5 α , 17 β)-1,1-dimethylethyl 3-oxo 4-aza- Δ^1 -androstene-17 carboxamide (or Finasteride Merck, 11th ed.) or azelaic acid or a blocking agent of androgen receptors for the treatment of acne, alopecia or hirsutism, or with a product stimulating the growth of hair such as Minoxidil for the treatment of alopecia. The compositions can also be used in the veterinary domain and in the form of radioactive products, can also be used in diagnostics as specific labels for the androgen receptors. As radioac-

tive products, the products labelled with tritium, with carbon 14 or also with iodine 125 can be used.

The novel method of the invention for inducing anti-androgenic activity in warm-blooded animals, including humans, comprises administering to warm-blooded animals an anti-androgenically effective amount of at least one compound of formula I and its non-toxic, pharmaceutically acceptable acid addition salts. The compounds may be administered parenterally, buccally, perlingually, rectally or topically and the usual daily dose is 0.133 to 6.66 mg/kg depending on the condition treated, the specific compound and the method of administration.

The starting compounds of formula II may be prepared by reacting phosgene when X is oxygen or thiophosgene when X is sulfur with an amine of the formula



A product of this type is described in French Patent No. 2,329,276. The amines of formula A are described in EP Patent No. 0,002,892 and French Patent No. 2,142,804.

The products of formula III or III' are known or can be prepared from the corresponding cyanhydrin by the process of J. Am. Chem. Soc., Vol 75 (1953), p. 4841.

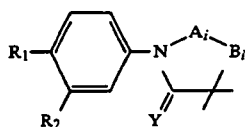
The compounds of formula III wherein R₃ is other than hydrogen may be obtained by reacting a compound of the formula R₃ Hal with 2-cyano-2-amino-propane under the conditions described above for reacting the said halide with the compounds of formula IV. An example is described by Jilek et al, Collect. Czech. Chem. Comm., Vol 54(8) (1989), p. 2248. The products of formula IV' are described in French Patent No. 2,329,276.

The compounds of formulae V and VI are commercially available known compounds and can be prepared by known methods.

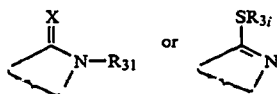
The preparation of the compounds of formula VI are described in the following publications: Zhur Preklad Khim., Vol. 28 (1955), p. 969-75 (CA, Vol. 50 (1956), p. 4881a); Tetrahedron, Vol. 43 (1987), p. 1753; J. Org. Chem., Vol. 52 (1987), p. 2407; Zh. Org. Khim., Vol. 21 (1985), p. 2006; J. Fluor. Chem., Vol. 17 (1981), p. 345; German Patent No. 637,318, European Patent No. 0,130,875 and Japanese Patent No. 81-121,525.

The products of formula VI which are derivatives of hydantoin are largely used and are known in the literature such as J. Pharm. Pharmacol., 67, Vol. 19(4) (1967), p. 209-16; J. Chem. Soc., Vol. 74(2) (1972), p. 219-221; Khim. Farm. Zh., Vol. 67(1)(5), p. 51-2; German Patent No. 2,217,914; European Patent No. 0,091,596 and J. Chem. Soc. Perkin. Trans. 1, Vol. 74(2), p. 48 and 219-221.

The novel intermediates of the invention are the compounds of the formula



wherein R_1 , R_2 and Y have the above definitions and —Ai—Bi— is



wherein X is oxygen or sulfur and R_{3i} is R_3 with the reactive groups protected among which are —OH or —NH₂ protected as above for R_3 .

In the following examples, there are described several preferred embodiments to illustrate the invention. However, it should be understood that the invention is not intended to be limited to the specific embodiments.

EXAMPLE 1

1-(4-nitro-3-trifluoromethyl-phenyl)-3,4,4-trimethyl-2,5-imidazolidinedione

A solution of 3.17 g of 1-(3-trifluoromethyl-4-nitro-phenyl)-4,4-dimethyl-imidazoline-2,5-dione (French Patent No. 2,329,276) and 32 ml of dimethylformamide were added at 23° C. to 26° C. to a 50% suspension of 492 mg of sodium hydride in oil and 3 ml of dimethylformamide and after stirring for 15 minutes, a solution of 0.7 ml of methyl iodide in 2 ml of dimethylformamide was added. The mixture was stirred for 25 minutes at 24° C. to 28° C. and was then poured into 200 g of a 1—1 water-ice mixture. The mixture was extracted with ether and the organic phase was washed with saturated aqueous sodium chloride, dried, filtered and evaporated to dryness under reduced pressure to obtain 3.6 g of the desired product melting at 116° C. An analytical sample was crystallized from isopropyl alcohol to obtain 2.73 g of the product melting at 116° C.

Analysis: $C_{13}H_{12}F_3N_3O_4$; molecular weight = 331.25				
	% C	% H	% F	% N
Calculated:	47.14	3.65	17.20	12.68
Found:	47.0	3.5	17.1	12.5

IR Spectrum ($CHCl_3$):	
C=O	1780, 1727 cm^{-1}
aromatics	1615, 1596, 1497 cm^{-1}
NO ₂	1545, 1357 cm^{-1}

EXAMPLE 2

5,5-dimethyl-1-ethyl-3-(4-nitro-3-trifluoromethyl-phenyl)-2,4-imidazolidinedione

Using the procedure of Example 1, 1 g of 1-(3-trifluoromethyl-4-nitro-phenyl)-4,4-dimethyl imidazoline-2,5-dione prepared as in French Patent No. 2,329,276 was reacted with 0.37 ml of ethyl iodide and a 50% suspension of 166 mg of sodium hydride in oil to obtain 1.19 g of the desired product melting at 110° C. to 111°

rvi

C. which was crystallized from isopropanol to obtain 934 mg of the product melting at 110° C. to 111° C.

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Analysis: $C_{14}H_{14}F_3N_3O_4$; molecular weight = 345.28				
	% C	% H	% F	% N
Calculated:	48.70	4.09	16.51	12.17
Found:	48.6	4.0	16.8	12.1

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IR Spectrum ($CHCl_3$):

C=O	1777, 1724 cm^{-1}
NO ₂	1545, 1356 cm^{-1}
aromatics	1614, 1596, 1497 cm^{-1}

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EXAMPLE 3

5,5-dimethyl-3-(4-nitro-3-trifluoromethyl-phenyl)-1-propyl-2,4-imidazolidinedione

Using the procedure of Example 1, 1 g of 1-(3-trifluoromethyl-4-nitro-phenyl)-4,4-dimethyl imidazoline-2,5-dione was reacted with 0.35 ml of 1-iodopropane and a 50% suspension of 155 mg of sodium hydride in oil to obtain after chromatography on silica with an eluant of acetone-methylene chloride (1—99), 3.087 g of raw product melting at 102° C. The product was crystallized from isopropanol to obtain 945 mg of the desired product melting at 102° C.

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Analysis: $C_{15}H_{16}F_3N_3O_4$; molecular weight = 359.31				
	% C	% H	% F	% N
Calculated:	50.14	4.49	15.86	11.69
Found:	50.1	4.4	15.9	11.5

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IR Spectrum ($CHCl_3$):

C=O	1778, 1724 cm^{-1}
NO ₂	1544, 1358 cm^{-1}
aromatics	1615, 1596, 1497 cm^{-1}

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EXAMPLE 4

5,5-dimethyl-1-isopropyl-3-(4-nitro-3-trifluoromethyl-phenyl)-2,4-imidazolidinedione

Using the procedure of Example 1, 1 g of 1-(3-trifluoromethyl-4-nitro-phenyl)-4,4-dimethyl imidazoline-2,5-dione was reacted with 0.4 ml of 2-iodopropane and a 50% suspension of 166 mg of sodium hydride in oil for 18 hours at 50° C. to obtain after chromatography over silica (eluant methylene chloride-acetone 99—1), 685 mg of product melting at 130° C. which after crystallization from isopropanol yielded 661 of the desired product melting at 130° C.

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Analysis: $C_{15}H_{16}N_3F_3O_4$; molecular weight = 359.31				
	% C	% H	% F	% N
Calculated:	50.14	4.49	15.86	11.69
Found:	50.1	4.4	16.2	11.6

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IR Spectrum ($CHCl_3$):

C=O	1779, 1771, 1723 cm^{-1}
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-continued

IR Spectrum (CHCl ₃):	
NO ₂	1544, 1361 cm ⁻¹
aromatics	1615, 1596, 1497 cm ⁻¹

EXAMPLE 5

5,5-dimethyl-3-(4-nitro-3-trifluoromethyl-phenyl)-1-(2-propenyl)-2,4-imidazolidinedione

Using the procedure of Example 1, 1 g of 1-(3-trifluoromethyl-4-nitro-phenyl)-4,4-dimethyl imidazoline-2,5-dione was reacted with 0.35 ml of allyl bromide and a 50% suspension of 166 mg of sodium hydride in oil to obtain after chromatography over silica (eluant—methylene chloride-acetone (99-1)) 1.10 g of product which after crystallization from isopropanol yielded 1.01 g of the desired product melting at 105° C.

Analysis: C₁₅H₁₄F₃N₃O₄; molecular weight = 357.29

	% C	% H	% F	% N
Calculated:	50.42	3.95	15.95	11.76
Found:	50.4	3.8	15.8	11.7

IR Spectrum (CHCl₃):

C=O	1779, 1724 cm ⁻¹
NO ₂	1545, 1358 cm ⁻¹
aromatics	1615, 1596, 1497 cm ⁻¹
CH=CH ₂	1643, 930 cm ⁻¹

EXAMPLE 6

5,5-dimethyl-3-(3-trifluoromethyl-4-nitro-phenyl)-1-benzyl-2,4-imidazolidinedione

Using the procedure of Example 1, 2 g of 1-(3-trifluoromethyl-4-nitro-phenyl)-4,4-dimethyl imidazolidine-2,5-dione was reacted with 0.71 ml of benzyl bromide and a 50% suspension of 332 mg of sodium hydride in oil to obtain after chromatography on silica and elution with 99-1 methylene chloride-acetone 2.375 g of the desired product which as crystallized from isopropanol to obtain 2.165 g of product melting at 99° C.

Analysis: C₁₉H₁₆N₃F₃O₄; molecular weight = 407.3

	% C	% H	% F	% N
Calculated:	56.02	3.96	10.31	14.00
Found:	56.1	3.8	10.2	13.9

IR Spectrum (CHCl₃):

C=O	1799, 1723 cm ⁻¹
aromatics	1608 cm ⁻¹
+	1594 cm ⁻¹ (m)
NO ₂	1545 cm ⁻¹ (F)
	1497 cm ⁻¹

EXAMPLE 7

4-(4,4-dimethyl-5-imino-2-oxo-1-imidazolidinyl)-2-trifluoromethylbenzonitrile

A solution of 10 g of 4-cyano-3-trifluoromethyl-aniline (described in European Paten No. 0,002,892) in 30 ml of ethyl acetate was added at 0 to 5° C. to 33.6 ml of

a toluene solution of 1.93 M/1 of phosgene and after stirring at 0 to 5° C. for 30 minutes, the temperature was raised to 25° C. The mixture was distilled while introducing fresh toluene maintaining to constant level for compensate the distilled volume of toluene until a temperature of about 110° C. was reached. The mixture was held at reflux until the disengagement of hydrogen chloride ceased (4 ½ hours). The temperature returned to room temperature and the white solid was dried over sodium sulfate and was rinsed with toluene 3 times. The organic phase was evaporated to dryness under reduced pressure, heated at 60° C. for one hour and then cooled under argon to obtain 11.6 g of 4-isocyanate of 2-trifluoromethyl-benzonitrile.

IR Spectrum:

-NC=O	2268 cm ⁻¹
-CN	2233 cm ⁻¹

A solution of 6.6 g of 4-isocyanate of 2-trifluoromethyl-benzonitrile in 10 ml of dichloroethane was added at 5° C. to a solution of 2.63 g of 2-amino-2-cyano-propane and 36 ml of dichloroethane and 0.9 ml of triethylamine and after stirring 16 hours at room temperature, the mixture was evaporated to dryness. The 7.7 g of residue were chromatographed on silica and eluted with a 85-15 methylene chloride-acetone mixture to obtain 3.54 g of the desired product melting at 228° C. An analytical sample was prepared by crystallizing 300 mg from isopropanol to obtain 267 mg of the product melting at 228° C.

Analysis: C₁₃H₁₁F₃N₄O; molecular weight = 296.25

	% C	% H	% F	% N
Calculated:	52.71	3.74	19.24	18.91
Found:	52.7	3.6	19.1	18.6

IR Spectrum (Nujol):

NH/OH	3340, 3290 cm ⁻¹
CN	2240 cm ⁻¹
C=O	1760 cm ⁻¹
C=N	1655 cm ⁻¹
aromatics	1606, 1570, 1502 cm ⁻¹

EXAMPLE 8

4-(4,4-dimethyl-2,5-dioxo-1-imidazolidinyl)-2-trifluoromethylbenzonitrile

A solution of 2.76 g of the product of Example 7 and 60 ml of 0.5 hydrochloric acid was refluxed for 35 minutes and was poured into 100 g of water and ice. The mixture was extracted with ethyl acetate and the organic phase was washed with water, dried and evaporated to dryness under reduced pressure to obtain 2.70 g of the desired product melting at 210° C. An analytical sample was obtained by crystallizing 440 mg of product from isopropanol to obtain 383 mg of product melting at 210° C. to 211° C.

Analysis: C₁₃H₁₀F₃N₃O₂; molecular weight = 297.24

	% C	% H	% F	% N
Calculated:	52.53	3.39	19.17	14.14

-continued

Analysis: C ₁₃ H ₁₀ F ₃ N ₃ O ₂ ; molecular weight = 297.24				
	% C	% H	% F	% N
Found:	52.4	3.2	19.4	13.9

IR Spectrum (CHCl ₃):	
CN	2245 cm ⁻¹
C=O	1788, 1722 cm ⁻¹
aromatics	1610, 1572, 1502 cm ⁻¹
NH (max)	3340 cm ⁻¹

EXAMPLE 9

3-(4-cyano-3-trifluoromethyl-phenyl)-5,5-dimethyl-2,4-dioxo-1-imidazolidine acetic acid

A solution of 600 mg of the product of Example 8 in 6 ml of dimethylformamide was added with stirring over 15 minutes to a suspension of a 50% suspension of 210 mg of sodium hydride in oil in 3 ml of dimethylformamide and after the addition of 290 mg of bromoacetic acid, the mixture was stirred for 16 hours at room temperature. After another 105 mg of sodium hydride were added, 145 mg of bromoacetic acid were added to the mixture which was stirred for 30 minutes and then poured into a mixture of 50 ml of water and 5 ml of 2N hydrochloric acid. The mixture was extracted with ether and the organic phase was washed with saturated aqueous sodium chloride, dried, filtered and evaporated to dryness under reduced pressure. The 1.22 g of residue were chromatographed on silica and eluted with a 90-10-0.5 methylene chloride-methanol-acetic acid mixture to obtain 367 mg of the desired product.

IR Spectrum:	
CN	2238 cm ⁻¹
C=O hydantoin & acid	1784, 1725, 1710 cm ⁻¹
aromatic	1616, 1580, 1508 cm ⁻¹

Ultra-violet Spectrum:		
ETOH - 0.1N HCl	max 258 nm	ε = 13,300
	inflex 277 nm	ε = 5,000
	inflex 285 nm	ε = 2,600
ETOH 0.1N NaOH	max 287 nm	ε = 19,100
	max 342 nm	ε = 1,900

EXAMPLE 10

Ethyl3-(4-cyano-3-trifluoromethyl-phenyl)-5,5-dimethyl-2,4-dioxo-1-imidazolidine-acetate

A solution of 600 mg of the product of Example 8 in 6 ml of dimethylformamide was added to a 50% suspension of 100 mg of sodium hydride in oil and 3 ml of dimethylformamide and after stirring for 15 minutes, 0.25 ml of ethyl bromoacetate was slowly added at less than 30° C. The mixture was stirred for 30 minutes and then was poured into 50 g of a 1-1 ice-water mixture. 0.5 g of monopotassium phosphate was added and the mixture was extracted with ether. The organic phase was washed with water, dried and evaporated to dryness to obtain 1.1 g of residue which was chromatographed on silica and eluted with 97-3 methylene chloride-acetone to obtain 709 mg of the desired product melting at 152° C. An analytical sample was prepared

by crystallization from isopropanol to obtain 667 mg of the desired product melting at 152° C.

Analysis: C ₁₇ H ₁₆ N ₃ F ₃ O ₄ ; molecular weight = 383.33				
	% C	% H	% F	% N
Calculated:	53.21	4.21	14.83	10.96
Found:	53.3	4.0	14.9	10.8

IR Spectrum (CHCl ₃):	
CN	2225 cm ⁻¹
imidazolidine	1786, 1729 cm ⁻¹
COOEt	1751 cm ⁻¹
aromatics	1616, 1572, 1505 cm ⁻¹

EXAMPLE 11

4-(5-imino-2-thioxo-3,4,4-trimethyl-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile

2.23 of 1-trifluoromethyl-4-amino-benzonitrile (described in European Patent No. 0,002,892) were slowly added to a solution of 22 ml of distilled water and 1 ml of thiophosgene and after stirring for one hour, the mixture was extracted with chloroform. The organic phase was washed with aqueous sodium chloride, dried and evaporated to dryness under reduced pressure to obtain 3 g of isocyanate product which as used as is.

A mixture of the 3 g of product, 1.33 ml of 2-methylamino-2-cyano-propane, 23 ml of tetrahydrofuran and 0.23 ml of triethylamine was refluxed for 40 minutes and was evaporated to dryness. The 3.07 g of residue were chromatographed on silica and eluted with a 1-1 cyclohexane-ethyl acetate mixture and then a 95-5 methylene chloride-acetone mixture to obtain 2.83 g of product which was crystallized from isopropanol to obtain 2.63 g of the desired product melting at 173° C. to 174° C.

Analysis: C ₁₄ H ₁₃ F ₃ N ₄ S; molecular weight = 326.35					
	% C	% H	% F	% N	% S
Calculated:	51.53	4.01	17.17	17.46	9.82
Found:	51.7	3.9	17.2	17.2	9.9

IR Spectrum:	
C=NH	3308, 1679 cm ⁻¹
C=S + aromatics	1608, 1575, 1505, 1488 cm ⁻¹
CN	2230 cm ⁻¹
CF ₃	1185 cm ⁻¹

EXAMPLE 12

4-(5-oxo-2-thioxo-3,4,4-trimethyl-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile

A mixture of 2.21 g of the product of Example 11 and 44 ml of 0.5 N hydrochloric acid was refluxed with stirring for one hour and was then poured into 200 g of an ice-water (1-1) mixture. The mixture was extracted with methylene chloride and the organic phase was washed with saturated aqueous sodium chloride, dried and evaporated to dryness. The residue was chromatographed on silica and eluted with a 1-1 cyclohexane-ethyl acetate mixture to obtain 2.1 g of product melting

at 171° C. which was crystallized from isopropanol to obtain 1.99 g of the desired product melting at 171° C.

Analysis: C ₁₄ H ₁₂ F ₃ N ₃ O ₂ S; molecular weight = 327.33					
	% C	% H	% F	% N	% S
Calculated:	51.37	3.69	12.84	17.41	9.79
Found:	51.4	3.5	12.7	17.6	10.79

IR Spectrum (CHCl ₃):	
C=O	1761, 1756 cm ⁻¹
aromatics	1610, 1578, 1505 cm ⁻¹
CN	2230 cm ⁻¹
CF ₃	1178 cm ⁻¹

EXAMPLE 13

4-(2,5-dithio-3,4,4-trimethyl-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile

A mixture of 839 mg of the product of Example 12, 518 mg of Lawesson reagent and 4.7 ml of toluene was refluxed for 24 hours and was then evaporated to dryness under reduced pressure. The 1.36 g of residue were chromatographed on silica and eluted with a 99-1 methylene chloride-ethyl acetate mixture and then an 85-15 cyclohexane-ethylacetate mixture to obtain 783 mg of product which was crystallized from isopropanol to obtain 690 mg of the desired product melting at 211° C. to 212° C.

Analysis: C ₁₄ H ₁₂ F ₃ N ₃ S ₂ ; molecular weight = 343.40					
	% C	% H	% F	% N	% S
Calculated:	48.97	3.52	16.60	12.24	18.67
Found:	49.0	3.4	16.6	12.2	18.6

IR Spectrum (CHCl ₃):	
CN	2230 cm ⁻¹
aromatics + conjugated system	1612, 1582, 1508 cm ⁻¹
CF ₃	1178 cm ⁻¹

EXAMPLE 14

4-(4,4-dimethyl-5-imino-2-thioxo-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile

1 g of 2-amino-2-cyano-propane and 1 ml of tetrahydrofuran were added with stirring to a mixture of 2.54 g of the isocyanate product of Example 11, 20 ml of tetrahydrofuran and 0.2 ml of triethylamine at room temperature and was then evaporated to dryness. The 3.5 g of residue were chromatographed on silica and eluted with a 7-3 ethyl acetate-cyclohexane mixture and then a 1-1 ethyl acetate-cyclohexane mixture to obtain 940 mg of the desired product. 300 g were crystallized from isopropanol to obtain 263 mg of product melting at 296° C.

Analysis: C ₁₃ H ₁₁ F ₃ N ₄ S; molecular weight = 312.32					
	% C	% H	% F	% N	% S
Calculated:	50.00	3.55	18.25	17.94	10.27

-continued

Analysis: C ₁₃ H ₁₁ F ₃ N ₄ S; molecular weight = 312.32					
	% C	% H	% F	% N	% S
Found:	49.9	3.4	18.3	17.6	10.4

IR Spectrum (Nujol):	
OH/NH	3260 cm ⁻¹
CN	2230 cm ⁻¹
C=S	1764 cm ⁻¹
aromatic + C=C	1612, 1575, 1530, 1501 cm ⁻¹

A new preparation was effected using 1,2-dichloroethane in place of tetrahydrofuran to obtain the product in a 60% yield.

EXAMPLE 15

4-(4,4-dimethyl-5-oxo-3-thioxo-1-imidazolidinyl)-1-trifluoromethyl-benzonitrile

A mixture of 635 mg of the product of Example 14 and 14 ml of 0.5 N hydrochloric acid was stirred for one hour at reflux and after cooling, 100 ml of water were added. The mixture was extracted with ethyl acetate and the organic phase was washed with aqueous sodium chloride, dried and evaporated to dryness. The 600 mg of residue were chromatographed and eluted with a 95-5 methylene chloride-acetone mixture to obtain 590 mg of product melting at 190° C. to 191° C. The latter was crystallized from isopropanol to obtain 490 mg of product melting to 190° C. to 191° C.

Analysis: C ₁₃ H ₁₀ F ₃ N ₃ O ₂ S; molecular weight = 313.30					
	% C	% H	% F	% N	% S
Calculated:	49.84	3.22	18.19	13.41	10.23
Found:	49.6	3.1	18.4	13.2	10.0

IR Spectrum (CHCl ₃):	
=C-NH	3430 cm ⁻¹
CN	2230 cm ⁻¹
C=O	1766 cm ⁻¹
aromatics and conjugated system	1612, 1578, 1505 cm ⁻¹

EXAMPLE 16

5,5-dimethyl-3-(4-nitro-3-trifluoromethyl-phenyl)-1-pentyl-2,4-imidazolidine

Using the procedure of Example 1, 1 g of 1-(3-trifluoromethyl-4-nitro-phenyl)-4,4-dimethyl imidazolidin-2,5-dione was reacted with 170 mg of sodium hydride and 0.47 ml of 1-bromo-pentane to obtain after chromatography on silica and elution with an 8-2 methylene chloride-cyclohexane mixture 1.23 g of product which as crystallized from isopropanol to obtain 995 mg of the desired product melting at 84° C.

Analysis: C ₁₇ H ₂₀ O ₄ F ₃ N ₃ ; molecular weight = 387.35				
	% C	% H	% F	% N
Calculated:	52.71	5.20	14.71	10.85
Found:	52.8	5.1	14.8	10.7

IR Spectrum (CHCl ₃):	
C=O	1778, 1723 cm ⁻¹
NO ₂	1544, 1360 cm ⁻¹

EXAMPLE 17

5,5-dimethyl-3-(4-nitro-3-trifluoromethyl-phenyl)-1-nonyl-2,4-imidazolidinedione

Using the procedure of Example 1, 1 g of 1-(3-trifluoromethyl-4-nitro-phenyl)-4,4-dimethyl imidazolidine-2,5-dione was reacted with a 50% suspension of 170 mg of sodium hydride in oil and 0.7 ml of 1-bromononane to obtain after chromatography on silica 1.08 g of the desired product melting at 63° C.

Analysis: C ₂₁ H ₂₈ O ₄ F ₃ N ₃ ; molecular weight = 443.46				
	% C	% H	% F	% N
Calculated:	56.87	6.36	12.85	9.48
Found:	57.0	6.5	12.8	9.5

IR Spectrum (CHCl ₃):	
C=O	1788, 1723 cm ⁻¹
NO ₂	1544, 1359 cm ⁻¹
C=O	1778, 1723 cm ⁻¹
NO ₂	1544, 1360 cm ⁻¹

EXAMPLE 17

5,5-dimethyl-3-(4-nitro-3-trifluoromethyl-phenyl)-1-nonyl-2,4-imidazolidinedione

Using the procedure of Example 1, 1 g of 1-(3-trifluoromethyl-4-nitro-phenyl)-4,4-dimethyl imidazolidine-2,5-dione prepared from a 50% suspension of 170 mg of sodium hydride in oil and 0.7 ml of 1-bromononane were reacted to obtain after chromatography on silica 1.08 g of the desired product melting at 63° C.

Analysis: C ₂₁ H ₂₈ O ₄ F ₃ N ₃ ; molecular weight = 443.46				
	% C	% H	% F	% N
Calculated:	56.87	6.36	12.85	9.48
Found:	57.0	6.5	12.8	9.5

IR Spectrum (CHCl ₃):	
C=O	1788, 1723 cm ⁻¹
NO ₂	1544, 1359 cm ⁻¹

EXAMPLE 18

4-(3,4,4-trimethyl-2,5-dioxo-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile

Using the procedure of Example 1, 300 mg of the product of Example 8 were reacted to obtain 275 mg of the desired product melting at 158° C.

IR Spectrum (CHCl ₃):	
C=O	1780, 1727 cm ⁻¹
aromatics	1615, 1574, 1505 cm ⁻¹

-continued

IR Spectrum (CHCl ₃):	
CN	2238 cm ⁻¹

EXAMPLE 19

4-(5-thioxo-2-oxo-3,4,4-trimethyl-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile (product A),
4-(5-oxo-2-thioxo-3,4,4-trimethyl-1-imidazolidinyl)-2-trifluoromethyl benzonitrile (product B) and
4-(2,5-dithioxo-3,4,4-trimethyl-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile (product C)

A suspension of 230 mg of the product of Example 18, 1.4 ml of toluene and 78 mg of Lawesson reagent was refluxed for 9 hours and then returned to room temperature and evaporated to dryness. The 330 mg of residue was chromatographed on silica and eluted with a 99-1 methylenechloride-acetone mixture to obtain in the following order of elution 46 mg of product C with a melting point of 210° C. to 211° C. and a R_f=0.63 (identical to the product of Example 13), 26 mg of product B with a melting point of 170° C. to 171° C. and a R_f=0.49 (identical to the product of Example 12) and 42 mg of product A with a melting point of 194° C. and a R_f=0.34.

Analysis for Product A

IR Spectrum (CHCl ₃):	
C=O	1760 cm ⁻¹
CN	2235 cm ⁻¹
aromatics	1615, 1580, 1508 cm ⁻¹

UV Spectrum (ethanol):

max	228 nm	ε = 19,400
	256 nm	ε = 12,100
	298 nm	ε = 8,600
	390 nm	ε = 70

EXAMPLE 20

4-(4,5-dihydro-4,4-dimethyl-2-methylthio-5-oxo-1H-imidazolidin-1-yl)-2-trifluoromethyl benzonitrile

A solution of 626 mg of the product of Example 15 in 6 ml of dimethylformamide was added to a 50% suspension of 108 mg of sodium hydride in oil and 1.8 ml of dimethylformamide and after rinsing with 0.3 ml of dimethylformamide, the mixture was stirred for 10 minutes after cessation of hydrogen evolution. A mixture of 0.19 ml of methyl iodide in 1 ml of dimethylformamide was added dropwise and after 45 minutes of reaction, the mixture was poured into 50 g of an ice-water mixture containing 0.5 g of monosodium phosphate. The mixture was extracted 4 times with ether and the combined organic phases were washed with aqueous sodium chloride, dried over magnesium sulfate and evaporated to dryness. The 668 mg of residue were chromatographed on silica and eluted with a 95-5 dichloromethane-ethyl acetate mixture to obtain 640 mg of the desired product which chromatographed again on silica. Elution with a 7-3 cyclohexane-ethyl acetate mixture yielded after taking up in ether 507 mg of the desired product melting at 62° C.

IR Spectrum:	
C=O	1747 cm ⁻¹
C=N and aromatics	1614, 1581, 1563, 1503 cm ⁻¹

UV Spectrum (ethanol):		
max	209 nm	ε = 26,000
inflex.	236 nm	ε = 11,500
inflex.	264 nm	ε = 8,700

EXAMPLE 21

4-(4,5-dihydro-4,4-dimethyl-5-oxo-2-benzylthio)-1H-imidazol-1-yl) 2-trifluoromethyl-benzonitrile

A solution of 313 mg of 4-(4,4-dimethyl-5-oxo-2-thioxo-1-imidazolidinyl) 2-trifluoromethyl-benzonitrile in 3 ml of dimethylformamide were added to a suspension of 53 mg of sodium hydride in oil and 0.5 ml of dimethylformamide and after stirring for 10 minutes, 0.1 ml of benzyl bromide were added. The mixture was stirred for 30 minutes and then poured into an ice-water mixture containing 500 mg of monosodium phosphate. The mixture was extracted with ether and the organic phase was washed with aqueous sodium chloride, dried and evaporated to dryness. The 450 mg of residue were chromatographed on silica and eluted with a 97.5-2.5 methylene chloride-ethyl acetate mixture to obtain 316mg of the desired product with a RF=0.38.

	Analysis:			
	%C	%H	%F	%N
Calculated:	59.54	4.0	14.12	10.41
Found:	59.6	4.0	14.1	10.2

IR Spectrum (CHCl ₃):	
C=O	1746 cm ⁻¹
CN	2236 cm ⁻¹
aromatics and conjugated system	1614, 1580, 1570, 1503, 1499 cm ⁻¹

EXAMPLE 22

4-(4,4-dimethyl-3-(2-hydroxyethyl)-5-imino-2-thioxo-1-imidazolidinyl) 2-trifluoromethyl-benzonitrile

8 ml of ethanoline were added dropwise at 20° C. to 30° C. to 12.3 ml of the cyanhydrin of acetone and after stirring for 18 hours, the mixture was distilled to obtain 2.3 g of a mixture of 2-(2-hydroxyethyl)-amino-2-methyl-propanenitrile and 2,2-dimethyl-oxazolidine which was used as is for the next step.

A mixture of 1.18 g of the said mixture, 2.11 g of the isothiocyanate of Example 11 and 20 ml of tetrahydrofuran and 0.5 ml of triethylamine was refluxed for 30 minutes and then evaporated to dryness. The residue was chromatographed on silica and eluted with a 95-5 methylene chloride-acetone mixture to obtain 1.26 g of the desired product and 686 mg of N-(4-cyano-2-trifluoromethyl-phenyl)-2,2-dimethyl-3-oxazolidine carbothioamide. The 686 mg were dissolved in 10 ml of ethyl acetate and after the addition of 30 ml of cyclohexane, the mixture was concentrated to 4 ml and vacuum filtered and dried to obtain another 518 mg of

product. The raw product was dissolved in 20 ml of isopropanol and the solution was concentrated to 5 ml, vacuum filtered and dried to obtain 1.04 g of the desired product melting at 181° C.

	Analysis:				
	%C	%H	%F	%N	%S
Calculated:	50.55	4.24	16.00	15.72	9.00
Found:	50.4	4.1	15.9	15.6	9.0

IR Spectrum (CHCl ₃):	
OH	3630 cm ⁻¹
=NH	3314, 1677 cm ⁻¹
CN	2230 cm ⁻¹
aromatics	1611, 1576, 1504 cm ⁻¹

EXAMPLE 23

4-(4,4-dimethyl-3-(2-hydroxyethyl)-5-oxo-2-thioxo-1-imidazolidinyl) 2-trifluoromethyl-benzonitrile (Product A) and 4-(4,4-dimethyl-2,5-dioxo-3-(2-mercaptoethyl)-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile (Product B)

A mixture of 680 mg of the product of Example 22, 7 ml of water and 7 ml of hydrochloric acid was refluxed for 10 minutes and after cooling to room temperature, the mixture was extracted with ethyl acetate. The organic phase was washed with aqueous sodium chloride, dried and evaporated to dryness. The residue was chromatographed on silica and eluted with a 1-1 cyclohexane-ethyl acetate mixture to obtain 119 mg of product B with a Rf=0.35 and 569 mg of product A with a Rf=0.14 and a melting point of ≈130° C.

	Analysis: C ₁₅ H ₁₄ F ₃ N ₃ O ₂ S; molecular weight = 357.36				
	%C	%H	%F	%N	%S
Calculated:	50.42	3.95	15.95	11.76	8.97
Product A Found:	50.7	4.0	15.7	11.5	9.1
Product B Found:	50.6	3.8	15.9	11.6	9.1

IR Spectrum (CHCl ₃):	
Product A:	
OH	3626 cm ⁻¹
CN	2236 cm ⁻¹
C=O	1763 cm ⁻¹
aromatics	1615, 1578, 1504 cm ⁻¹
Product B:	
Absence of OH	
CN	2228 cm ⁻¹
C=O	1780, 1726 cm ⁻¹
aromatics	1615, 1578, 1505 cm ⁻¹

Using 4-(4,4-dimethyl-2,5-dioxo-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile of Example 8 and the appropriate reactants, the following products were prepared.

EXAMPLE 24

4-(4,4-dimethyl-2,5-dioxo-3-ethyl-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile with a melting point of 100° C. to 101° C.

Analysis: C ₁₅ H ₁₄ F ₃ N ₃ O ₂ ; molecular weight = 325.29				
	%C	%H	%F	%N
Calculated:	55.39	4.34	17.52	12.92
Found:	55.7	4.3	17.6	12.8

IR Spectrum (CHCl ₃):	
CN	2238 cm ⁻¹
C=O	1777, 1724 cm ⁻¹
aromatics	1617, 1575, 1505 cm ⁻¹

EXAMPLE 25

4-(4,4-dimethyl-2,5-dioxo-3-(2-propenyl)-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile melting at 109° C. to 110° C.

Analysis: C ₁₆ H ₁₄ F ₃ N ₃ O ₂ ; molecular weight = 337.35				
	%C	%H	%F	%N
Calculated:	56.97	4.18	16.90	12.46
Found:	57.0	4.1	16.2	12.3

IR Spectrum (CHCl ₃):	
CN	2238 cm ⁻¹
C=O	1728, 1725 cm ⁻¹
HC=CH ₂	1645 cm ⁻¹
aromatics	1616, 1575, 1505 cm ⁻¹

EXAMPLE 26

4-(4,4-dimethyl-2,5-dioxo-3-benzyl-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile melting at 98° C. to 99° C.

Analysis: C ₂₀ H ₁₆ F ₃ N ₃ O ₂ ; molecular weight = 387.36				
	%C	%H	%F	%N
Calculated:	62.01	4.16	14.71	10.85
Found:	62.0	4.1	14.7	10.8

IR Spectrum (CHCl ₃): C—NH: 3430 cm ⁻¹	
CN	2238 cm ⁻¹
C=O	1779, 1724 cm ⁻¹
aromatics	1615, 1605, 1575, 1504, 1497 cm ⁻¹

EXAMPLE 27

4-(4,4-dimethyl-2,5-dioxo-3-(4-fluorobenzyl)-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile melting at 101° C. to 102° C.

Analysis: C ₂₀ H ₁₅ F ₄ N ₃ O ₂ ; molecular weight = 405.35				
	%C	%H	%F	%N
Calculated:	59.26	3.73	18.75	10.37
Found:	59.1	3.5	18.9	10.3

IR Spectrum (CHCl ₃):	
CN	2238 cm ⁻¹
C=O	1780, 1724 cm ⁻¹
aromatics	1615, 1612, 1505 cm ⁻¹

EXAMPLE 28

4-(4,4-dimethyl-2,5-dioxo-3-(4-methoxybenzyl)-1-imidazolidinyl)-benzonitrile melting at 95° C. to 96° C.

Analysis: C ₂₁ H ₁₈ F ₃ N ₃ O ₃ ; molecular weight = 417.39				
	%C	%H	%F	%N
Calculated:	60.43	4.35	13.65	10.07
Found:	59.1	3.5	10.9	10.3

IR Spectrum (CHCl ₃):	
CN	2238 cm ⁻¹
C=O	1778, 1723 cm ⁻¹
aromatics	1615, 1584, 1514, 1505 cm ⁻¹

EXAMPLE 29

4-(4,4-dimethyl-2,5-dioxo-3-(4-trifluoromethyl-benzyl)-1-imidazolidinyl)-2-trifluoromethyl benzonitrile melting at ≈89° C. to 90° C.

Analysis: C ₂₁ H ₁₅ F ₆ N ₃ O ₂ ; molecular weight = 313.30				
	%C	%H	%F	%N
Calculated:	55.39	3.32	25.03	9.23
Found:	55.2	3.2	25.3	9.2

IR Spectrum (CHCl ₃):	
CN	2238 cm ⁻¹
C=O	1615, 1505 cm ⁻¹
aromatics	1615, 1505 cm ⁻¹

EXAMPLE 30

4-(4,4-dimethyl-2,5-dioxo-3-(2-epoxymethyl)-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile melting at 112° C. to 113° C.

Analysis: C ₁₆ H ₁₄ F ₃ N ₃ O ₃ ; molecular weight = 353.30				
	%C	%H	%F	%N
Calculated:	54.39	3.99	16.13	11.89
Found:	54.7	4.0	16.1	11.8

IR Spectrum (CHCl ₃):	
CN	2235 cm ⁻¹
C=O	1781, 1725 cm ⁻¹
aromatics	1615, 1576, 1505 cm ⁻¹

EXAMPLE 31

4-(4,4-dimethyl-2,5-dioxo-3-propyl-1H-imidazolidinyl)-2-trifluoromethyl benzonitrile melting at 113° C. to 114° C.

Analysis: C ₁₆ H ₁₆ F ₃ N ₃ O ₂ ; molecular weight = 339.32				
	%C	%H	%F	%N
Calculated:	56.64	4.75	16.80	12.38
Found:	56.7	4.7	16.7	12.2

IR Spectrum (CHCl ₃):	
CN	2236 cm ⁻¹
C=O	1778, 1725 cm ⁻¹
aromatics	1616, 1505 cm ⁻¹

EXAMPLE 32

4-(4,4-dimethyl-2,5-dioxo-3-isopropyl-1-imidazolidinyl)-2-trifluoromethyl benzonitrile melting at 138° C. to 139° C.

Analysis: C ₁₆ H ₁₆ F ₃ N ₃ O ₂ ; molecular weight = 339.32				
	%C	%H	%F	%N
Calculated:	56.64	4.75	16.80	12.38
Found:	56.5	4.7	17.1	12.3

IR Spectrum (CHCl ₃):	
CN	2236 cm ⁻¹
C=O	1778, 1724 cm ⁻¹
aromatics	1616, 1575, 1505 cm ⁻¹

Using 4-(4,4-dimethyl-5-oxo-2-thioxo-1-imidazolidinyl)-2-trifluoromethyl benzonitrile of Example 15 and the appropriate reactants, the following compounds were prepared:

EXAMPLE 33

4-(4,5-dihydro-4,4-dimethyl-2-nonylthio-5-oxo-1H-imidazol-1-yl) 2-trifluoromethyl-benzonitrile were a Rf=0.35 (97.5-2.5 methylene chloride-ethyl acetate eluant).

EXAMPLE 34

4-(4,5-dihydro-4,5-dimethyl-2-(3-hydroxypropylthio)-5-oxo-1H-imidazol-1-yl) 2-trifluoromethyl-benzonitrile with a Rf=0.17 (8-2 methylene chloride-ethyl acetate eluant).

EXAMPLE 35

Ethyl

[1-(4-cyano-3-trifluoromethyl-phenyl)-4,5-dihydro-4,4-dimethyl-5-oxo-1H-imidazol-2-yl]-thio]-acetate with a Rf=0.20 (65-35 cyclohexane-ethyl acetate eluant).

Using the isocyanate of Example 11 and the appropriate reactants, the following compounds were prepared.

EXAMPLE 36

4-(4,4-dimethyl-3-ethyl-5-imino-2-thioxo-1-imidazolidinyl)-2-trifluoromethyl benzonitrile with a Rf=0.16 (95-5 methylene chloride-acetone eluant).

EXAMPLE 37

4-(4,4-dimethyl-5-imino-3-pentyl-2-thioxo-1-imidazolidinyl)-2-trifluoromethyl benzonitrile with a Rf=0.35 (8-2 ethyl acetate-cyclohexane eluant)

Using the 4-(4,4-dimethyl-3-ethyl-5-imino-2-thioxo-1-imidazolidinyl)-2-trifluoromethyl benzonitrile of Example 36 and the 4-(4,4-dimethyl-5-imino-3-pentyl-2-thioxo-1-imidazolidinyl) 2-trifluoromethyl-benzonitrile of Example 37 and 0.5 N hydrochloric acid, the following compounds were prepared.

EXAMPLE 38

4-(4,4-dimethyl-3-ethyl-5-oxo-2-thioxo-1-imidazolidinyl)-2-trifluoromethyl benzonitrile with a Rf=0.38 (1-1 ethyl acetate-cyclohexane eluant).

EXAMPLE 39

4-(4,4-dimethyl-5-oxo-3-pentyl-2-thioxo-1-imidazolidinyl)-2-trifluoromethyl benzonitrile with a melting point of 78° C. and a Rf=0.66 (8-2 ethyl acetate-cyclohexane eluant)

Using 4-(4,5-dihydro-4,4-dimethyl-2-methylthio-5-oxo-1H-imidazol-1-yl) 2-trifluoromethyl-benzonitrile of Example 20 and 4-(4,5-dihydro-4,4-dimethyl-5-oxo-2-benzylthio-1H-imidazol-1-yl) 2-trifluoromethyl-benzonitrile of Example 21 and the Lawesson reagent, the following compounds were prepared.

EXAMPLE 40

4-(4,5-dihydro-4,4-dimethyl-2-methylthio-5-thioxo-1H-imidazol-1-yl) 2-trifluoromethyl-benzonitrile with a Rf=0.36 (97.5-2.5 methylene chloride-ethyl acetate eluant).

EXAMPLE 41

4-(4,5-dihydro-4,4-dimethyl-2-benzylthio-5-thioxo-1H-imidazol-1-yl) 2-trifluoromethyl-benzonitrile with a Rf=0.62 (98-2 methylene chloride-ethyl acetate eluant).

EXAMPLE 42

3-(4-cyano-3-trifluoromethyl-phenyl)-5,5-dimethyl-2,4-dioxo-N-methyl-N-isopropyl-1-imidazolidine acetamide

0.1 ml of N-methyl-morpholine was added to a suspension of 3-(4-cyano-3-trifluoromethyl-phenyl)-5,5-dimethyl-2,4-dioxo-1-acetic acid in 4 ml of methylene chloride and after cooling the solution to -10° C., 0.1 ml of isobutyl chloroformate was added dropwise. After stirring for 25 minutes at -10° C., 0.15 ml of N-methyl-N-isopropylamine was added and the mixture was allowed to return to room temperature over 40 minutes. 5 ml of an aqueous saturated sodium bicarbonate solution were added and after stirring for 30 minutes, the mixture was extracted with methylene chloride. The organic phase was washed with water, dried and evaporated to dryness under reduced pressure. The residue was chromatographed on silica and eluted with a 96-4 methylene chloride-acetone mixture to obtain 147 mg of the desired product.

IR Spectrum (CHCl ₃):	
CN	2236 cm ⁻¹
hydantoin C=O	1783, 1728 cm ⁻¹
amide C=O	1661 cm ⁻¹

-continued

IR Spectrum (CHCl ₃):	
aromatics	1615, 1575, 1505 cm ⁻¹

EXAMPLE 43

4-(4,4-dimethyl-2,5-dioxo-3-(2-hydroxyethyl)-1-imidazolidinyl) 2-trifluoromethyl-benzonitrile

Using the procedure of Example 9, 900 mg of the product of Example 8 and 1.91 g of 2-bromoethane tert.-butyldimethylsilyl ether were reacted to obtain 1 g of the silyloxy ether derivative melting at 86° C. to 87° C. after chromatography on silica and elution with a 7 g cyclohexane-ethyl acetate mixture.

1 ml of 2 N hydrochloric acid were added to a mixture of 380 mg of the silyloxy ether, 4 ml of methanol and 1 ml of methylene chloride and after stirring for 40 minutes at room temperature, the mixture was poured into 15 ml of water and was extracted with methylene chloride. The organic phase was washed with water, dried and evaporated to dryness and the residue was chromatographed on silica. Elution with a 7-3 methylene chloride-ethyl acetate mixture yielded the desired product which after crystallization from isopropanol melted at 109° C. to 110° C. and had a R_f=0.9.

	Analysis:			
	%C	%H	%F	%N
Calculated:	52.79	4.23	16.70	12.31
Found:	52.5	4.2	16.7	12.1

EXAMPLE 44

Using the procedure of Example 43, 2-bromopropanol tert.-butyldimethylsilyl ether was reacted to obtain 4-(4,4-dimethyl-2,5-dioxo-3-(3-hydroxypropyl)-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile melting at 131° C. to 132° C. and a R_f=0.13 (3-1 methylene chloride-ethyl acetate eluant).

EXAMPLE 45

4-[3-(2-acetyloxyethyl)-4,4-dimethyl-2,5-dioxo-1-imidazolidinyl]2-trifluoromethyl-benzonitrile

A mixture of 215 mg of the product of Example 43, 15 mg of 4-dimethylamino-pyridine, 1 ml of pyridine and 0.5 ml of acetic acid anhydride was stirred at room temperature for 30 minutes and was then poured into 20 ml of a saturated aqueous sodium bicarbonate solution. After stirring for 20 minutes, the mixture was extracted with ethyl acetate. The organic phase was washed with water and evaporated to dryness and the pyridine and residual acetic acid were distilled. The residue was chromatographed on silica and eluted with a 65-35 methylene chloride-ethyl acetate mixture. The residue with a R_f=0.35 was taken up in isopropanol, partially concentrated, iced and vacuum filtered to obtain after drying 210 mg of the desired product melting at 99° C. to 100° C.

	Analysis:			
	%C	%H	%F	%N
Calculated:	53.27	4.21	14.87	10.96
Found:	53.5	4.3	15.2	10.9

Using the above procedure, the following products were prepared.

EXAMPLE 46

4-(4,4-dimethyl-2,5-dioxo-3-(5-hydroxypropyl)-1-imidazolidinyl) 2-trifluoromethyl-benzonitrile melting at 101° C. to 102° C.

EXAMPLE 47

4-(4,4-dimethyl-2,5-dioxo-3-(2-methoxyethyl)-1-imidazolidinyl) 2-trifluoromethyl-benzonitrile melting at 68° C. to 69° C.

EXAMPLE 48

4-(4,4-dimethyl-2,5-dioxo-3-cyanomethyl-1-imidazolidinyl)-2-trifluoromethyl benzonitrile melting at 186° C. to 187° C.

EXAMPLE 49

4-(4,4-dimethyl-2,5-dioxo-3-[(1,3-dioxolan-2-yl)-methyl]-1-imidazolidinyl) 2-trifluoromethyl-benzonitrile melting at 135° C. to 136° C.

EXAMPLE 50

4-(4,4-dimethyl-2,5-dioxo-3-(2-chloroethyl)-1-imidazolidinyl) 2-trifluoromethyl-benzonitrile melting at 120° C. to 121° C.

EXAMPLE 51

1-(3,4-dichlorophenyl)-5-imino-3,4,4-trimethyl-2-imidazolidine thione

A mixture of 2.4 g of the isocyanate of 3,4-dichlorophenyl, 1.3 ml of 2-methylamino-2-cyano-propane, 23 ml of tetrahydrofuran and 0.23 ml of triethylamine was refluxed for 16 hours and then evaporated to dryness under reduced pressure. The residue was chromatographed on silica and eluted with a 96-4 methylene chloride-acetone mixture to obtain after crystallization from ether, 2.54 g of the desired product melting at 133° C.

EXAMPLE 52

3-(3,4-dichlorophenyl)-2-thioxo-1,5,5-trimethyl-1-imidazolidinone

A suspension of 1.88 g of the product of Example 51 in 14 ml of 6 N hydrochloric acid was refluxed for 45 minutes and after the addition of another 14 ml of 6 N hydrochloric acid, the mixture was refluxed for 2 more hours. Another 4 ml of 6 N hydrochloric acid were added and the mixture was refluxed for 90 minutes and then returned to room temperature. 100 g of ice were added and the mixture was extracted with ethyl acetate. The organic phase was washed with water, dried and evaporated to dryness. The residue was chromatographed on silica and eluted with a 1-1 cyclohexane-ethyl acetate mixture to obtain 1.84 g of the desired product melting at 129° C. after crystallization from isopropanol.

	Analysis: C ₁₂ H ₁₂ Cl ₂ N ₂ O ₂ S; molecular weight = 303.21				
	%C	%H	%Cl	%N	%S
Calculated:	47.54	3.99	23.38	9.24	10.57
Found:	47.5	3.8	23.2	9.3	10.5

IR Spectrum (CHCl ₃):	
C=O	1753 cm ⁻¹
C=S + aromatics	1595, 1570, 1496 cm ⁻¹

Using the above procedures, the following compounds were prepared:

EXAMPLE 53

3-(3,4-dichlorophenyl)-3,5-dihydro-5,5-dimethyl-2-methylthio-4H-imidazol-4-one melting at 110° C.

EXAMPLE 54

1-(3,4-dichlorophenyl)-3,4,4-trimethyl-2,5-imidazolidine-dithione melting at ≈146° C.

EXAMPLE 55

1-(4-chloro-3-trifluoromethyl-phenyl)-4,4-dimethyl-2-thioxo-5-imidazolidinone melting at 176° C.

EXAMPLE 56

1-(4-chloro-3-trifluoromethyl-phenyl)-4,4-dimethyl-5-imino-2-imidazolidinethione melting at 173° C. to 174° C.

EXAMPLE 57

3-(3,4-dichlorophenyl)-3,5-dihydro-5,5-dimethyl-2-benzylthio 4H-imidazol-4-one

IR Spectrum (CHCl ₃):	
C=O	1736 cm ⁻¹
CN + aromatics	1578, 1496 cm ⁻¹

EXAMPLE 58

4-(4,4-dimethyl 2,5-dioxo 3-(4-hydroxy butyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

a) Condensation

600 mg of 4-(4,4-dimethyl-2,5-dioxo-1-imidazolidinyl)-2-(trifluoromethyl) benzonitrile obtained as in Example 8—in 5 ml of dimethylformamide were added to a suspension of 104 mg of sodium hydride in 0.8 ml of dimethylformamide, while maintaining the temperature below 20° C. After 10 minutes of stirring, 445 mg of 4-chloro-t-butyl-dimethylsilylether and 300 mg of sodium iodide were added. The mixture was heated for 16 hours at 50° C. and then, cooled to ambient temperature. 87 mg of sodium hydride were added followed by another 400 mg of the chlorinated ether and 267 mg of sodium hydride were added. The mixture was heated for another hour and then, returned to ambient temperature, and poured into 60 ml of water containing 600 mg of monopotassium phosphate. Extraction was carried out with ether and the organic phase was washed with water, dried and the solvent was evaporated. The residue was chromatographed on silica (eluant: methylene chloride—acetone (99-1)) to obtain 526 mg of product which as used as is for the stage following the cleavage.

The said product was mixed in 5 ml of methanol and 1.5 ml of 2 N hydrochloric acid and the mixture was stirred for 40 minutes at ambient temperature. The mixture was poured into 30 ml of water and was extracted with methylene chloride. The organic phase was washed with water, dried and the solvent was evaporated. After chromatographing the residue on silica

(eluant methylene chloride—acetone (9-1), the fractions with a Rf=0.15, were recovered, and after crystallization from isopropyl ether, 307 mg of the expected product melting at 102°–103° C. were obtained.

Analysis; C ₁₇ H ₁₈ F ₃ N ₃ O ₃ ; molecular weight = 369.35				
	C%	H%	F%	N%
Calculated	55.28	4.91	15.43	11.38
Found	55.2	4.9	15.3	11.1

IR Spectrum (CHCl ₃):	
OH	3628 cm ⁻¹
C=N	2236 cm ⁻¹
C=O	1778-1724 cm ⁻¹
Aromatics	1615-1575-1505 cm ⁻¹

Preparation of the 4-chloro t-butyl dimethylsilylether used at the start of Example 58.

9.9 ml of 4-chloro-1-butanol and 24.3 g of imidazole in 50 ml of tetrahydrofuran were stirred and 2.82 g of tertbutyldimethylsilyl chloride in 20 ml of tetrahydrofuran were added dropwise at a temperature of less than 20° C. The mixture was stirred for 18 hours at ambient temperature, followed by separating, rinsing with tetrahydrofuran and eliminating the solvent under reduced pressure. The residue was purified by chromatography on silica (eluant: cyclohexane—ethyl acetate (95-5)) to obtain 17.5 g of the expected product.

EXAMPLE 59

(1,1-dimethyl) ethyl 3-(4-cyano 3-trifluoro-methyl-phenyl)5,5-dimethyl 2,4-dioxo-1-imidazolidine acetate

450 mg of the product of Example 8—in solution in 4 ml of dimethylformamide were added to a suspension of 78 mg of sodium hydride at 50% in oil and 0.5 ml of dimethylformamide. The mixture was stirred for 15 minutes and then without exceeding 30° C., 0.22 ml of tertbutyl bromoacetate were slowly added. The mixture was stirred for 16 hours and then, was poured into 50 g of a water and ice mixture (1—1). 0.5 g of monopotassium phosphate were added and extraction was carried out with ether. The organic phase was washed with water, dried and evaporated to dryness. The 1.1 g of crude product was chromatographed on silica (eluant: methylene chloride—acetone (99-1)) to obtain 425 mg of the expected product melting at 122°–123° C. with a Rf=0.28 (eluant: methylene chloride—acetone (99-1))

IR Spectrum (CHCl ₃):	
C=O	1788-1729 cm ⁻¹ (hydantoin) 1745 cm ⁻¹ (ester)
C=N	2235 cm ⁻¹
Aromatics	1616-1505 cm ⁻¹

UV Spectrum (EtOH)	
Max. 258 nm	= 16100
Infl. 277 nm	= 6000
Infl. 285 nm	= 3000

b) Substitution

548 mg of 4-[4,4-dimethyl-2,5-dioxo-3-(2-chloroethyl) 1-imidazolidinyl]-2-(trifluoromethyl) benzonitrile (Example 50—in solution in 4 ml of dimethylformamide) were added to the solution of a) and the mixture was stirred for 2 hours, poured into 50 ml of water with 0.5 g of monopotassium phosphate. Extraction was carried out with ether and the organic phase was washed with water and dried and the solvent was evaporated. After chromatographing the residue on silica (eluant: cyclohexane—ethyl acetate (75-25)), 570 mg of the expected product melting at 93°–94° C. with a Rf=0.29 (eluant: cyclohexane—ethyl acetate (75-25)) were obtained.

IR Spectrum (CHCl ₃)	
C=O	1780-1726 cm ⁻¹
C≡N	2238 cm ⁻¹
Aromatics	1616-1579-1506 cm ⁻¹
(fluorophenyl) thio	1591-1492 cm ⁻¹

UV Spectrum (EtOH)	
Max. 254 nm	= 18600
Infl. 277 nm	= 7500
Infl. 286 nm	= 4200

EXAMPLE 66

4-(4,4-dimethyl-2,5-dioxo-3-(2-(4-fluorophenyl sulfonyl) ethyl)-1-imidazolidinyl-2-(trifluoromethyl) benzonitrile

1.21 g of metachloroperbenzoic acid in 24 ml of methylene chloride were added dropwise at a temperature of less than 29° C. to 222 mg of the product of Example 65 in 4.4 ml of methylene chloride. After 30 minutes of stirring, the mixture was poured into 30 ml of sodium thiosulfate (0.5 M/1). The mixture was stirred for 10 minutes, followed by decanting and extracting with methylene chloride. The organic phase was washed with a saturated aqueous solution of sodium bicarbonate, then with water, dried, and the solvent was evaporated. After chromatographing the residue on silica (eluant: cyclohexane—ethyl acetate (1—1)), 220 mg of product were obtained which was crystallized from isopropanol to obtain 196 mg of the expected product melting at 155°–156° C. with a Rf=0.22 (eluant: ethyl acetate—cyclohexane (1—1)).

IR Spectrum (CHCl ₃)	
C=O	1783-1727 cm ⁻¹
C≡N	2236 cm ⁻¹
Aromatics	1615-1593-1505-1497 cm ⁻¹
SO ₂	1314-1150 cm ⁻¹

UV Spectrum (EtOH)	
Max. 258 nm	= 16700
Infl. 286 nm	

EXAMPLE 67

4-(4,4-dimethyl 2,5-dioxo 3-(2-((4-fluorophenyl) sulfinyl) ethyl) 1-imidazolidinyl 2-(trifluoromethyl) benzonitrile

222 mg of the product of Example 65 in 15 ml of methanol were stirred for 30 minutes at ambient temperature in the presence of 5 ml of an aqueous solution of sodium metaperiodate (0.1 M/1). The suspension was heated for one hour at 40° C. and 10 ml of methanol and 5 ml of oxidizing solution were added. The methanol was evaporated off and after 10 ml of a saturated solution of sodium chloride were added, extraction was carried out with ethyl acetate. The organic phase was washed with salt water, dried, and the solvent was evaporated. After chromatographing the residue on silica (eluant: methylene chloride—acetone (9-1)), 205 mg of product were obtained which was crystallized from isopropanol to obtain 180 mg of the expected product melting at 145°–146° C. with a Rf=0.10 (eluant: methylene chloride—acetone (9-1)).

IR Spectrum (CHCl ₃)	
C=O	1782-1727 cm ⁻¹
C≡N	2236 cm ⁻¹
Aromatics	1615-1592-1505-1493 cm ⁻¹

UV Spectrum (EtOH)	
Max. 258 nm	ε = 17600
Infl. 285 nm	

Using the procedure of the preceding examples, 4-(4,4-dimethyl-2,5-dioxo-1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile of Example 8—and the appropriate reagents, the compounds of the following examples were obtained:

EXAMPLE 68

4-(4,4-dimethyl 2,5-dioxo 3-((3-methoxyphenyl) methyl) 1-imidazolidinyl 2-(trifluoromethyl) benzonitrile Melting at 88°–89° C. with a Rf=0.21 (eluant: cyclohexane—ethyl acetate (7-3))

IR Spectrum (CHCl ₃)	
C=O	1779-1724 cm ⁻¹
C≡N	2238 cm ⁻¹
Aromatics	1614-1602-1588-1575-1504-1491

UV Spectrum (EtOH)	
Max. 260 nm	ε = 16800
Infl. 210 nm	ε = 28500
Infl. 280 nm	ε = 8900

EXAMPLE 69

4-(4,4-dimethyl 2,5-dioxo 3-(2-(4-morpholinyl) ethyl) 1-imidazolidinyl 2-(trifluoromethyl) benzonitrile with a Rf=0.20 (eluant: methylene chloride—acetone (70-30))

IR Spectrum (CHCl ₃)	
C=O	1779-1725 cm ⁻¹
C=N	2235 cm ⁻¹
Aromatics	1616-1576-1505 cm ⁻¹
morpholinyl	1117 cm ⁻¹

UV Spectrum (EtOH)	
Max. 261 nm	ε = 14000
Infl. 277 nm	ε = 6900
Infl. 286 nm	ε = 3600

EXAMPLE 70

4-(4,4-dimethyl 3-(2-hydroxyethyl) 5-imino 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl)-benzonitrile

a) Preparation of the isothiocyanate

2.23 g of 1-trifluoromethyl-4-amino benzonitrile (prepared accord to EP 0002892) were added slowly to a solution of 22 ml of distilled water and 1 ml of thiophosgene and the mixture was stirred for one hour and then extracted with chloroform. The extracts were washed with salt water, dried and evaporated to dryness under reduced pressure to obtain 3 g of product which was used as is for obtaining the imine.

b) Obtaining the imine

5 g of the said isothiocyanate were mixed with 37 ml of tetrahydrofuran in the presence of 1.5 ml of triethylamine and 2.8 g of 2-[(2-hydroxy ethyl) amino] 2-methyl propane nitrile (prepared in Example 22)—in solution in 10 ml of tetrahydrofuran were added all at once. The temperature spontaneously increased to 34° C. and the resultant mixture was allowed to return to ambient temperature while stirring for one hour. The solvent was evaporated off and the residue was chromatographed on silica (eluant: methylene chloride—methanol (7-3)) to obtain 5.87 g of the expected product melting at 181° C., after crystallization from isopropanol.

EXAMPLE 71

4-(4,4-dimethyl 3-(2-hydroxyethyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

4.6 g of the product of Example 70 in 65 ml of methanol was refluxed for one hour in the presence of 10 ml of 2 N hydrochloric acid. The mixture was cooled to ambient temperature and poured into 300 ml of ice-cooled water. Extraction was carried out with ethyl acetate and the organic phase was washed with salt water, dried, and the solvent was evaporated off. The residue was chromatographed on silica (ethyl acetate—cyclohexane (1—1)) and the fractions were collected with a R_f=0.14. After crystallization from methylene chloride and cyclohexane, 4.37 g of the expected product melting at 130° C. were obtained

Analysis; C ₁₅ H ₁₄ F ₃ N ₃ O ₂ S; molecular weight = 357.36					
	C %	H %	F %	N %	S %
Calculated	50.42	3.95	15.95	11.76	8.97
Found	50.3	3.9	15.9	11.6	8.9

IR Spectrum (CHCl ₃);	
OH	3626 cm ⁻¹
C=N	2236 cm ⁻¹
C=O	1763 cm ⁻¹
Aromatics	1615-1578-1504 cm ⁻¹

EXAMPLE 72

4-(4,4-dimethyl-3-(2-hydroxyethyl)-5-imino-2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl)-5-³H-benzonitrile

a) Preparation of the tritiated benzonitrile

15 mg of 2-trifluoromethyl 4-amino 5-bromo benzonitrile were mixed with 200 μl of ethyl acetate in the presence of 6.5 μl of triethylamine and 2 mg of palladium on activated charcoal and then tritium (1.42 bar) was introduced. After filtering, rinsing with ethyl acetate and evaporating to dryness at ambient temperature, approximately 66.6 G.Bq (1.8 Ci) of product were obtained.

b) Preparation of the tritiated isothiocyanate

150 μl of a 10% solution of thiophosgene in chloroform were added to the above product, in 150 μl of water and the mixture was stirred for 45 minutes at ambient temperature. Dilution was carried out with 0.5 ml of water and 1 ml of chloroform, followed by extraction with chloroform. The solvent was evaporated off under reduced pressure and the residue was taken up in toluene to obtain 50.7 G.Bq (1.37 Ci) of the expected product which was kept at -80° C.

c) Preparation of the tritiated imine

Having eliminated the toluene from the above mixture under reduced pressure, 130 μl of tetrahydrofuran with 1% triethylamine were added and 13 μl of 2-[(2-hydroxyethyl)-amino] 2-methylpropane-nitrile (Example 22)—were added. Then, another 130 μl of tetrahydrofuran with 1% triethylamine were added and the mixture was stirred for 30 minutes at ambient temperature and the solvents were eliminated under reduced pressure.

Preparation of the 2-trifluoromethyl 4-amino 5-bromo benzonitrile used in Example 72.

A solution of 2-trifluoromethyl 4-amino benzonitrile (prepared according to EP 0002892) (5 moles) in 25 ml of methanol was cooled to 0° C. and bismine was added (5.2 moles). The mixture was allowed to return to ambient temperature, stirred for 3 hours, alkalized with triethylamine and then an aqueous solution of sodium thiosulfate was added. The solvents were eliminated and extraction was carried out with chloroform. The organic phase was washed with water, dried, and the solvent was evaporated to obtain the product which was used as is for the following stage.

IR Spectrum (CHCl ₃);	
NH ₂	3612-3408 cm ⁻¹
C=N	2230 cm ⁻¹
Aromatics	1621-1556-1506 cm ⁻¹

EXAMPLE 73

4-(4,4-dimethyl-3-(2-hydroxyethyl)-5-oxo-2-thioxo-1-imidazolidinyl)-2-(trifluoromethyl) 5-³H-benzonitrile

The product of Example 72 in 180 μ l of water was heated to 100° C. and 60 μ l of 2 N hydrochloric acid was added. The mixture was stirred for 5 minutes at reflux and then approximately 600 mg of ice were added. Extraction is carried out with ethyl acetate and the extracts were washed with salt water and dried to obtain 34.7 G.Bq (937 mCi) of product. After chromatography on silica (eluant: cyclohexane—ethyl acetate (60-40)), 19 G.Bq (513 mCi) of the expected product were obtained.

EXAMPLE 74

4-(4,4-dimethyl-3-hydroxypropyl)-5-imino-2-thioxo-1-imidazolidinyl) 2-(trifluoromethyl)-benzonitrile

Using the procedure of Example 22—2 g of the isothio-cyanate of Example 70 (a) and 1.2 g of the appropriate aminonitrile were reacted to obtain 1.70 g of the expected product with a Rf=0.25 (methylene chloride—acetone (65-35)).

IR Spectrum (CHCl ₃);	
OH	3630 cm ⁻¹
=NH	3314-1676 cm ⁻¹
C=N	2235 cm ⁻¹

EXAMPLE 75

4-(4,4-dimethyl-3-(3-hydroxypropyl)-5-oxo-2-thioxo-1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

Using the procedure of Example 71, 240 mg of the product of Example 74 were reacted to obtain 226 mg of the expected product melting at 149°-150° C. with a Rf=0.32 (eluant; methylene chloride—acetone (75-25)).

IR Spectrum (CHCl ₃);	
OH	3626 cm ⁻¹
C=O	1763 cm ⁻¹
C=N	2236 cm ⁻¹
Aromatics	1613-1580-1504-1483 cm ⁻¹

EXAMPLE 76

4-(4,4-dimethyl 3-(4-hydroxybutyl)-5-imino 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

Using the procedure of Example 22,—2 g of isothio-cyanate and 1.38 g of the appropriate aminonitrile were reacted to obtain 2.08 g of the expected product with a Rf=0.25 (methylene chloride—acetone (65-35)).

IR Spectrum (CHCl ₃);	
OH	3630 cm ⁻¹
=NH	3314-1675 cm ⁻¹
C=N	2235 cm ⁻¹
Aromatics	1614-1577-1504 cm ⁻¹

EXAMPLE 77

4-(4,4-dimethyl 3-(4-hydroxybutyl)-5-oxo 2-thioxo-1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

Using the procedure of Example 71, 300 mg of the product of Example 76 were reacted to obtain 236 mg of the expected product melting at 78°-79° C. with a Rf=0.31 (eluant: methylene chloride—acetone (75-25)).

IR Spectrum (CHCl ₃);	
OH	3624 cm ⁻¹
C=O	1762 cm ⁻¹
C=N	2237 cm ⁻¹
Aromatics	1615-1580-1504 cm ⁻¹

UV Spectrum (EtOH)	
Max. 232 nm	$\epsilon = 19500$
Max. 254 nm	$\epsilon = 24000$
Infl. 266 nm	

EXAMPLE 78

4-(4,4-dimethyl 3-(2-methoxyethyl) 5-imino 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

Using the procedure of Example 22,—2.5 g of isothio-cyanate and 1.56 g of the appropriate aminonitrile were reacted to obtain 2.36 g of the expected product with a Rf=0.23 (methylene chloride—acetone (92.5-7.5)).

IR Spectrum (CHCl ₃);	
=NH	3314 cm ⁻¹
C=N	2236 cm ⁻¹
Aromatics	1614-1578-1504 cm ⁻¹
C=N	1675 cm ⁻¹

EXAMPLE 79

4-(4,4-dimethyl 3-(2-methoxyethyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

Using the procedure of Example 71, the product of Example 78 was reacted to obtain the expected product melting at 98°-99° C. with a Rf=0.32 (eluant: methylene chloride—acetone (99-1))

IR Spectrum (CHCl ₃);	
C=O	1757 cm ⁻¹
C=N	2236 cm ⁻¹
Aromatics	1615-1580-1504 cm ⁻¹

UV Spectrum (EtOH)	
Max. 232 nm	$\epsilon = 18200$
Max. 254 nm	$\epsilon = 22400$
Infl. 265 nm	

EXAMPLE 80

4-(4,4-dimethyl 3-(1-methylethyl) 5-imino 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

Using the procedure of Example 22,—2.5 g of the isothiocyanate and 1.32 g of the appropriate aminonitrile were reacted to obtain 880 mg of the expected product with a $R_f=0.20$ (eluant: methylene chloride—acetone (96-4)).

IR Spectrum (CHCl ₃);	
=NH	3310-1675 cm ⁻¹
C=N	2236 cm ⁻¹
Aromatics	1614-1580-1504 cm ⁻¹

EXAMPLE 81

4-(4,4-dimethyl 3-(1-methylethyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

Using the procedure of Example 71, 880 mg of the product of Example 80 and 35 ml of 6 N hydrochloric acid were reacted to obtain after extraction with chloroform, 744 mg of the expected product melting at 203°-204° C. with a $R_f=0.45$ (eluant: cyclohexane—ethyl acetate (1—1)).

IR Spectrum (CHCl ₃);	
OH	3626 cm ⁻¹
C=O	1753 cm ⁻¹
C=N	2232 cm ⁻¹
Aromatics	1615-1580-1504 cm ⁻¹

UV Spectrum (EtOH)	
Max. 232 nm	$\epsilon = 18900$
Max. 235 nm	$\epsilon = 22500$
Infl. 273 nm	

EXAMPLE 82

3-(3,4-dichlorophenyl 5,5-dimethyl 1-(3-hydroxypropyl) 4-imino 2-imidazolidine thione

Using the procedure of Example 51,—2.4 g of 3,4-dichlorophenyl isocyanate and 1.6 g of the appropriate aminonitrile were reacted to obtain, after chromatography on silica (eluant: methylene chloride—acetone (6-4)), 2.16 g of expected product with a $R_f=0.25$

IR Spectrum (CHCl ₃);	
OH	3630 cm ⁻¹ + associated
C=NH	3294-1676 cm ⁻¹ (F)
Aromatics	1595-1569-1482 cm ⁻¹

EXAMPLE 83

3-(3,4-dichlorophenyl 5,5-dimethyl 1-(3-hydroxypropyl) 2-thioxo 4-imidazolidinone

Using the procedure of Example 52,—0.88 g of the product of Example 82 and 35 ml of 6 N hydrochloric acid were reacted to obtain, after extraction with chloroform, 0.79 g of the expected product melting at 202°-203° C.

IR Spectrum (CHCl₃);

C=O	1753 cm ⁻¹
C=N	2232 cm ⁻¹
Aromatics	1615-1580-1504 cm ⁻¹

UV Spectrum (EtOH)

Max. 232 nm	$\epsilon = 18900$
Max. 235 nm	$\epsilon = 22500$
Infl. 273 nm	

EXAMPLE 84

4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-imino 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) (5-³H) benzonitrile

a) 4-amino 2-(trifluoromethyl) (5-³H) benzonitrile

The following were cooled to -180° C. and mixed under an inert atmosphere: 16 mg of 2-trifluoromethyl 4-amino 5-bromo benzonitrile, 2 mg of palladium on activated charcoal, 200 μ l of ethyl acetate and 6.5 μ l of triethylamine. Then the mixture was left under a tritium atmosphere and taken to 20° C. and the pressure was then 1.68 bar. The mixture was stirred until absorption was complete ($p=0.42$ bar), followed by cooling to -180° C. The excess tritium was recovered, taken to 20° C. and then filtered. The filtrate was rinsed with ethyl acetate and concentrated at 40° C. under reduced pressure to obtain 68 G.Bq of the expected product.

b) 4-thioisocyanate 2-(trifluoromethyl) (5-³H) benzonitrile

The following were mixed under an argon atmosphere: 34 G.Bq of the above tritiated amino derivative, 150 μ l of demineralized water and 150 μ l of 10% thiophosgene solution in chloroform. The mixture was stirred at 20° C. for 45 minutes, decanted and reextraction was carried out with chloroform. The extracts were dried over magnesium sulfate, filtered and concentrated under reduced pressure. The thioisocyanate obtained was used as is for the following stage.

c) 4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-imino 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) (5-³H) benzonitrile

The following were mixed under an argon atmosphere with the thioisocyanate of stage b): 350 μ l of tetrahydrofuran with 1% triethylamine and 20 μ l of propanonitrile prepared as indicated below. The mixture was stirred for 2 hours at 20° C., followed by concentration at 20° C. under reduced pressure. The imine was used as is for the following stage. Preparation of the 2-(4-hydroxybutylamino) 2-methylpropano-nitrile used in stage c)

550 μ l of acetone cyanohydrin and 500 μ l of 4-amino 1-butanol were mixed together and the mixture was stirred for 16 hours at 20° C. to obtain the desired product which was used as is for the following stage.

EXAMPLE 85

4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-oxo 2-thioxo
1-imidazolidinyl) 2-(trifluoromethyl) (5-³H)
benzonitrile

200 μl of 2 N hydrochloric acid were added to the imine of Example 84 and the mixture was refluxed for 5 minutes, then returned to 20° C. and diluted with 1 ml of water. Extraction was carried out with ethyl acetate and the extracts were washed with water and concentrated under reduced pressure. The crude product was purified by chromatography on silica (eluant: cyclohexane—ethyl acetate (6-4)) to obtain 2.8 G.Bq of the expected product.

EXAMPLE 86

4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-imino 2-thioxo
1-imidazolidinyl) 2-(trifluoromethyl) benzo (¹⁴C) nitrile

a) 4-amino 2-(trifluoromethyl) benzo (¹⁴C) nitrile

377 mg of cuprous cyanide ¹⁴C (9 G.Bq) and 1.0732 g of 4-bromo 3-(trifluoromethyl) benzenamine were mixed together under a nitrogen atmosphere in 8 ml of dimethylformamide and the mixture was refluxed for 4 hours, then cooled to 0° C. and diluted with 20 ml of acetone. The insoluble part was filtered off and the filtrate was concentrated at 70° C. under reduced pressure. The residue was taken up in methylene chloride, filtered and the filtrate was concentrated under reduced pressure. The benzonitrile (¹⁴C) was purified by chromatography on silica (eluant: methylene chloride—cyclohexane (70-30)) to obtain 0.558 g (6.62 G.Bq) of the expected product.

b) 4-thioisocyanate 2-(trifluoromethyl) benzo (¹⁴C) nitrile

The following were mixed under a nitrogen atmosphere: 189 mg of benzonitrile (¹⁴C) of stage a), 2.7 ml of water and 85 μl of thiophosgene. The mixture was agitated vigorously stirred for 5 minutes, and after 30 μl of thiophosgene were added, stirring was continued for one hour at 20° C. Then extraction was carried out with chloroform and the extracts were washed with water, dried and concentrated under reduced pressure. The thioisocyanate obtained was used as is for the following stage.

c) 4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-imino 2-thioxo
1-imidazolidinyl) 2-(trifluoromethyl) benzo (¹⁴C) nitrile

2 ml of tetrahydrofuran, the propanonitrile prepared below in solution in 1.5 ml of methylene chloride and 150 μl of triethylamine were added under a nitrogen atmosphere to the thioisocyanate of stage b). The mixture was heated for 30 minutes under reflux and concentrated under reduced pressure to obtain the imine which was used as is for the following stage.

Preparation of the 2-(4-hydroxybutylamino)
2-methylpropano-nitrile of stage c

220 μl of acetone cyanohydrin and 200 μl of 4-amino 1-butanol were mixed together with stirring for 16 hours at 20° C. and then was diluted with 2 ml of methylene chloride, dried, filtered and the filtrate was concentrated under reduced pressure to obtain the propanonitrile which was used as is for the following stage.

EXAMPLE 87

4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-oxo 2-thioxo
1-imidazolidinyl) 2-(trifluoromethyl) benzo (¹⁴C) nitrile

6 ml of methanol and 1.6 ml of 2 N hydrochloric acid were added to the imine of Example 86 and the mixture was refluxed for 45 minutes, cooled to 20° C. and diluted with 10 ml of water. Extraction was carried out with methylene chloride and the extracts were washed with water and concentrated under reduced pressure. The crude product was purified by chromatography on silica (eluant: ether—acetonitrile—cyclohexane (50-15-35)) to obtain 328 mg of the expected product.

EXAMPLE 88

4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-imino 2-oxo
1-imidazolidinyl) 2-(trifluoromethyl) (5-³H)
benzonitrile

a) 4-amino 2-(trifluoromethyl) (5-³H) benzonitrile

Using the procedure of stage a) of Example 84, 16 mg of 4-amino 5-bromo 2-trifluoromethyl benzonitrile, 2 mg of palladium on activated charcoal, 200 μl of ethyl acetate and 6.5 μl of triethylamine were reacted to obtain 68 G.Bq of the expected product.

b) 4-isocyanate 2-(trifluoromethyl) (5-³H) benzonitrile

34 G.Bq of tritiated amino derivate of step a) and 100 μl of 20% phosgene in toluene were mixed together under an argon atmosphere and the mixture was taken to 80° C. for one hour. A further 100 μl of phosgene were added and the mixture heated for one hour at 80° C. This operation was repeated one more time, then concentration was carried out at 20° C. under reduced pressure to obtain the isocyanate which was used as is for the following stage.

c) 4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-imino 2-oxo
1-imidazolidinyl) 2-(trifluoromethyl) (5-³H)
benzonitrile

The following were added under an argon atmosphere to the isocyanate of stage b): 200 μl of methylene chloride, 50 μl of the propanonitrile chloromethylene solution prepared as below and 20 μl of triethylamine and the mixture was stirred for 30 minutes. A further 50 μl of the propanonitrile solution were added and stirring was continued for 30 minutes followed by concentration at 20° C. under reduced pressure. The imine was used as is for the following stage. Preparation of the 2-(4-hydroxybutylamino) 2-methyl propano-nitrile, of stage c)

220 μl of acetone cyanohydrin and 200 μl of 4-amino 1-butanol were mixed together and the mixture was stirred for 16 hours at 20° C., then diluted with 3 ml of methylene chloride and dried over magnesium sulfate. The decanted solution was used as is for the following stage.

EXAMPLE 89

4-(4,4-dimethyl 2,5-dioxo 3-(4-hydroxybutyl)
1-imidazolidinyl) 2-(trifluoromethyl) (5-³H)
benzonitrile

200 μl of methanol and 50 μl of 2 N hydrochloric acid were added to the imine of Example 88 and the mixture was refluxed for 45 minutes, then returned to 20° C. and diluted with 1 ml of water. Extraction was carried out with methylene chloride and the extracts were washed

with water and concentrated at 20° C. under reduced pressure. The crude product was purified by chromatography on silica (eluant: methylene chloride—ethyl acetate (7-3 then 5—5)) to obtain 16 G.Bq of the expected product.

EXAMPLE 90

4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-imino 2-oxo 1-imidazolidinyl) 2-(trifluoromethyl) benzo (¹⁴C) nitrile

a) 4-amino 2-(trifluoromethyl) benzo (¹⁴C) nitrile

Using the procedure of Example 86, stage a), 377 mg of cuprous cyanide ¹⁴C, 1.0732 g of 4-bromo 3-trifluoromethyl benzenamine and 8 ml of dimethylformamide were reacted to obtain 0.558 g (6.62 G.Bq) of the expected product.

b) 4-isocyanato 2-(trifluoromethyl) benzo (¹⁴C) nitrile

182.4 mg of benzonitrile (¹⁴C) (0.97 mmole), 2 ml of dioxane and 1 ml of 20% phosgene in toluene were mixed together under a nitrogen atmosphere and the solution was heated at 60° C. for 22 hours, then concentrated at 60° C. under reduced pressure. The isocyanate was used as is for the following stage.

c) 4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-imino 2-oxo 1-imidazolidinyl) 2-(trifluoromethyl) benzo (¹⁴C) nitrile

1.5 ml of methylene chloride (on siliporite NK 30), the propanonitrile of Example 88 in solution in 1.5 ml of methylene chloride, and 150 μl of triethylamine were added under a nitrogen atmosphere to the isocyanate of stage b). The mixture was stirred for one hour at 20° C. and concentrated under reduced pressure. The imine was used as is for the following stage.

EXAMPLE 91

4-(4,4-dimethyl 2,5-dioxo 3-(4-hydroxybutyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzo (¹⁴C) nitrile

5 ml of methanol and 1.2 ml of 1 N hydrochloric acid were added to the imine of Example 90 and the mixture was refluxed for 40 minutes, then returned to 20° C. and diluted with 10 ml of water. Extraction was carried out with methylene chloride and the extracts were washed with water and concentrated under reduced pressure. The crude product was purified by chromatography on silica (eluant: ether—acetonitrile—cyclohexane (50-15-35)) to obtain 289 mg (1.26 G.Bq) of the expected product.

EXAMPLE 92

4-(2,5-dioxo 4,4-dimethyl 3-(4-triphenylmethoxy-butyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

370 mg of the product of Example 58, 307 mg of trityl chloride in the presence of 10 mg of 4-dimethylaminopyridine, 0.25 ml of triethylamine and 4 ml of dimethylformamide were stirred at ambient temperature for 16 hours. The mixture was heated to 40° C. for 4 hours, poured into water and extraction was carried out with ether. The extracts were washed with water and dried and the solvent was eliminated under reduced pressure. The residue was chromatographed on silica (eluant: cyclohexane—ethyl acetate 75-25) to obtain 467 mg of the expected product with a R_f=0.25.

IR Spectrum (CHCl₃);

C=O 1778, 1725 cm⁻¹ (F)

-continued

IR Spectrum (CHCl₃);

C=N 2235 cm⁻¹
Aromatics 1615, 1597, 1505, 1490 cm⁻¹

EXAMPLE 93

4-(2,5-dioxo 4,4-dimethyl 3-(4-phenylmethoxy-butyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

48 mg of sodium hydride were added in several lots to 370 mg of the product of Example 58 in solution in 4 ml of dimethylformamide and the mixture was stirred for 30 minutes. Then, 0.12 ml of benzyl bromide and 40 mg of tetrabutylammonium iodide were added and after 90 minutes of reaction, the same amount of each reagent was added. The mixture was stirred for one hour and the reaction medium was poured into an ice-cooled aqueous solution of monopotassium phosphate. Extraction was carried out with ether and the extracts were washed with water and dried. The solvent was eliminated under reduced pressure and the residue was chromatographed on silica (eluant: methylene chloride—acetone 99-1) to obtain 140 mg of the expected product melting at 75°-76° C.

IR Spectrum (CHCl₃);

C=O 1779, 1725 cm⁻¹
C=N 2235 cm⁻¹
Aromatics 1615, 1580, 1505, 1497 cm⁻¹

EXAMPLE 94

4-[4,4-dimethyl 2,5-dioxo 3-(4-methoxybutyl) 1-imidazolidinyl] 2-(trifluoromethyl) benzonitrile

50 mg of sodium hydride were added in several lots to 370 mg of the product of Example 58 in solution in 3 ml of dimethylformamide and the mixture was stirred for 20 minutes. 0.06 ml of methyl iodide were added and the mixture was stirred for one hour. A further 50 mg of sodium hydride were added and then after 20 minutes, 0.06 ml of methyl iodide were added. The reaction medium was poured into water and extracted with ether. The extracts were washed with water, dried and the solvent was evaporated. The residue was chromatographed on silica (eluant: methylene chloride—acetone 98-2) to obtain 135 mg of the expected product melting at 80°-81° C.

IR Spectrum (CHCl₃);

C=O 1779, 1725 cm⁻¹ (F)
C=N 2234 cm⁻¹
Aromatics 1616, 1576, 1505 cm⁻¹
OCH₃ approx. 2830 cm⁻¹

EXAMPLE 95

4-[3-(4-chlorobutyl) 4,4-dimethyl 2,5-dioxo 1-imidazolidinyl] 2-(trifluoromethyl) benzonitrile


Using the procedure of Example 59, 600 mg of the product of Example 8—and 660 mg of 1-chloro 4-iodobutane in solution in 1 ml of dimethylformamide cooled down to +5° C. were reacted to obtain 604 mg of the expected product melting at 80°-81° C.

IR Spectrum (CHCl ₃);	
C=O	1779, 1725 cm ⁻¹ (F)
C≡N	2238 cm ⁻¹
Aromatics	1616, 1575, 1505 cm ⁻¹

EXAMPLE 96

4-[3-[4-[(methylsulphonyl) oxy] butyl] 4,4-dimethyl 2,5-dioxo 1-imidazolidinyl] 2-(trifluoromethyl) benzonitrile

0.17 ml of methanesulfonyl chloride were added to 740 mg of the product of Example 58 in solution in 7.4 ml of pyridine and 24 mg of 4-dimethylamino-pyridine and the mixture was stirred for one hour. The mixture was poured into ice-cooled water and extraction was carried out with methylene chloride. The extracts were washed with water and the residual pyridine was eliminated by distillation. The residue was chromatographed on silica (eluant: methylene chloride—ethyl acetate 8-2) to obtain 771 mg of the expected product.

IR Spectrum (CHCl ₃);	
C=O	1779, 1725 cm ⁻¹
C≡N	2235 cm ⁻¹
Aromatics	1615, 1575, 1505 cm ⁻¹
	1361, 1175 cm ⁻¹

UV Spectrum (EtOH)		
max.	261 nm	ε = 14900
infl.	279-297 nm	

EXAMPLE 97

4-(3-acetyl 4,4-dimethyl 2,5-dioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

Using the procedure of Example 59, 420 mg of the product of Example 8—and two lots of 0.1 ml of acetyl chloride were reacted to obtain after chromatography on silica (eluant: methylene chloride—ethyl acetate 98-2), 334 mg of the expected product melting at 129°-130° C.

IR Spectrum (CHCl ₃);	
C=O	1800, 1740, 1717 cm ⁻¹
C≡N	2240 cm ⁻¹
Aromatics	1616, 1505 cm ⁻¹

UV Spectrum (EtOH)		
max	250 nm	ε = 12000
infl.	274-284 nm	

EXAMPLE 98

4-(3-benzoyl 4,4-dimethyl 2,5-dioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

Using the procedure of Example 59, 300 mg of the product of Example 8—and two lots of 0.12 ml of benzoyl chloride in solution in 0.5 ml of dimethylformamide were reacted to obtain after chromatography on silica (eluant: cyclohexane—ethyl acetate 8-2), 285 mg of the expected product melting at 179°-180° C.

IR Spectrum (CHCl ₃);	
C=O	1800, 1780, 1746, 1699 cm ⁻¹
C≡N	2235 cm ⁻¹
Aromatics	1617, 1600, 1580, 1504 cm ⁻¹

UV Spectrum (EtOH)	
max. 250 nm	ε = 28500
infl. 275 nm	ε = 6500
infl. 263 nm	ε = 3850

EXAMPLE 99

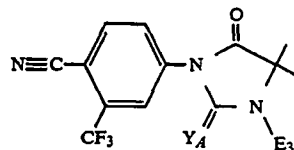
4-[3-[dimethyl (1,1-dimethylethyl) silyl] 4,4-dimethyl 2,5-dioxo 1-imidazolidinyl] 2-(trifluoromethyl) benzonitrile

Using the procedure of Example 59, 450 mg of the product of Example 8—and 300 mg of dimethyl t-butylsilyl chloride in 2 ml of dimethylformamide were reacted to obtain after chromatography on silica (eluant: methylene chloride—acetone 99-1), 527 mg of the expected product melting at 147°-148° C.

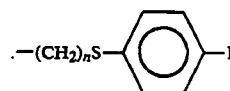
IR Spectrum (CHCl ₃);	
C≡N	2236 cm ⁻¹
Aromatics	1615, 1579, 1505 cm ⁻¹

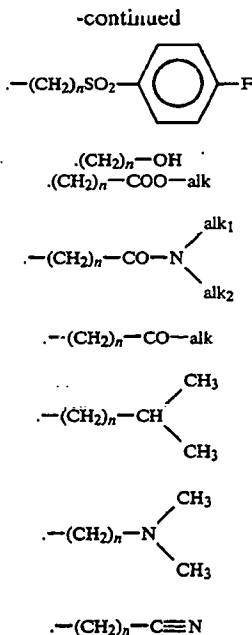
UV Spectrum (EtOH)	
max. 258 nm	ε = 17000
infl. 275-285 nm	

In addition to the products described above, the following products are products which can be obtained within the scope of the present invention, namely the products of the formula:



in which Y_A is oxygen or sulfur and R_{3A} has the following values:





alk, alk₁ and alk₂ are alkyl of 1 to 4 carbon atoms and n is an integer between 1 and 4.

EXAMPLE 100

Tablets were prepared with a composition of 100 mg of 4-(5-oxo-2-thioxo-3,4,4-trimethyl-1-imidazoliny)-2-trifluoromethyl-benzonitrile and sufficient excipient of lactose, starch, talc and magnesium stearate for a final tablet weight of 300 mg.

PHARMACOLOGICAL DATA

Study of the affinity of the products of the invention for the androgenic receptor.

1) Androgenic receptor

Male rats of the Sprague Dawley EOPS strain weighing 180 to 200 g, castrated 24 hours previously, were killed and the prostate was removed, weighed and homogenized at 0° C. with a potter glass in a buffered solution (Tris 10 mM, saccharose 0.25 M, PMSF (phenyl methane sulfonyl fluoride) 0.1 mM, sodium molybdate 20 mM, HCl pH 7.4 into which was added extemporaneously 2 M of DTT (DL dithiothreitol) at a rate of 1 g of tissue per 8 ml of buffer solution. The homogenate was then ultracentrifuged at 0° C. for 45 minutes at 105,000 g and the aliquots of supernatant (=cytosol) were incubated for 30 minutes and 24 hours with a concentration (T) of tritiated testosterone and in the presence of increasing concentrations (0 to 2500.10⁻⁹M) of cold testosterone or the test products. The concentration of bound tritiated Testosterone (B) was then measured for each incubate by adsorption method of carbon-dextran. The relative affinity of bonding (RBA) was calculated.

The following two curves were graphed: the percentage of the bound tritiated hormone B/T as a logarithm function of the concentration of the cold hormone and B/T as a logarithm function of the concentration of the tested cold product. The line of the equation

$$I_{50} = \frac{(B/T_{max} + B/T_{min})}{2}$$

was determined. B/T max = % of the bound tritiated hormone for an incubation of this tritiated hormone at the concentration (T). B/T min = % of the bound tritiated hormone for an incubation of this tritiated hormone at the concentration (T) in the presence of a large excess of cold hormone (2,500 × 10⁻⁹M).

The intersections of the straight line I₅₀ and the curves permit an evaluation of the concentrations of the cold reference hormones (CH) and the cold test product (CX) which inhibit by 50% the bonding of the tritiated hormone on the receptor. The RBA of the test product was determined by the equation

$$RBA = (CH)/(CX)$$

and the following results expressed in ARL were obtained with testosterone = 100.

	Incubation 30 minutes	Incubation 24 hours
Product Example 1	27.5	3
Product Example 2	22	6
Product Example 4	21	5
Product Example 11	28	8
Product Example 12	128	92
Product Example 13	31	39
Product Example 14	27	7
Product Example 15	69	24

2) Study of the affinity of the products of the invention for the androgen receptor.

Male rats of the Sprague Dawley EOPS strain weighing 180 to 200 g, castrated 24 hours previously, were killed and the prostate was removed, weighed and homogenized at 0° C. with a potter glass in a buffered solution (Tris 10 mM, saccharose 0.25 M, PMSF (phenyl methane sulfonyl fluoride) 0.1 mM, sodium molybdate 20 mM, HCl pH 7.4 into which was added extemporaneously 2 mM of DTT (DL dithiothreitol) at a rate of 1 g of tissue per 8 ml of buffer solution. The homogenate was then ultracentrifuged at 0° C. for 30 minutes at 209,000 g and the aliquots of supernatant (=cytosol) were incubated for 30 minutes and 24 hours with a concentration (T) of tritiated testosterone and in the presence of increasing concentrations (0 to 2500.10⁻⁹M) of cold testosterone or the test products. The concentration of bound tritiated Testosterone (B) was then measured for each incubate by adsorption method of carbon-dextran. The relative affinity of bonding (RBA) was calculated.

The following two curves were graphed: the percentage of the bound tritiated hormone B/T as a logarithm function of the concentration of the cold hormone and B/T as a logarithm function of the concentration of the tested cold product. The line of the equation

$$I_{50} = \frac{(B/T_{max} + B/T_{min})}{2}$$

was determined. B/T max = % of the bound tritiated hormone for an incubation of this tritiated hormone at the concentration (T). B/T min = % of the bound tritiated hormone for an incubation of this tritiated hor-

none at the concentration (T) in the presence of a large excess of cold hormone ($2,500 \times 10^{-9}M$).

The intersections of the straight line I_{50} and the curves permit an evaluation of the concentrations of the cold reference hormones (CH) and the cold test product (CX) which inhibit by 50% the bonding of the tritiated hormone on the receptor. The RBA of the test product was determined by the equation

$$RBA = 100(CH)/(CX)$$

and the following results expressed in RBA were obtained with testosterone = 100.

	Incubation 24 hours
Example 59	31
Example 71	163
Example 77	300
Example 79	81
Example 81	28

3) Determination of the androgen or anti-androgen activity by the dosage of ornithine carboxylase.

Six week old male Swiss mice castrated 24 hours receive oral does of the test products as a 0.5% suspension in methyl cellulose simultaneously with a subcutaneous injection of 3 mg/kg of testosterone propionate in solution in sesame oil containing 5% of benzyl alcohol to determine the anti-androgen activity. Active agonists were determined in the absence of testosterone propionate. The test compounds as well as testosterone were administered in a volume of 10 ml/kg. 16 hours after the treatments, the animals were killed, the kidneys were removed and then homogenized at 0° C. with a teflon-glass grinding apparatus in 10 volumes of buffer Tris-HCl 50 mM at a pH 7.4 containing 250 mM of pyridoxal phosphate, 0.1 mM EDTA and 5 mM of dithiothreitol. The homogenate was centrifuged at 105,000 g for 45 minutes.

At 37° C., renal ornithine decarboxylase transforms an isotropic mixture of cold ornithine and tritiated ornithine in cold putrescine and tritiated putrescine. The putrescine was then collected on selective ion-exchange papers. After drying, excess non-transformed cold and tritiated ornithine were eliminated by washing 3 times with 0.1 M ammonium hydroxide. The papers were dried and the radioactivity was determined after addition of an Aqualite sample. The results expressed in fmoles ($10^{-15}M$) of tritiated putrescine formed per hour mg of protein are reported in the following Table

PRODUCT OF EXAMPLE	ANTAGONISM IN MG/KG	PERCENT
11	3	83
12	0.1	12
	0.3	36
	1	68
	3	94
	10	99
12	(Agonism) 10	0
14	Antagonism 3	87
15	0.3	4
	1	82

4) Determination of the androgen or anti-androgen activity by the dosage of ornithine carboxylase.

Swiss six week old male mice castrated 24 hours received oral or percutaneous doses of the test products as a 0.5% suspension in methyl cellulose or in ethanol simultaneously with a sub-cutaneous injection of 3 mg/kg of testosterone propionate in solution in corn oil to determine the anti-androgen activity. Active agonists were determined in the absence of testosterone propionate. The test compounds as well as testosterone were administered in a volume of 10 ml/kg. 20 hours after the treatments, the animals were killed, the kidneys were removed and then homogenized at 0° C. with a teflon-glass grinding apparatus in 10 volumes of buffer Tris-HCl 50 mM at a pH 7.4 containing 250 mM of pyridoxal phosphate, 0.1 mM EDTA and 5 mM of dithiothreitol. The homogenate was centrifuged at 209,000 g for 45 minutes.

Principle of dosage

At 37° C., renal ornithine decarboxylase transforms an isotopic mixture of cold ornithine and tritiated ornithine in cold putrescine and tritiated putrescine. The putrescine was then collected on selective ion-exchange papers. After drying, excess non-transformed cold and tritiated ornithine were eliminated by washing 3 times with 0.1 M ammonium hydroxide. The papers were dried and the radioactivity was determined after addition of an Aqualite sample. The results expressed in fmoles ($10^{-15}M$) of tritiated putrescine formed per hour/mg of protein are reported in the following Table.

The same test were repeated with the following changes:

Test A: the products were administered percutaneously at 1.5 mg/kg at a volume of 10 μ l.

Test B: the products were administered orally at 1 mg/kg.

Test C: the products are administered orally at 3 mg/kg. The results are in the following Table.

The results are expressed in % of inhibition of the ODL the samples receiving only the testosterone propionate:

Products of example	ODL		
	Test A	Test B	Test C
58	40	36	
71	32		67
75	41		
78	78		
80	62		
81	35		
83	58		

CONCLUSION

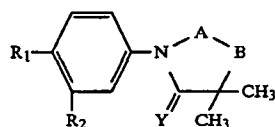
The tests show that the tested compounds of the invention possess a strong anti-androgen activity and do not have agonist activity.

Various modifications of the compounds and method of the invention may be made without departing from the spirit or scope thereof and it is to be understood that the invention is intended to be limited only as defined in the appended claims.

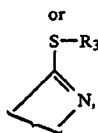
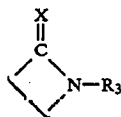
We claim:

1. A compound selected from the group consisting of a compound of the formula

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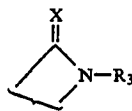
wherein R_1 is selected from the group consisting of $-\text{CN}$, $-\text{NO}_2$ and halogen, R_2 is $-\text{CF}_3$ or halogen, $-\text{A-B}$ is



X is $-\text{O}-$ or $-\text{S}-$, R_3 is selected from the group consisting of a) hydrogen, b) alkyl, alkenyl and alkynyl of up to 12 carbon atoms, c) phenyl and phenylalkyl unsubstituted or substituted with at least one member of the group consisting of $-\text{OH}$, halogen, $-\text{OCH}_3$, $-\text{CN}$ and haloalkyl, d) acyl of an organic carboxylic acid of up to 7 carbon atoms, e) free or salted carboxy, carboxy esterified with alkyl and amidified carboxy, f) amino and mono and dialkylamino of 1 to 4 carbon atoms and g) $-\text{S}-$ phenyl unsubstituted or substituted with at least one member of the group consisting of $-\text{CF}_3$ and alkyl, alkenyl, alkoxy, alkenyloxy, alkynyl and alkynyloxy of up to 12 carbon atoms with the sulfur unoxidized or oxidized to sulfone or sulfoxide, the alkyl, alkenyl and alkynyl being uninterrupted or interrupted with oxygen, sulfur or nitrogen and Y is $-\text{O}-$, $-\text{S}-$ or $-\text{NH}-$ with the provisos that when X is oxygen, R_3 is hydrogen and Y is $-\text{O}-$ or $-\text{NH}-$, then R_1 is NO_2 or $-\text{CN}$ and when X is sulfur and Y is $-\text{O}-$ then at least one of the following conditions is satisfied, R_1 is $-\text{CN}$ and R_2 is $-\text{CF}_3$ and their non-toxic, pharmaceutically acceptable acid addition salts.

2. A compound of claim 1 wherein Y is oxygen.

3. A compound of claim 1 wherein $-\text{A-B}-$ is



and X is sulfur.

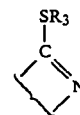
4. A compound of claim 3 wherein R_3 is hydrogen or alkyl of 1 to 4 carbon atoms optionally substituted with a $-\text{OH}$ or methoxy.

5. A compound of claim 1 wherein R_1 is $-\text{CN}$ or halogen.

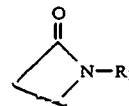
6. A compound of claim 1 wherein R_1 wherein R_1 is chlorine.

7. A compound of claim 1 wherein $-\text{A-B}-$ is

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or



and R_3 is alkyl or alkenyl of up to 6 carbon atoms unsubstituted or substituted or uninterrupted or interrupted by oxygen or unoxidized or oxidized sulfur or unsubstituted or substituted aralkyl or acyl or trialkylsilyl.

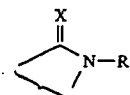
8. A compound of claim 7 wherein R_3 is alkyl of 1 to 6 carbon atoms unsubstituted or substituted by at least one member of the group consisting of halogen, $-\text{OH}$, $-\text{O}$ acyl, carboxy, carboxy esterified with alkyl, a heterocycle, O-alkyl and unoxidized or oxidized S-aryl with the aryl unsubstituted or substituted with at least one member of the group consisting of halogen and alkoxy.

9. A compound of claim 8 wherein R_3 is alkyl of 2 to 4 carbon atoms substituted by a member selected from the group consisting of chlorine, ethoxycarbonyl, tert-butoxy carbonyl, cyclopentylloxycarbonyl, unoxidized or oxidized 4-fluorophenylthio, morpholino, phenylmethoxy, triphenylmethoxy and methylsulfonyloxy.

10. A compound of claim 7 wherein R_3 is acetyl or benzoyl or (1,1-dimethylethyl)dimethylsilyl.

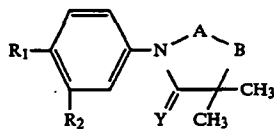
11. A compound of claim 1 selected from the group consisting of 4-(5-oxo-2-thioxo-3,4,4-trimethyl-1-imidazolidinyl)-2-(trifluoromethyl) benzonitrile, 4-(4,4-dimethyl-5-oxo-2-thioxo 1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile, 4,4,4-dimethyl-3-(2-hydroxyethyl)-5-oxo-2-thioxo-1-imidazolidinyl 2-(trifluoromethyl)-benzonitrile, 3-(3,4-dichlorophenyl)-2-thioxo-1,5,5-trimethyl-4-imidazolidinone, 1-(4-nitro-3-(trifluoromethyl) phenyl-3,4,4-trimethyl-2,5-imidazolidinedione, 4,4,5-dihydro-4,4-dimethyl-5-oxo-2-(phenylmethyl) thio-1H-imidazol-1-yl-2-(trifluoromethyl) benzonitrile, 4,4,4-dimethyl 3-(2-hydroxyethyl) 5-oxo 2-thioxo 1-imidazolidinyl 2-(trifluoromethyl) benzonitrile, 4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-oxo 2-thioxo 1-imidazolidinyl 2-(trifluoromethyl) benzonitrile, 3-(4-cyano 3-trifluoromethyl) phenyl) 5,5-dimethyl 2,4-dioxo 1-imidazolidinebutanoic acid and 4-(4,4-dimethyl 2,5-dioxo 3-(4-hydroxybutyl) 1-imidazolidinyl) 2-(trifluoromethyl benzonitrile.

12. A compound of claim 1 wherein Y is $-\text{O}-$ except the compounds wherein the $-\text{A-B}-$ group is

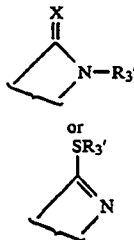


in which X is oxygen and R_3 is hydrogen, R_2 is halogen or trifluoromethyl and R_1 is nitro or halogen.

13. A compound of the formula



wherein R_1 , R_2 and Y have the definitions of claim 1, $-A-i-B_1$ is



Y is oxygen or sulfur and R'_3 is R_3 with any reactive functions protected.

14. An anti-androgenic composition comprising an anti-androgenically effective amount of at least one compound of claim 1 and an inert pharmaceutical carrier.

15. A composition of claim 14 wherein the active compound is selected from the group consisting of 4-(5-oxo-2-thioxo-3,4,4-trimethyl-1-imidazolidinyl) 2-(trifluoromethyl)-benzotrile, 4-(4,4-dimethyl-5-oxo-2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl)-benzotrile, 4-(4,4-dimethyl-3-(2-hydroxyethyl)-5-oxo-2-thioxo 1-imidazolidinyl-2-(trifluoromethyl)-benzotrile, 3-(3,4-dichlorophenyl) 2-thioxo--1,5,5-trimethyl-4-imidazolidinone, 1-(4-nitro-3-(trifluoromethyl)-phenyl-3,4,4-trimethyl-2,5-imidazoli-

16. A method of inducing anti-androgenic activity in warm-blooded animals comprising administering to warm-blooded animals an anti-androgenically effective amount of at least one compound of claim 1.
17. A method of claim 16 wherein Y is oxygen.
18. A method of claim 16 wherein R_1 is $-CN$ or halogen.
19. A method of claim 16 wherein R_1 is chlorine.
20. A method of claim 14 wherein the active compound is selected from the group consisting of 4-(5-oxo-2-thioxo-3,4,4-trimethyl-1-imidazolidinyl) 2-(trifluoromethyl)-benzotrile, 4-(4,4-dimethyl-5-oxo-2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl)-benzotrile, 4-(4,4-dimethyl-3-(2-hydroxyethyl) 5-oxo-2-thioxo-1-imidazolidinyl-2-(trifluoromethyl)-benzotrile, 3-(3,4-dichlorophenyl)-2-thioxo--1,5,5-trimethyl-4-imidazolidinone, 1-(4-nitro-3-(trifluoromethyl)-phenyl-3,4,4-trimethyl-2,5-imidazolidinedione, 4-4,5-dihydro-4,4-dimethyl-5-oxo-2-(phenylmethyl)-thio-1H-imidazol-1-yl-2-(trifluoromethyl) benzotrile-4,4,4-dimethyl 3-(2-hydroxyethyl) 5-oxo 2-thioxo 1-imidazolidinyl 2-(trifluoromethyl) benzotrile, 4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzotrile-3-(4-cyano 3-trifluoromethyl) phenyl) 5,5-dimethyl 2,4-dioxo 1-dioxo 3-(4-hydroxybutyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzotrile.
21. A method of inducing anti-androgenic activity in warm-blooded animals comprising administering to warm-blooded animals an anti-androgenically effective amount of at least one compound of claim 1.
22. A method of claim 21 wherein Y is oxygen.
23. A method of claim 21 wherein R_1 is $-CN$ or halogen.
24. A method of claim 21 wherein R_1 is chlorine.
25. A method of claim 21 wherein the active compound is selected from the group consisting of 4-(5-oxo-2-thioxo-3,4,4-trimethyl-1-imidazolidinyl) 2-(trifluoromethyl)-benzotrile, 4-(4,4-dimethyl-5-oxo-2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl)-benzotrile, 4-(4,4-dimethyl-3-(2-hydroxyethyl)-5-oxo-2-thioxo 1-imidazolidinyl-2-(trifluoromethyl)-benzotrile, 3-(3,4-dichlorophenyl) 2-thioxo--1,5,5-trimethyl-4-imidazolidinone, 1-(4-nitro-3-(trifluoromethyl)-phenyl-3,4,4-trimethyl-2,5-imidazolidinedione, 4-4,5-dihydro-4,4-dimethyl-5-oxo-2-(phenylmethyl)-thio-1H-imidazol-1-yl-2-(trifluoromethyl) benzotrile-4,4,4-dimethyl 3-(2-hydroxyethyl) 5-oxo 2-thioxo 1-imidazolidinyl 2-(trifluoromethyl) benzotrile, 4-(4,4-dimethyl 3-(4-hydroxybutyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzotrile-3-(4-cyano 3-trifluoromethyl) phenyl) 5,5-dimethyl 2,4-dioxo 1-imidazolidinebutanoic and acid and 4-(4,4-dimethyl 2,5-dioxo 3-(4-hydroxybutyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzotrile.

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US006087509A

United States Patent [19][11] **Patent Number:** 6,087,509

Claussner et al.

[45] **Date of Patent:** Jul. 11, 2000[54] **1-IMIDAZOLIDINYL-PHENYLS**5,434,176 7/1995 Claussner et al. 514/391
5,656,651 8/1997 Sovak et al. 514/396[75] **Inventors:** Andre Claussner, Villemomble;
Francois Goubet, Paris; Jean-George
Teutsch, Pantin, all of France**FOREIGN PATENT DOCUMENTS**494819 7/1992 European Pat. Off. .
578516 1/1994 European Pat. Off. .
0704448 4/1996 European Pat. Off. .
2693461 1/1994 France .
52-000270 1/1977 Japan 548/320.1
95187947 7/1995 WIPO .[73] **Assignee:** Hoechst Marion Roussel, France[21] **Appl. No.:** 09/077,223[22] **PCT Filed:** Nov. 21, 1996[86] **PCT No.:** PCT/FR96/01846

§ 371 Date: Jun. 23, 1998

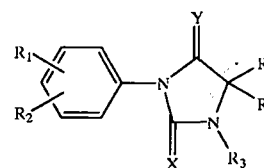
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[87] **PCT Pub. No.:** WO97/19064

PCT Pub. Date: May 29, 1997

[30] **Foreign Application Priority Data**

Nov. 22, 1995 [FR] France 9513836

[51] **Int. Cl.⁷** C07D 233/76; C07D 233/72;
C07D 233/78; C07D 235/02; C07D 405/12;
A61K 31/4166; A61N 5/28; A61N 17/10[52] **U.S. Cl.** 548/307.1; 514/396; 514/397;
514/398; 514/399; 514/400; 548/219.1;
548/320; 548/320.5; 548/321.1[58] **Field of Search** 548/320.1, 321.1,
548/320.5, 317.1, 300.7; 514/398, 399,
396, 397[56] **References Cited****U.S. PATENT DOCUMENTS**4,407,814 10/1983 Bernauer et al. 548/321.1 X
4,473,393 9/1984 Nagpal 548/320.1 X
4,944,791 7/1990 Schroder et al. 548/321.1 X
4,958,028 9/1990 Prisbylla 548/321.1
5,166,358 11/1992 Seuron et al. 548/321.1
5,411,981 5/1995 Gaillard-Kelly et al. 514/386*Primary Examiner*—Floyd D. Higel
Attorney, Agent, or Firm—Bierman, Muserlian and Lucas[57] **ABSTRACT**A particular subject of the invention is the products of
formula (I):

(I)

in which:

R₁ and R₂ represent in particular cyano and
trifluoromethyl,R₃ represents in particular alkyl, alkenyl or alkynyl,
optionally substituted by one or more halogen, cyano or
hydroxyl radicals,R₄ and R₅ either represent in particular methyl optionally
substituted by fluorine,

or form in particular a cyclohexyl radical,

X and Y represent in particular oxygen,

as well as their salts and isomers.

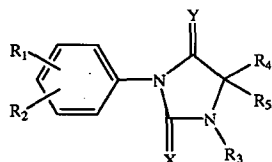
3 Claims, No Drawings

1-IMIDAZOLIDINYL-PHENYLS

This application is a 371 PCT/FR96/01846 filed Nov. 21, 1996.

The present invention relates to new fluorinated or hydroxylated phenylimidazolidines, their preparation process, the new intermediates obtained, their use as medicaments, their new use and the pharmaceutical compositions containing them.

A subject of the present invention is the products of formula (I):



in which:

R_1 and R_2 , identical or different, are chosen from cyano, nitro, trifluoromethyl radicals and halogen atoms,

R_3 represents a linear or branched aryl, arylalkyl, alkyl, alkenyl or alkynyl radical containing at most 10 carbon atoms and optionally substituted by one or more radicals chosen from halogen atoms and cyano, hydroxyl, alkoxy, carboxy, acyl and acyloxy radicals, in which, if appropriate, the alkyl, alkoxy and acyl radicals are linear or branched, containing at most 10 carbon atoms, the carboxy radical is free, salified, esterified or amidified and the hydroxy radical is free, esterified, etherified or protected,

R_4 and R_5 identical or different, represent a linear or branched alkyl radical containing at most 4 carbon atoms and optionally substituted by a halogen atom, or form with the carbon atom to which they are linked a cyclic radical constituted by 3 to 7 members and optionally containing one or more identical or different heteroatoms, chosen from oxygen, sulphur or nitrogen atoms,

X and Y, identical or different, represent an oxygen or sulphur atom,

said products of formula (I) being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with mineral and organic acids or with mineral and organic bases of said products of formula (I).

In the products of formula (I) and in what follows:

the term halogen designates fluorine, chlorine, bromine or iodine atoms,

The fluorine, chlorine or bromine atoms are preferred.

The term linear or branched alkyl radical designates the following radicals: methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl, tert-butyl, pentyl, isopentyl, hexyl, isohexyl and also heptyl, octyl, nonyl and decyl, as well as their linear or branched position isomers,

Alkyl radicals having at most 6 carbon atoms are preferred and in particular the methyl, ethyl, propyl, isopropyl, n-butyl, n-pentyl and n-hexyl radicals.

The term linear or branched alkenyl radical designates the vinyl, allyl, 1-propenyl, butenyl, 1-butenyl, pentenyl or hexenyl radicals as well as their linear or branched position isomers.

Among the alkenyl radicals, the vinyl, allyl, n-butenyl or isobutenyl values are preferred.

The term alkynyl designates a linear or branched radical having at most 12 carbon atoms such as for example ethynyl, propargyl, butynyl, pentynyl or hexynyl.

Among the alkynyl radicals, those with 4 carbon atoms are preferred and in particular the propargyl radical.

The term linear or branched alkoxy radical designates the methoxy, ethoxy, propoxy, isopropoxy, Linear, secondary or tertiary butoxy, pentoxy or hexoxy radicals as well as their linear or branched position isomers.

The term cyclic radical constituted by 3 to 7 members and optionally containing one or more identical or different heteroatoms, chosen from oxygen, sulphur or nitrogen atoms, designates on the one hand a cycloalkyl radical which itself designates in particular the cyclobutyl, cyclopentyl and cyclohexyl radicals and on the other hand a carbocyclic radical interrupted by one or more heteroatoms chosen from oxygen, nitrogen or sulphur atoms such as quite particularly the saturated monocyclic heterocyclic radicals such as for example the following radicals: oxetanyl, oxolanyl, dioxanyl, dithiolane, thiooxolane, thiooxane, pyrrolidinyl, piperidyl, piperazinyl, morpholinyl, azetidine, oxetane and thietane.

The term acyl radical preferably designates the formyl, acetyl, propionyl, butyryl and benzoyl radicals, but also the valeryl, hexanoyl, acryloyl, crotonoyl and carbamoyl radicals.

The term acyloxy radical designates the radicals in which the acyl radicals have the meaning indicated above and for example the acetoxyl or propionyloxy radicals.

The term aryl designates the carbocyclic aryl radicals such as phenyl or naphthyl and the heterocyclic monocyclic aryl radicals with 5 or 6 members or constituted by condensed rings, containing one or more heteroatoms preferably chosen from oxygen, sulphur and nitrogen. Among the heterocyclic aryl radicals with 5 members the following radicals can be mentioned: furyl, thienyl, pyrrolyl, thiazolyl, oxazolyl, imidazolyl, thiadiazolyl, pyrazolyl, isoxazolyl, tetrazolyl.

Among the heterocyclic aryl radicals with 6 members, the pyridyl, pyrimidinyl, pyridazinyl, pyrazinyl radicals can be mentioned.

Among the condensed aryl radicals, the indolyl, benzofuranyl, benzothieryl, quinolinyl radicals can be mentioned.

The phenyl, tetrazolyl and pyridyl radicals are preferred.

The term arylalkyl designates the radicals resulting from the combination of the alkyl radicals and the aryl radicals mentioned above.

The benzyl, phenylethyl, pyridylmethyl, pyridylethyl or tetrazolylmethyl radicals are preferred.

As particular examples of alkyl radicals substituted by one or more halogens, the monofluoro-, chloro- or bromomethyl, difluoro-, dichloro- or dibromo-methyl and trifluoromethyl radicals can be mentioned.

The carboxy radical or radicals of the products of formula (I) can be free, salified, esterified or amidified by the various groups known to a man skilled in the art.

There can be mentioned, for example:

the carboxy radicals salified by mineral bases such as, for example, an equivalent of sodium, potassium, lithium, calcium, magnesium or ammonium or organic bases such as, for example, methylamine, propylamine, trimethylamine, diethylamine, triethylamine, N,N-

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dimethylethanolamine, tris (hydroxymethyl) amino methane, ethanolamine, pyridine, picoline, dicyclohexylamine, morpholine, benzylamine, procaine, lysine, arginine, histidine, N-methylglucamine.

The sodium or potassium salts are preferred.

the carboxy radicals esterified by alkyl radicals in order to form alkoxy carbonyl groups such as, for example, methoxycarbonyl, ethoxycarbonyl, propoxycarbonyl, butoxy-, isobutoxy- and tert-butoxy-carbonyl or benzyloxycarbonyl, these alkyl radicals being able to be substituted by radicals chosen for example from halogen atoms, hydroxyl, alkoxy, acyl, acyloxy, alkylthio, amino or aryl radicals such as, for example, in the chloromethyl, hydroxypropyl, methoxymethyl, propionyloxymethyl, methylthiomethyl, dimethylaminoethyl, benzyl or phenethyl groups.

The radicals formed with the easily cleavable ester remainders can also be mentioned, such as the methoxymethyl, ethoxymethyl radicals; the acyloxyalkyl radicals such as pivaloyloxymethyl, pivaloyloxyethyl, acetoxyethyl or acetoxyethyl; the alkyloxycarbonyloxy alkyl radicals such as the methoxycarbonyloxy methyl or ethyl radicals, the isopropylloxycarbonyloxy methyl or ethyl radicals.

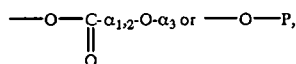
A list of such ester radicals can be found for example in the European Patent EP 0,034,536.

By amidified carboxy is meant the groups of $-\text{CON}(\text{R}_6)$ (R_7) type in which the R_6 and R_7 radical identical or different represent a hydrogen atom or an alkyl radical having 1 to 4 carbon atoms such as the following radicals: methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl or tert-butyl.

Among the $-\text{CON}(\text{R}_6)(\text{R}_7)$ groups defined above, those in which the $-\text{N}(\text{R}_6)(\text{R}_7)$ represents the amino, mono- or dimethylamino radical are preferred.

The $\text{N}(\text{R}_6)(\text{R}_7)$ radical can also represent a heterocycle which may or may not contain an additional heteroatom. The pyrrolyl, imidazolyl, indolyl, piperidino, morpholino, piperazinyloxy radicals can be mentioned. The piperidino or morpholino radicals are preferred.

By esterified, etherified or protected hydroxyl radical is meant the



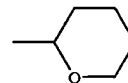
radicals respectively, formed from an $-\text{OH}$ hydroxyl radical, according to the usual methods known to a man skilled in the art and in which P represents a protective group, α_1 , α_2 and α_3 represent in particular an alkyl, alkenyl, alkynyl, aryl or arylalkyl radical, having at most 12 carbon atoms and optionally substituted as is defined above in particular for R_3 .

Examples of the protective group P, as well as the formation of the protected hydroxyl radical, are given in particular in the usual book of a man skilled in the art: Protective Groups in Organic Synthesis, Theodora W. Greene, Harvard University, printed in 1981 by Wiley-Interscience Publishers, John Wiley & Sons.

The protective group of the hydroxyl radical which can be represented by P, can be chosen from the list below: for example formyl, acetyl, chloroacetyl, bromoacetyl, dichloroacetyl, trichloroacetyl, trifluoroacetyl, methoxyacetyl, phenoxyacetyl, benzoyl, benzoylformyl, p-nitrobenzoyl. The following groups can also be men-

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tioned: ethoxycarbonyl, methoxycarbonyl, propoxycarbonyl, $\beta\beta\beta$ -trichloroethoxy-carbonyl, benzyloxycarbonyl, tertbutoxycarbonyl, 1-cyclopropylethoxycarbonyl, tetrahydropyranyl, tetrahydrothiopyranyl, methoxytetrahydropyranyl, trityl, benzyl, 4-methoxybenzyl, benzhydryl, trichloroethyl, 1-methyl 1-methoxyethyl, phthaloyl, propionyl, butyryl, isobutyryl, valeryl, isovaleryl, oxalyl, succinyl and pivaloyl, phenylacetyl, phenylpropionyl, mesyl, chlorobenzoyl, para-nitrobenzoyl, para-tert-butylbenzoyl, caprylyl, acryloyl, methylcarbamoyl, phenylcarbamoyl, naphthylcarbamoyl. P can in particular represent the radical



or also a silicon derivative such as trimethylsilyl.

The addition salts with mineral or organic acids of the products of formula (I) can be, for example, the salts formed with the following acids: hydrochloric, hydrobromic, hydroiodic, nitric, sulphuric, phosphoric, propionic, acetic, formic, benzoic, maleic, fumaric, succinic, tartaric, citric, oxalic, glyoxylic, aspartic, ascorbic, alkyldisulphonic such as for example methanesulphonic, ethanesulphonic, propanesulphonic, alkyldisulphonic such as for example methanedisulphonic, alpha, beta-ethanedisulphonic, arylmonosulphonic such as benzenesulphonic and arylsulphonic.

There can be mentioned more particularly the salts formed with the hydrochloric or methanesulphonic acids for example.

It can be remembered that stereoisomerism can be defined as the isomerism of compounds having the same developed formulae, but the various groups of which are arranged differently in space, such as in particular in the boat and chair shapes of cyclohexane and mono-substituted cyclohexanes whose substituent can be in axial or equatorial position, and the various possible rotational conformations of ethane derivatives. However, another type of stereoisomerism exists, due to the different spatial arrangements of fixed substituents, either on double bonds, or on rings, which is often called geometrical isomerism or cis-trans isomerism. The term stereoisomeric is used in the present Application in its broadest sense and therefore relates to all of the compounds indicated above.

In the products of formula (I) and in what follows, it can be noted that:

the hydrogen atoms which are contained by the optionally substituted alkyl or alkenyl radicals which can be represented by R_3 can be deuterium atoms,

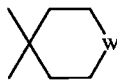
the fluorine atoms which can be represented by halogen atoms can be an ^{18}F atom which is useful for medical imagery.

Thus a subject of the present invention is the products of formula (I) as defined above, in which: R_1 and R_2 both represent a chlorine atom, or being identical or different are chosen from the cyano, nitro and trifluoromethyl radical,

R_3 represents a linear or branched phenyl, pyridyl, phenylalkyl, pyridylalkyl, alkyl, alkenyl or alkynyl radical containing at most 4 carbon atoms and optionally substituted by one or more radicals chosen from halogen atoms and cyano, hydroxyl, alkoxy, acyl and acyloxy radicals, in which if appropriate, the acyl and alkoxy radicals are linear or branched, containing at most 6 carbon atoms and the hydroxyl radical is free, esterified or protected,

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R_4 and R_5 , identical or different, represent a methyl radical optionally substituted by a halogen atom, or form with the carbon atom to which they are linked a cyclobutyl, cyclopentyl, cyclohexyl, dioxane radical, or a



radical in which W represents an oxygen or sulphur atom or the $-NH$ radical,

X and Y, identical or different, represent an oxygen or sulphur atom,

said products of formula (I) being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with mineral and organic acids or with mineral and organic bases of said products of formula (I).

The



radical represents in particular the piperidyl or tetrahydropyran radical.

A particular subject of the present invention is the products of formula (I) as defined above, in which:

R_1 and R_2 represent a cyano radical and a trifluoromethyl radical,

R_3 represents a linear or branched alkyl, alkenyl or alkynyl radical, containing at most 4 carbon atoms and optionally substituted by one or more radicals chosen from halogen atoms, the cyano radical and the free, esterified or protected hydroxyl radical,

R_4 and R_5 , identical or different, represent a methyl radical optionally substituted by a fluorine atom, or form with the carbon atom to which they are attached a cyclohexyl radical

X and Y represent an oxygen atom, said products of formula (I) being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with mineral and organic acids or with mineral and organic bases of said products of formula (I).

Among the preferred products of the invention, there can be mentioned more particularly the products of formula (I) as defined above the names of which follow:

4-(4,4-bis(fluoromethyl)-2,5-dioxo-3-(2-fluoroethyl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzotrile,

4-(2,5-dioxo-4,4-bis(fluoromethyl)-3-ethyl-1-imidazolidinyl)-2-(trifluoromethyl)-benzotrile,

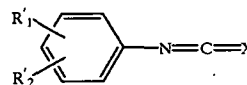
4-(4,4-bis(fluoromethyl)-2,5-dioxo-3-(4-hydroxy-2-butyn-1-yl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzotrile,

4-(3-(4-hydroxy-2-butyn-1-yl)-4,4-dimethyl-2,5-dione-1-imidazolidinyl)-2-(trifluoromethyl)-benzotrile,

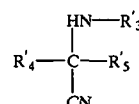
4-[2,4-dioxo-1-(4-hydroxybutyl)-1,3-diazaspiro[4.5]decan-3-yl]-2-(trifluoromethyl)-benzotrile, said products of formula (I) being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with mineral and organic acids or with mineral and organic bases of said products of formula (I).

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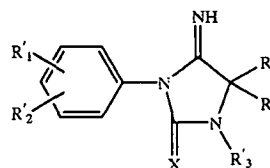
Also a subject of the present invention is a preparation process for the products of formula (I), as defined above, characterized in that, in the presence of a tertiary base, a product of formula (II):



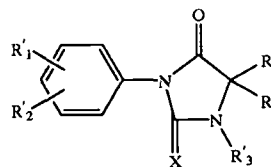
in which R'_1 and R'_2 have the meanings indicated above, for R_1 and R_2 respectively, in which the optional reactive functions are optionally protected and X has the meaning indicated above, is reacted with a product of formula (III):



in which R'_3 , R'_4 and R'_5 have the meanings indicated above, for R_3 , R_4 and R_5 respectively in which the optional reactive functions are optionally protected, in order to obtain a product of formula (IV):



in which X, R'_1 , R'_2 , R'_3 , R'_4 and R'_5 have the meanings indicated above, which is converted into a product of formula (V):



in which X, R'_1 , R'_2 , R'_3 , R'_4 and R'_5 have the meanings indicated above,

which products of formulae (IV) and (V), if necessary or if desired, in order to obtain products of formula (I) as defined above, can be subjected to any one or more of the following reactions, in any order:

- if appropriate conversion of the $>C=S$ group which can be represented by $>C=X$ in the $>C=O$ group,
- the action on the products of formula (V) in which R'_3 represents a hydrogen atom, of a reagent of formula $Hal-R''_3$ in which R''_3 has the values indicated above for R_3 , with the exception of the hydrogen value, and in which the optional reactive functions are optionally protected, and Hal represents a

- halogen atom, in order to obtain the corresponding products, in which R'_3 is replaced by R''_3 ,
- c) release of the OH radical which can be carried by one or both of R'_4 and R'_5 and/or R'_3 ,
 - d) esterification or conversion of the OH radical into a halogen radical,
 - e) elimination reaction of the optional protective groups,
 - f) if appropriate the action of an esterification, amidification or salification agent,
 - g) resolution reaction of the racemic forms, said products of formula (I) thus obtained being in all the possible racemic, enantiomeric and diastereoisomeric forms.

The action of the products of formula (II) with the products of formula (III) in order to obtain the products of formula (IV) can be carried out in an organic solvent such as tetrahydrofuran or dichloroethane but ethyl ether or isopropyl ether can also be used.

The operation is carried out in the presence of a tertiary base such as triethylamine or also pyridine or methylethylpyridine.

The optional reactive functions which are optionally protected can be in particular the hydroxy or amino functions. The usual protective groups are used to protect these functions. The following protective groups of the amino radical can for example be mentioned: tert-butyl, tertamyl, trichloroacetyl, chloroacetyl, benzhydryl, trityl, formyl, benzyloxycarbonyl, tertbutyloxycarbonyl.

As the protective group of the hydroxy radical there can be mentioned for example the tetrahydropyranyl, trimethylsilyl, triphenylmethyl or tert-butyl dimethylsilyl radicals.

Of course the above list is not limitative and other protective groups, for example known in the chemistry of the peptides, can be used. A list of such protective groups is found for example in the French Patent BF 2,499,995 whose content is incorporated here by reference.

In the product of formula (III), R_4 and R_5 can form a cyclic radical with the carbon atom that carries them, such as in particular a cyclohexyl radical.

Also, in the product of formula (III), one or both of R_4 and R_5 may or may not carry a hydroxyl radical which can be protected in particular as an -O-tetrahydropyranyl (OTHP).

The reaction of the product of formula (II) as defined above with a product of formula (III) as defined above in order to produce the corresponding product of formula (IV), can then be carried out in particular in the presence of methylene chloride at a temperature of about -30°C .

The optional elimination reactions of the protective groups are carried out according to the usual methods known to a man skilled in the art or, for example, as indicated in the Patent BF 2,499,995. The preferred method of elimination is acid hydrolysis using acids chosen from hydrochloric, benzene sulphonic or paratoluene sulphonic, formic or trifluoroacetic acids. Hydrochloric acid is preferred.

The optional hydrolysis reaction of the $>\text{C}=\text{NH}$ group into the carbonyl group in order to convert in particular the product of formula (IV) into the product of formula (V) is preferably carried out using an acid such as aqueous hydrochloric acid, for example under reflux.

The action on the products of formula (V) of the reagent of formula $\text{Hal}-\text{R}''_3$ is carried out in the presence of a strong base such as sodium or potassium hydride. The operation can be carried out by phase transfer reaction in the presence of quaternary ammonium salts such as tetrabutyl ammonium dihydrogen phosphate salts.

The conversion of the OH radical into the halogen radical can be carried out under the usual conditions known to a man skilled in the art such as in particular in a solvent such as for example tetrahydrofuran and by the action of a halogenated derivative such as in particular, when the halogen atom is a fluorine atom, diethylaminosulphide trifluoride (DAST).

Triflic anhydride can also be reacted beforehand in order to obtain the corresponding triflate which is then exchanged with the corresponding fluoride as described hereafter in the examples and in particular by the action of tetrabutylammonium fluoride.

When the halogen atom is a bromine, chlorine or iodine atom, the action can be carried out according to the usual conditions known to a man skilled in the art such as in particular by the action, in the presence of triphenylphosphine, of the corresponding halogenating agent such as for example carbon tetrabromide, carbon tetrachloride or also iodine.

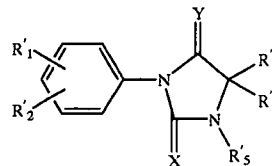
The optional esterification of the products of formula (IV), (V) or (I) as defined above, which contain one or more free OH radicals is carried out under standard conditions. For example an acid or a functional derivative can be used, for example an anhydride such as acetic anhydride in the presence of a base such as pyridine.

The optional esterification or salification of the products of formula (IV), (V) or (I) as defined above, which contain one or more COOH groups, is carried out under standard conditions known to a man skilled in the art.

The optional amidification of the products of formula (IV), (V) or (I) as defined above, which contain a COOH radical, is carried out under standard conditions. A primary or secondary amine can be used on a functional derivative of the acid, for example a symmetrical or mixed anhydride.

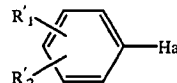
Also a subject of the present invention is a preparation process for the products of formula (I):

(I)

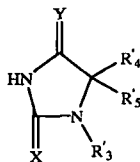


in which X, Y, R'_1 , R'_2 , R'_3 , R'_4 and R'_5 are as defined above, process characterized in that a product of formula

(VII)



in which R'_1 and R'_2 have the previous meanings and Hal represents a halogen atom, is reacted with a product of formula (VI):



in which X, Y, R₃, R₄ and R₅ have the meanings indicated above, the reaction being carried out in the presence of a catalyst and optionally of a solvent.

With regard to the products of formula (VI), the term Hal preferably designates the chlorine atom, but can also represent a bromine or iodine atom.

The reaction conditions of such a process are in particular those described in EP 0,494,819.

The products which are a subject of the present invention possess useful pharmacological properties, in particular they fix themselves on the androgen receptor and they have an anti-androgenic activity.

Tests given in the experimental part illustrate these properties.

These properties make the products of formula (I) as defined above of the present invention of use as medicaments mainly for:

the treatment of adenomas and neoplasias of the prostate as well as benign hypertrophy of the prostate, on its own or combined with analogues of LHRH. They can also be used in the treatment of benign or malignant tumours possessing androgen receptors and more particularly cancers of the breast, the skin, the ovaries, the bladder, the lymphatic system, the kidneys and the liver,

the treatment of cutaneous affections such as acne, hyperseborrhea, alopecia or hirsutism. These products can therefore be used in dermatology on their own or combined with antibiotics such as derivatives of azelaic and fusidic acids, erythromycin, as well as derivatives of retinoic acid or an inhibitor of Salpha-reductase such as (Salpha,17beta)-1,1-dimethylethyl 3-oxo 4-aza-androst-1-ene 17-carboxamide (or Finasteride, Merck 11th Ed.) for the treatment of acne, alopecia or hirsutism. They can also be combined with a product stimulating hair growth such as Minoxidil for the treatment of alopecia.

The products of formula (I), as defined above, in radioactive form (tritium, carbon 14, iodine 125 or fluorine 18) can also be used as specific labels for the androgen receptors. They can also be used in diagnostics for medical imagery.

The products of formula (I) as defined above can also be used in the veterinary domain for the treatment of behavioural disorders such as aggressiveness, androgen-dependent affections, such as circum analum in dogs and tumours having androgen receptors. They can also be used to bring about a chemical castration in animals.

Therefore a subject of the invention is the use, as medicaments, of the products of formula (I) as defined above, said products of formula (I) being in all the possible racemic or optically-active isomer forms, as well as the addition salts with pharmaceutically acceptable mineral or organic acids or mineral and organic bases of said products of formula (I).

A particular subject of the invention is the use, as medicaments of the products of formula (I) as defined above, in which:

(VI)

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R₁ and R₂ represent a cyano radical and a trifluoromethyl radical,

R₃ represents a linear or branched alkyl, alkenyl or alkynyl radical, containing at most 4 carbon atoms and optionally substituted by one or more radicals chosen from halogen atoms, the cyano radical and the free, salified or protected hydroxyl radical,

R₄ and R₅ are either identical or different and represent a methyl radical optionally substituted by a fluorine atom, or form with the carbon atom to which they are attached a cyclohexyl radical,

X and Y represent an oxygen atom, said products of formula (I) being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with pharmaceutically acceptable mineral and organic acids or mineral and organic bases of said products of formula (I).

Also a subject of the invention is the use, as medicaments, of the following products:

4-(4,4-bis(fluoromethyl)-2,5-dioxo-3-(2-fluoroethyl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzotrile,

4-(2,5-dioxo-4,4-bis(fluoromethyl)-3-ethyl-1-imidazolidinyl)-2-(trifluoromethyl)-benzotrile,

4-(4,4-bis(fluoromethyl)-2,5-dioxo-3-(4-hydroxy-2-butyn-1-yl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzotrile,

4-(3-(4-hydroxy-2-butyn-1-yl)-4,4-dimethyl-2,5-dioxo-1-imidazolidinyl)-2-(trifluoromethyl)-benzotrile,

4-[2,4-dioxo-1-(4-hydroxybutyl)-1,3-diazaspiro[4.5]decan-3-yl]-2-(trifluoromethyl)-benzotrile, said products of formula (I) being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with pharmaceutically acceptable mineral and organic acids or mineral and organic bases of said products of formula (I).

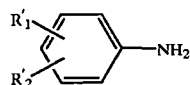
The products can be administered by parenteral, buccal, perlingual, rectal or topical route.

Also a subject of the invention is the pharmaceutical compositions, characterized in that they contain, as active ingredient, at least one of the medicaments of formula (I), as defined above.

These compositions can be presented in the form of injectable solutions or suspensions, tablets, coated tablets, capsules, syrups, suppositories, creams, ointments and lotions. These pharmaceutical forms are prepared according to the usual methods. The active ingredient can be incorporated with excipients usually employed in these compositions, such as aqueous or non-aqueous vehicles, talc, gum arabic, lactose, starch, magnesium stearate, cocoa butter, fatty substances of animal or vegetable origin, paraffin derivatives, glycols, various wetting, dispersing or emulsifying agents, preservatives.

The usual dose, variable according to the patient treated and the affection in question, can be, for example, from 10 mg to 500 mg per day in man, by oral route.

The products of formula (II) used at the start of the invention can be obtained by the action of phosgene when X represents an oxygen atom or thiophosgene when X represents a sulphur atom on the corresponding amine of formula (A):



in which R_1 and R_2 have the meanings indicated above.

A product of this type is in particular described in the French Patent BF 2,329,276.

Amines of formula (A) are described in the European Patent EP 0,002,892 or the French Patent BF 2,142,804. The products of formula (III) are known or can be prepared from the corresponding cyanohydrin according to the process described in the publications: J. Am. Chem. Soc. (1953), 75, 4841, BEIL I 4 526 or J. Org. Chem. 27 2901 (1962).

The products of formula (III) in which R_3 is different from a hydrogen atom can be obtained by the action of a product of formula R_3 Hal on 2-cyano 2-amino propane under the conditions stated above for the action of R_3 Hal on the products of formula (V). An example of this type of preparation is described in the reference:

Jilek et al. Collect. Czech. Chem. Comm. 54(8) 2248 (1989).

The products of formulae (VII) and (VI), used at the start of the process indicated above, for obtaining products of formula (I), as defined above, are known and commercially available or can be prepared according to methods known to a man skilled in the art.

The preparation of products of formula (VI) is described in particular in the following publications:

Zhur. Prékklad. Khim. 28, 969-75 (1955) (CA 50, 4881a, 1956)

Tetrahedron 43, 1753 (1987)

J. Org. Chem. 52, 2407 (1987)

Zh. Org. Khim. 21, 2006 (1985)

J. Fluor. Chem. 17, 345 (1981) or in the:

German Patent DRP 637,318 (1935)

European Patent EP 0,130,875

Japanese Patent JP 81,121,524.

The products of formula (VI) which are hydantoin derivatives are widely used and mentioned in the literature such as for example in the following articles:

J. Pharm. Pharmacol., 67, Vol. 19(4), p. 209-16 (1967)

Khim. Farm. Zh., 67, Vol. 1 (5) p. 51-2

German Patent 2,217,914

European Patent 0,091,596

J. Chem. Soc. Perkin. Trans. 1, p. 219-21 (1974).

Also a subject of the invention is, as new industrial products and in particular as new industrial products which can be used as intermediate products for the preparation of the products of formula (I) as defined above, the products of formulae (IV) and (V) as defined above and notably the products of formula (V) in which R_4 and R_5 represent an alkyl radical substituted by a free, esterified, etherified or protected hydroxyl radical.

Also a subject of the present invention is the use of the products of formula (I) as defined above, for the preparation of pharmaceutical compositions intended for the treatment of adenomas and neoplasias of the prostate as well as of benign hypertrophy of the prostate, on their own or combined with analogues of LHRH, for the treatment of cutaneous affections such as acne, hyperseborrhea, alopecia or hirsutism or in diagnostics for medical imagery.

The following examples illustrate the invention without however limiting it.

Preparation 1

2-(trifluoromethyl) benzonitrile 4-isocyanate.

10 g of 4-cyano 3-(trifluoromethyl) aniline (described in the European Patent EP 0,002,892) in solution in 30 ml of ethyl acetate is added over 20 minutes to 33.6 ml of a toluenic solution of phosgene at 1.93 M/l cooled down to 0/5° C. Agitation is carried out for 30 minutes at this temperature then the reaction medium is left to rise to 25° C. It is heated until distillation is achieved, compensating for the volume distilled off with toluene until the distillation temperature reaches 110° C. Reflux is maintained until the release of hydrochloric acid has stopped (i.e. 4 hours 30 minutes). The medium is returned to ambient temperature, the insoluble part is separated out over sodium sulphate under an inert atmosphere, followed by rinsing three times with 10 ml of toluene and evaporating to dryness under reduced pressure. After heating at 60° C. for one hour, the temperature is returned to ambient under inert atmosphere and 11.6 g of expected product is obtained.

ANALYSES:

Infra-red (cm ⁻¹)	
-N=C=O	2268
-CN	2233

EXAMPLE 1

4-(4,4-bis(fluoromethyl)-2,5-dioxo-3-(2-fluoroethyl)-1-imidazolidinyl)-2-(trifluoromethyl) benzonitrile

STAGE 1: 1,3 bis [(tetrahydro-2H-pyran-2-yl)oxy]-2-propanone

9 g of 2,5-dihydroxy-1,4-dioxane-2,5-dimethanol is introduced into 60 ml of dioxane and the suspension is taken to about 70° C. for 15 minutes then returned to ambient temperature. 20 ml of 3,4-dihydro 2H-pyran and 300 mg of monohydrated paratoluene sulphonic acid are then added and the temperature is maintained at about 40° C. then the whole is left for 16 hours at ambient temperature. The reaction medium is poured into a mixture of 300 ml of saturated sodium bicarbonate solution +10 ml of triethylamine and extraction is carried out 4 times with methylene chloride. The organic phase is washed with salt water and dried. After purification by passage through silica, eluting with cyclohexane-ethyl acetate: 8-2 with 0.5% of triethylamine, 17 g of expected product (pale yellow syrup) is obtained.

ANALYSES:

IR CHCl ₃ (cm ⁻¹)	
Absence OH	
O=C	1736

STAGE 2: 2-amino-3-((tetrahydro-2H-pyran-2-yl) oxy)-2-(((tetrahydro-2H-pyran-2-yl)-oxy)-methyl)-propanenitrile.

5.6 g of the product obtained in Stage 1 above is introduced into 8 ml of ammonium hydroxide, the mixture is taken to about -5° C. and 1.58 g of ammonium chloride and 1.23 g of sodium cyanide are added successively and the reaction medium is left to rise to ambient temperature for about 40 minutes then heated at 40° C. ± 5° C. under agitation

overnight. It is returned to ambient temperature and extraction is carried out 3 times with chloroform, the organic phase is washed with salt water and dried. After purification on silica, eluting with cyclohexane-ethyl acetate: 3-7 with 0.5% of triethylamine, 4.41 g of expected product (pale yellow syrup) is obtained.

ANALYSES:

IR CHCl ₃ (cm ⁻¹)	
—CN	2235
NH ₂	3390-3317

STAGE 3: 4-(5-imino-2-oxo-4,4-bis(((tetrahydro-2H-pyran-2-yl)-oxy)-methyl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

570 mg of the product obtained in Stage 2 above is introduced into 5 ml of isopropyl ether and 0.28 ml of triethylamine and the mixture is taken to -30° C. then a solution of 12.6 ml of 1,2-dichloroethane containing 2.32 g of the product obtained in Preparation 1 is added over one hour. 4 ml of methylene chloride is added then the reaction medium is allowed to rise to ambient temperature, left for about 2 hours and dried. After purification on silica, eluting with methylene chloride-acetone: 9-1, 700 mg of expected product is obtained.

ANALYSES:

IR CHCl ₃ (cm ⁻¹)	
NH	3442-3317
—CN	2235
C=O	1757
C=N	1670
Aromatic	1614-1575-1505

STAGE 4: 4-(4,4-bis(hydroxymethyl)-2,5-dioxo-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

300 mg of the product obtained in Stage 3 above is introduced into 3 ml of methanol and 1.5 ml of 2N hydrochloric acid and the whole is taken to reflux for one hour 30 minutes. It is returned to ambient temperature, poured into 5 ml of bicarbonate, extraction is carried out 4 times with ethyl acetate then the extracts are washed with a saturated sodium chloride solution and dried. 5 ml of methanol is added and purification is carried out on silica eluting with methylene chloride-methanol: 9-1. The residue is taken up in 20 ml of isopropanol under reflux then concentration is carried out and 225 mg of expected product (white crystals) is obtained. M.p.=207-208° C.

IR NUJOL (cm ⁻¹)	
OH/NH	3525-3365-3250
CN	2240
C=O	1778-1738
Aromatic	1618-1578-1506

STAGE 5: 4-[4,4-bis(((tetrahydro-2H-pyran-2-yl)oxy)methyl)-2,5-dioxo-1-imidazolidinyl]-2-(trifluoromethyl)-benzonitrile

331 mg of the product obtained in Stage 4 above, 4 ml of tetrahydrofuran, 1 ml of 3,4-dihydro-2H-pyran and 16 mg of p. toluene sulphonic acid, and H₂O are introduced. After 35 minutes, the reaction medium is poured into 10 ml of sodium bicarbonate+1 ml of triethylamine and extraction is carried

out with 3×10 ml of chloroform, the extracts are washed with a saturated aqueous solution of sodium chloride, dried and evaporated to dryness. After chromatography on silica, eluting with methylene chloride-acetone: 9-1, 500 mg of expected product (friable white foam) is obtained.

ANALYSES:

IR (CHCl ₃) cm ⁻¹	
Absence OH,	
=C—NH	3440
C≡N	2236
>C=O	1791-1736
Aromatics	1615-1576-1505

STAGE 6: 4-(3-(2-fluoroethyl)-4,4-bis(hydroxymethyl)-2,5-dioxo-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

A) Condensation of 2-bromo-1-fluoroethane

530 mg of 50% sodium hydride is introduced and 5 g of the product obtained in Stage 5 above and 30 ml of dimethylsulphoxide dried over silporite are added dropwise over about 40 minutes, and rinsing is carried out with 2 ml of dimethylsulphoxide. 20 minutes after the release of hydrogen has stopped, 1.1 ml of 2-bromo-1-fluoroethane is added in one lot. After reacting for 2 hours 30 minutes, the medium is poured into 250 ml of water containing 1 g of monopotassium phosphate, extraction is carried out 4 times with ether, the organic phase is washed with water then with salt water, dried and evaporated to dryness. Purification is carried out on silica, eluting with methylene chloride-acetone: 92.5-7.5 and in this way 5.31 g of expected product (white friable foam) is obtained.

ANALYSES:

IR (CHCl ₃) cm ⁻¹	
Absence OH and NH	
C≡N	2238
>C=O	1783-1728
Aromatics	1616-1575-1505

b) Hydrolysis of the tetrahydropyranic ethers

550 mg of the product obtained above in a) is taken up in 6 ml of methanol and 2 ml of hydrochloric acid (2N) and the solution obtained is taken to 40° C. for 40 minutes. It is then poured into 15 ml of a saturated solution of sodium bicarbonate, extraction is carried out 4 times with ethyl acetate, the extracts are dried and evaporated to dryness. Purification on silica is carried out eluting with methylene chloride-acetone: 8-2 and in this way 351 mg of expected product (white crystals) is obtained. M.p.=138-139° C.

ANALYSES:

M.A.	% calculated	found
C	48.09	48.1
H	3.49	3.5
F	20.25	19.9
N	11.20	11.5

I.R. Nujol (cm ⁻¹)	
OH/NH	3580-3505
C≡N	2245

dimethylformamide and 0.24 ml of ethyl iodide. In this way 1.1 g of product is obtained which is taken up in 0.2 ml of methanol and 4 ml of hydrochloric acid (2N). In this way 608 mg of expected product (white crystals) is obtained.

M.p.=155-156° C.

ANALYSES:

M.A.	% calculated	found
C	50.42	50.5
H	3.95	3.9
F	15.95	15.6
N	11.76	11.6

I.R. Nujol (cm⁻¹)

Absorption OH/NH approx.	3330
C≡N	2238
C=O	1784-1722
Aromatics	1620-1580-1510

STAGE 2: 4-(2,5-dioxo-4,4-bis(fluoromethyl)-3-ethyl-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

The operation is carried out as in Stage 7 of Example 1, starting with 1 ml of tetrahydrofuran, 0.66 ml of diethylaminotrifluorosulphide and 349 mg of the product obtained in Stage 1 above in 4 ml of tetrahydrofuran. In this way 319 mg of expected product (white crystals) is obtained. M.p.=129-130° C.

ANALYSES:

M.A.	% calculated	found
C	49.89	50.0
H	3.35	3.2
F	26.29	25.95
N	11.63	11.6

I.R. CHCl₃ (cm⁻¹)

C≡N	2238
> = O	1787-1734
Aromatics	1615-1578-1505

EXAMPLE 4

4-(4,4-bis(fluoromethyl) 2,5-dioxo-3-(1-methylethyl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

STAGE 1: 4-(4,4-bis(hydroxymethyl)-2,5-dioxo-3-(1-methylethyl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

The operation is carried out as in a) and b) of Stage 6 of Example 1 starting with 110 mg of 50% sodium hydride, 1 g of the product obtained in Stage 5 of Example 1, 5.5 ml of dimethylsulphoxide and 0.3 ml of isopropyl iodide. 1.1 g of product is obtained which is taken up in 12 ml of methanol and 4 ml of 2N hydrochloric acid. In this way 574 mg of expected product (white crystals) is obtained. M.p.=172-173° C.

ANALYSES:

M.A.	% calculated	found
C	51.75	51.9
H	4.34	4.1
F	15.35	15.2
N	11.32	11.2

I.R. nujol (cm⁻¹)

OH/NH	3390
C≡N	2240
> = O	1782-1720
Aromatics	1618-1575-1508

STAGE 2: 4-(4,4-bis(fluoromethyl)-2,5-dioxo-3-(1-methylethyl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

The operation is carried out as in Stage 7 of Example 1, starting with 1 ml of tetrahydrofuran, 0.66 ml of diethylaminotrifluorosulphide and 316 mg of the product obtained in Stage 1 above in 4 ml of tetrahydrofuran. In this way 298 mg of expected product (white crystals) is obtained. M.p.=153-154° C.

ANALYSES:

M.A.	% calculated	found
C	51.21	51.1
H	3.76	3.6
F	25.01	24.9
N	11.20	11.3

I.R. CHCl₃ (cm⁻¹)

C≡N	2238
C=O	1787-1734
Aromatics	1616-1578-1505

EXAMPLE 5

4-(4,4-bis(fluoromethyl)-2,5-dioxo-3-(2-propynyl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

STAGE 1: 4-(4,4-bis(hydroxymethyl)-2,5-dioxo-3-(2-propynyl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

The operation is carried out as in a) and b) of Stage 6 of Example 1 starting with 420 mg of 50% sodium hydride, 3.86 g of the product obtained in Stage 5 of Example 1, 15 ml of dimethylformamide and 1.30 g of propargyl bromide, in 2 ml of dimethylformamide. 3.872 g of product is obtained which is taken up in 18 ml of methanol and 6 ml of 2N hydrochloric acid. In this way 993 mg of expected product (white crystals) is obtained. M.p.=125-126° C.

ANALYSES:

M.A.	% calculated	found
C	52.32	52.4
H	3.29	3.2
F	15.52	15.5
N	11.44	11.4

-continued

I.R. nujol (cm ⁻¹)	
OH/NH	3495-3360
C≡CH	3308
C≡N	2236
C=O	1784-1730
Aromatics	1666-1576-1506

STAGE 2: 4-[4,4-bis[[[(trifluoromethyl)sulphonyloxy]methyl] 2,5-dioxo-3-(2-propynyl)-1-imidazolidinyl]-2-(trifluoromethyl)-benzonitrile

500 mg of product obtained in Stage 1 above, 8 ml of methylene chloride, 1.2 ml of pyridine and 61 mg of 4-dimethylaminopyridine are introduced, the whole is taken to about -10° C., 1 ml of triflic anhydride is added and the reaction medium is left to react at 0° C. for about 45 minutes. It is then poured into 20 ml of sodium bicarbonate, extraction is carried out 3 times with ethyl acetate, the extracts are washed with salt water and dried. In this way 982 mg of expected product (inert friable foam) is obtained.

STAGE 3: 4-(4,4-bis(fluoromethyl) 2,5-dioxo-3-(2-propynyl) 1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

982 mg of the crude product obtained in Stage 2 above and 8 ml of tetrahydrofuran are introduced and a 1.1 M solution in tetrahydrofuran of 3 ml of tetrabutylammonium fluoride is added over about 5 minutes. After 30 minutes, the reaction medium is poured into 25 ml of 50% sodium bicarbonate, extraction is carried out 3 times with methylene chloride, the extracts are washed with water and dried. A first purification is carried out on silica eluting with methylene chloride-acetone: 99-1, then a second purification is carried out on silica eluting with cyclohexane-ethyl acetate: 7-3. In this way, after evaporation of the fractions and trituration in ether, 252 mg of expected product (white crystals) is obtained. M.p.=126-127° C.

ANALYSES:

M.A.	% calculated	found
C	51.76	52.0
H	2.71	2.7
F	25.58	25.3
N	11.32	11.1

I.R. CHCl ₃ (cm ⁻¹)	
≡CH	3307
C≡N	2238
C=O	1792-1738
Aromatics	1615-1575-1505

EXAMPLE 6

4-(4,4-bis(fluoromethyl)-2,5-dioxo-3-(4-hydroxy-2-butyn-1-yl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

STAGE 1: 4-[3-[4-(acetyloxy)-2-butyn-1-yl]-4,4-bis[[[(tetrahydro-2H-pyran-2-yl)oxy]methyl]-2,5-dioxo-1-imidazolidinyl]-2-(trifluoromethyl)-benzonitrile

416 mg of 50% sodium hydride and dropwise 4 g of the product obtained in Stage 5 of Example 1 and 15 ml of dimethylformamide are introduced and the resultant mixture is rinsed with 1 ml of dimethylformamide. 10 minutes after the end of the release of hydrogen, 3.2 g of 4-bromo-2-

butyne-1-ol acetate prepared as described in J. W. Lown GENE 149, 81 (1994) and 2 ml of dimethylformamide are added, and rinsing is carried out with 0.5 ml of dimethylformamide. After one hour 30 minutes, the reaction medium is poured into 100 ml of water containing 0.5 g of monopotassium phosphate, extraction is carried out 4 times with ether, the organic phase is washed with water and with salt water and dried. Purification is carried out by chromatography on silica eluting with methylene chloride-acetone: 95-5 and 4.47 g of expected product (orange resin) is obtained.

ANALYSES:

I.R. CHCl ₃ (cm ⁻¹)	
C≡N	2236
> = O	1735-1729
Aromatics	1616-1575-1505

STAGE 2: 4-[3-[4-(acetoxo)-2-butyn-1-yl]-4,4-bis[[[(hydroxymethyl)-2,5-dioxo-1-imidazolidinyl]-2-(trifluoromethyl)-benzonitrile

4.4 g of the product obtained in Stage 1 above; 14 ml of tetrahydrofuran, 28 ml of acetic acid and 7 ml of water are introduced and the whole is heated to 60° C.±5° C. for 4 hours. The acetic acid is evaporated off, 100 ml of ethyl acetate is added, followed by washing with bicarbonate and drying. After chromatography on silica eluting with methylene chloride-acetone: 85-15, 1.99 g of expected product (white foam) is obtained.

ANALYSES:

I.R. CHCl ₃ (cm ⁻¹)	
OH	3618-3548
C≡N	2235
> = O	1784-1729
Aromatics	1617-1607-1572-1505

STAGE 3: 4-[3-[4-(acetoxo)-2-butyn-1-yl]-4,4-bis(fluoromethyl)-2,5-dioxo-1-imidazolidinyl]-2-(trifluoromethyl)-benzonitrile

The operation is carried out as in Stages 2 and 3 of Example 5, starting with 439 mg of the product obtained in Stage 2 above, 6 ml of methylene chloride, 1.35 ml of 2,6-lutidine, 50 mg of 4-dimethylaminopyridine and 1 ml of trifluoromethane sulphonic anhydride is added at a temperature of 0 to 5° C. The reaction medium is left to react for one hour at this temperature then poured into a mixture of 50 ml of sodium bicarbonate-ethyl acetate: 1-1, followed by decanting, washing with salt water and drying. Then the product is taken up in 6 ml of tetrahydrofuran, and a 1.1 M solution of 2.3 ml of tetrabutylammonium fluoride in tetrahydrofuran is added. The medium is poured into 60 ml of sodium bicarbonate-ethyl acetate: 1-1, followed by decanting, washing with salt water and drying. After purification by chromatography on silica eluting with methylene chloride-ethyl acetate: 95-5, 177 mg of expected product (resin) is obtained.

ANALYSES:

I.R. CHCl ₃ (cm ⁻¹)	
C≡N	2238
> = O	1792-1757
Aromatics	1615-1575-1505

STAGE 4: 4-(4,4-bis(fluoromethyl)-2,5-dioxo-3-(4-hydroxy-2-butyn-1-yl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

1 g of the product obtained in Stage 3 above, 18 ml of methanol and 4.5 ml of 2N hydrochloric acid are introduced then left for one hour 30 minutes at 50° C. The reaction medium is then returned to ambient temperature, poured into 30 ml of sodium bicarbonate, extraction is carried out with ethyl acetate 3 times, the extracts are washed with salt water and dried. After chromatography on silica eluting with methylene chloride-acetone: 85-15, 780 mg of expected product (white crystals) is obtained. M.p.=131-132° C.

ANALYSES:

I.R. CHCl ₃ (cm ⁻¹)	
OH	3610
C≡N	2238
> = O	1792-1736
Aromatics	1615-1576-1505

EXAMPLE 7

4-(4,4-bis(fluoromethyl)-2,5-dioxo-3-methyl-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

1 ml of tetrahydrofuran is introduced, taken to -30° C., 3.24 ml of diethylaminotrifluorosulphide, then 0.12 g of the product of Example 24 of FR 2715402 are added and the whole is rinsed with 0.5 ml of tetrahydrofuran, then left to rise to ambient temperature and taken to +30° C. After 40 minutes, the reaction medium is poured over 5 g of ice in 20 ml of sodium bicarbonate, extraction is carried out 5 times with methylene chloride, the organic phase is washed with salt water and dried. Purification is carried out by chromatography on silica eluting with methylene chloride-ethyl acetate: 99-1 and in this way 111 mg of expected product (white crystals) is obtained. M.p.=137-138° C.

ANALYSES:

M.A.	% calculated	found
C	48.42	48.2
H	2.90	2.8
F	27.35	26.9
N	12.10	11.9

I.R. CHCl ₃ (cm ⁻¹)	
C≡N	2235
> = O	1790-1735
Aromatics	1617-1580-1505

EXAMPLE 8

4-(3-(4-hydroxy-2-butyn-1-yl)-4,4-dimethyl-2,5-dione-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

- a) Condensation of the 4-tertbutyldimethylsiloxy-2-butyn chain

103 mg of 50% sodium hydride is introduced, 570 mg of the product of Example 8 of EP 0494819, 3.5 ml of dimethylformamide are added and the whole is rinsed with 0.5 ml of dimethylformamide. 20 minutes after the release of hydrogen has stopped, 0.5 g of 1-bromo-4-tertbutyldimethylsiloxy-2-butyn prepared as indicated in J. W. Lown et al. GENE 149, 81 (1994) is added and the reaction medium is taken to 40° C. After 50 minutes, it is poured into 40 ml of water containing about 0.5 g of monopotassium phosphate, extraction is carried out 4 times with ether, the organic phase is washed with water then with salt water and dried. Purification is carried out by chromatography on silica eluting with methylene chloride-acetone: 98-2 and 732 mg of pale syrup is obtained.

- b) Hydrolysis of the silylated ether

The 732 mg of product obtained in a) above is taken up in 7.5 ml of methanol and 1.5 ml of 2N hydrochloric acid. After 40 minutes, the reaction medium is poured into 30 ml of 50% sodium bicarbonate, extraction is carried out 3 times with ethyl acetate, the extracts are washed with salt water and dried. After purification by chromatography on silica eluting with methylene chloride-acetone: 92.5-7.5, 516 mg of expected product (white crystals) is obtained. M.p.=125-126° C.

ANALYSES:

M.A.	% calculated	found
C	55.89	56.0
H	3.86	3.8
F	15.60	15.9
N	11.50	11.7

I.R. CHCl ₃ (cm ⁻¹)	
OH	3608
C≡N	2230
> = O	1783-1726
Aromatics	1616-1575-1505

EXAMPLE 9

4-[2,4-dioxo-1-(4-hydroxybutyl)-1,3-diazaspiro[4.5]decan-3-yl]-2-(trifluoromethyl)-benzonitrile

STAGE 1: 1-amino-cyclohexanecarbonitrile

1.23 g of sodium cyanide, 1.58 g of ammonium chloride and 8 ml of ammonium hydroxide are introduced, the whole is taken to 0° C. and 2.1 g of cyclohexanone is added. The reaction medium is left to return to ambient temperature under agitation for 18 hours, diluted in a small amount of water and decanted. The aqueous phase is extracted with methylene chloride, the organic phases are combined, washed with salt water, dried, filtered and concentrated. In this way 2.38 g of expected product (colourless oil) is obtained.

ANALYSES:

I.R. CHCl ₃ (cm ⁻¹)	
NH ₂	3380-3315
CN	2225

STAGE 2: 4-[2-imino-4-oxo-1,3-diazaspiro[4.5]decan-3-yl]-2-(trifluoromethyl)-benzonitrile

3.75 ml of an 18% 1,2-dichloroethane solution of the product of Preparation 1 is introduced, 10 ml of dry dichlo-

roethane and 0.22 ml of triethylamine are added, the whole is taken to -30°C ., 558 mg of the product obtained in Stage 1 above in 2 ml of dichloroethane is added and the reaction medium is left at ambient temperature for 18 hours. After chromatography on silica eluting with methylene chloride-acetone: 8-2, 1.10 g of expected product (white solid) is obtained.

ANALYSES:

I.R. (cm^{-1})	
OH/NH	3370-3290
CN	2240
C=O	1743
C=N	1678
Aromatics	1615-1606-1572-1542-1508

STAGE 3: 4-(2,4-dioxo-1,3-diazaspiro(4.5)decan-3-yl)-2-(trifluoromethyl)-benzonitrile

1.10 g of the product obtained in Stage 2 above, 3 ml of 6N hydrochloric acid and 8 ml of ethanol are introduced, the mixture is taken to reflux for one hour, cooled down to ambient temperature, neutralized by the addition of sodium hydrogen carbonate, extracted with ethyl acetate. The organic phases are combined, washed with water, dried, filtered and concentrated. After chromatography on silica eluting with methylene chloride-acetone: 9-1, then recrystallization from isopropanol, 470 mg of expected product (white solid) is obtained. M.p.= 187°C .

ANALYSES:

M.A.	% calculated	found
C	57.0	56.8
H	4.15	4.0
N	12.50	12.5
F	16.9	16.8

I.R. CHCl_3 (cm^{-1})	
NH	3450
CN	2235
C=O	1787-1727
Aromatics	1617-1575-1505

STAGE 4: 4-[2,4-dioxo-1-(4-hydroxybutyl)-1,3-diazaspiro[4.5]decan-3-yl]-2-(trifluoromethyl)-benzonitrile

28 mg of 50% sodium hydride and 3 ml of dimethylformamide are introduced, agitation is carried out at ambient temperature for 5 minutes, 170 mg of the product obtained in Stage 3 above is added and the whole is left under agitation for 20 minutes until the release of hydrogen has stopped. 202 mg of O-trimethylsilyl-4-iodo-n-butanol is then added and the reaction medium is left under agitation for 2 hours. After hydrolysis using saturated ammonium chloride, extraction is carried out with ethyl acetate. The organic phases are combined, washed with water, dried, filtered and concentrated. The residue is solubilized in 10 ml of methanol and 0.5 ml of 2N hydrochloric acid is added. After agitation for 5 minutes, the solution is neutralized by the addition of saturated sodium hydrogen carbonate and extraction is carried out with ethyl acetate. The organic phases are combined, dried, filtered and concentrated. After chromatography on silica eluting with methylene chloride-acetone: 9-1, 158 mg of expected product (white solid) is obtained. M.p.= 145°C .

ANALYSES:

M.A.	% calculated	found
C	58.8	58.8
H	5.4	5.2
F	13.9	13.6
N	10.3	10.1

I.R. CHCl_3 (cm^{-1})	
OH	3626
CN	2236
C=O	1772-1719
Aromatics	1615-1575-1505

EXAMPLE 10

Tablets were prepared having the following composition:

Product of Example 6 . . . 100 mg

Excipient s.q. for a tablet completed at . . . 300 mg (Detail of the excipient: lactose, starch, talc, magnesium stearate).

PHARMACOLOGICAL STUDY OF THE PRODUCTS OF THE INVENTION

1) Study of the Affinity of the Products of the Invention for the Androgen Receptor

Male Sprague Dawley EOPS rats weighing 180-200 g, castrated 24 hours previously, are sacrificed, the prostates are removed, weighed and homogenized at 0°C using a Potter glass flask, in a buffered solution (10 mM Tris, 0.25M saccharose, 0.1 mM PMSF (phenylmethanesulphonyl fluoride), 20 mM sodium molybdate, HCl pH 7.4; to which 2 mM of DTT (DL dithiothreitol) is added extemporaneously), at the rate of 1 g of tissue per 8 ml of buffer.

The homogenate is then ultracentrifuged at 0°C for 30 minutes at 209,000 g. Aliquots of the supernatant obtained (=cytosol), are incubated for 30 minutes and 24 hours at 0°C ., with a constant concentration (T) of tritiated testosterone and in the presence of increasing concentrations (0 to $2500 \times 10^{-9}\text{M}$), either of unlabelled testosterone, or of the products to be tested. The concentration of bound tritiated testosterone (B) is then measured in each incubate by the method of adsorption with carbon dextran.

Calculation of the Relative Bond Affinity (RBA).

The following 2 curves are drawn: the percentage of bound tritiated hormone B/T as a function of the logarithm of the concentration of unlabelled reference hormone and B/T as a function of the logarithm of the concentration of unlabelled product tested. The straight line of the equation $I_{50} = (B/T_{\text{max}} + B/T_{\text{min}})/2$ is determined.

B/T_{max} = % of bound tritiated hormone for an incubation of this tritiated hormone at the concentration (T).

B/T_{min} = % of bound tritiated hormone for an incubation of this tritiated hormone at the concentration (T) in the presence of a large excess of unlabelled hormone ($2500 \times 10^{-9}\text{M}$).

The intersections of the straight line I_{50} and the curves allow the evaluation of the concentrations of the unlabelled reference hormone (CH) and of the tested unlabelled product (CX) which inhibit by 50% the binding of the tritiated hormone on the receptor. The relative bond affinity (RBA) of the tested product is determined by the equation $\text{RBA} = 100 (\text{CH})/(\text{CX})$.

The following results are obtained, expressed in RBA.
Reference product (Testosterone): 100

TABLE 1

Product of example	RBA: Incubation 24 hours
1	42
3	43

2) Determination of the Reducing Effect on the Costovertebral Gland of the Hamster

The local activity (topical) of an antiandrogen is determined by the reduction which it brings about in the surface area of the costovertebral gland of the hamster (hereafter C.V.G.), an androgen-dependent organ situated on the flanks of the animal.

The animals are male hamsters weighing about 140 g, 14 weeks old and originating from the Charles River breed (USA), they are subjected to a long photoperiod (16 hours of light, 8 hours of darkness). The animals are treated every day, except for the weekend, for 3 weeks (14 administrations). The product to be tested is applied, by topical route, on the right-hand C.V.G., the left-hand one serving as the control. The surface of the gland has been shaved beforehand. The animals are sacrificed by bleeding the carotid artery 24 hours after the last treatment. The C.V.G.'s are removed, measured and weighed. The local activity of a product is determined by the % of reduction in the surface area of the C.V.G. which it induces in comparison with the 1st day of the experiment and compared to the animals treated with solvent only.

TABLE 2

Product of example	% CVG reduction with 3 µg/jour
1	-32
3	-33
9	-25

3) Determination of the Reducing Effect of the Weight of the Prostate in an Intact Male Rat

The systemic activity of an antiandrogen is determined by the reduction in the weight of the prostate which it brings about in an intact animal.

The animals used are male rats of the Sprague Dawley strain weighing about 200 g, 7 weeks old, originating from the Iffa Credo breed (France). The experiment is carried out over two weeks, except for the weekend.

The product can be administered by oral, sub-cutaneous or percutaneous route.

The solvents used are then: by oral route: 0.5% aqueous solution of methylcellulose under a volume of 5 ml/kg, by sub-cutaneous route: wheatgerm oil in 10% ethanol under a volume of 0.2 ml/kg, and by percutaneous route: ethanol under a volume of 50 µl on previously-shaved skin.

The treatment is carried out from day 0 to day 4 then (after the weekend) from day 7 to day 10. The animals are sacrificed the day after the last treatment by bleeding the carotid artery, the prostates are removed and fixed in demineralized water containing 10% formol for 72 hours. They are then dissected and weighed. The blood is removed in order to determine, by radioimmunological assay, the amount of seric testosterone. The antiandrogen activity of the product is expressed as % reduction in the weight of the prostates and as % variation in the amounts of testosterone compared to the animals treated by the solvent only.

TABLE 3

Product of example	% reduction in the prostate weight with 3 mg/kg P.O.
1	-27
3	-20
8	-35

What is claimed is:

1. A compound selected from the group consisting of 4-(4,4-bis(fluoromethyl)-2,5-dioxo-3-(4-hydroxy-2-butyn-1-yl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzoxonitrile, 4-(3-(4-hydroxy-2-butyn-1-yl)-4,4-dimethyl-2,5-dioxo-1-imidazolidinyl)-2-(trifluoromethyl)-benzoxonitrile, 4-[2,4-dioxo-1-(4-hydroxybutyl)-1,3-diazaspiro[4,5]decan-3-yl]-2-(trifluoromethyl)-benzoxonitrile and the addition salts with non-toxic, pharmaceutically acids.

2. An antiandrogenic composition comprising an antiandrogenically effective amount of a compound of claim 1 and an inert pharmaceutical carrier.

3. A method of inducing antiandrogenic activity in warm-blooded animals comprising administering to warm-blooded animals an antiandrogenically effective amount of a compound of claim 1.

* * * * *

Using the procedure of Example 16, the product of Example 15 was reacted to obtain the expected product melting at 92° C.-93° C.

IR Spectrum CHCl₃:

OH	3626 cm ⁻¹
C≡N	2235 cm ⁻¹
C=O	1775(m), 1784 cm ⁻¹ (F)
aromatics	1616-1578-1505 cm ⁻¹

UV Spectrum (EtOH):

Max.	238 nm	ε = 11,200
Max.	262 nm	ε = 13,900
Inf.	288 nm	

Using the procedure of the above Examples, the appropriate isocyanate or thioisocyanate and 3,3,3-trifluoromethyl-2-trifluoromethyl-2-methylamino-propionitrile prepared according to J. Org. Chem., Vol. 35, p. 1485 (1970) were reacted to obtain the following compounds.

Example 18

4-(4,4-bis(trifluoromethyl)-3-methyl-5-imino-2-thioxo-1-imidazolidiny)-2-trifluoromethyl-benzonitrile

Example 19

4-(4,4-bis(trifluoromethyl)-3-methyl-5-oxo-2-thioxo-1-imidazolidiny)-2-trifluoromethyl-benzonitrile

Example 20

4-(4,4-bis(trifluoromethyl)-3-methyl-5-imino-2-oxo-1-imidazolidiny)-2-trifluoromethyl-benzonitrile

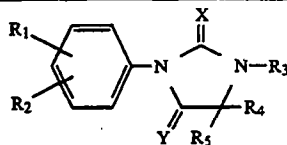
Example 21

4-(4,4-bis(trifluoromethyl)-3-methyl-2,5-dioxo-2-thioxo-1-imidazolidiny)-2-trifluoromethyl-benzonitrile

Example 22

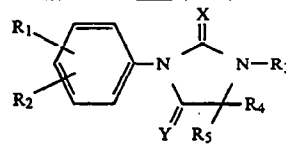
Tablets were prepared by combining 100 ml of the product of Example 3, with sufficient excipient comprising lactose amido talc and magnesium stearate to form a final tablet weight of 300 mg.

In addition to the above products, other compounds falling within the scope of the invention are those having the following formula wherein R₁, R₂, R₃, R₄, R₅ and X are as indicated in the following table.



R ₁	R ₂	X	Y	R ₄	R ₅
C≡N	CF ₃	S	O		
"	"	O	"	"	
"	"	S	"		

-continued



R ₁	R ₂	X	Y	R ₄	R ₅
"	"	O	"	"	

PHARMACOLOGICAL DATA

1) Study of androgenic Receptors

Male rats of the Sprague Dawley EOPS strain weighing 180 to 200 g, castrated 24 hours previously, were killed and the prostate was removed, weighed and homogenized at 0° C. with a potter glass in a buffered solution (Tris 10 mM, saccharose 0.25M, PMSF (phenyl methane sulfonyl fluoride) 0.1 mM, sodium molybdate 20 mM, HCl pH 7.4 into which was added extemporaneously 2 m of DTT (DL dithiothreitol) at a rate of 1 g of tissue per 8 ml of buffer solution. The homogenate was then ultracentrifuged at 0° C. for 30 minutes at 209,000 g and the aliquots of supernatant (=cytosol) were incubated for 30 minutes and 24 hours with a concentration (T) of tritiated testosterone and in the presence of increasing concentrations (0 to 2500 × 10⁻⁹M) of cold testosterone or the test products. The concentration of bound tritiated testosterone (B) was then measured for each incubate by adsorption method of carboxymethyl dextran. The relative affinity of bonding (RBA) was calculated.

The following two curves were graphed: the percentage of the bound tritiated hormone B/T as a logarithmic function of the concentration of the cold hormone and B/T as a logarithmic function of the concentration of the tested cold product. The line of the equation

$$I_{50} = \frac{(B/T_{max} + B/T_{min})}{2}$$

was determined.

B/T max = % of the bound tritiated hormone for an incubation of this tritiated hormone at concentration (T).

B/T min = % of the bound tritiated hormone for an incubation of this tritiated hormone at the concentration (T) in the presence of a large excess of cold hormone (2,500 × 10⁻⁹M).

The intersection of the straight line I₅₀ and the curves permit an evaluation of the concentrations of the cold reference hormones (CH) and the cold test product (CX) which inhibit by 50% the bonding of the tritiated hormone on the receptor. The RBA of the test product was determined by the equation

$$RBA = 100(CH)/(CX)$$

and the following results expressed in RBA were obtained with testosterone = 100.

Products of example	Incubation 24 H
3	27

OH/NH	3350, 3290 cm ⁻¹
C=N	2240 cm ⁻¹
C=O	1744 cm ⁻¹
C=N	1678 cm ⁻¹
Aromatics	1610-1574-1510 cm ⁻¹

Preparation of 1-amino-cyclopentane-carbonitrile

8.8 ml of cyclopentane were added dropwise at 0° to 8° C. to a mixture of 7.9 g of ammonium chloride, 6.14 g of sodium cyanide and 40 ml of ammonium hydroxide and, after returning to room temperature, the mixture was stirred for 16 hours and extracted with methylene chloride. The organic phase was washed with aqueous sodium chloride, dried, and evaporated to dryness at a temperature less than 30° C. The residue was distilled to obtain 11 g of the expected product with a boiling point of 55° C.±2° at 11 mm of Hg.

Example 13

4-(2,4-thioxo-1,3-diazaspiro(4,4))-nonan-3-yl)-2-trifluoromethyl-benzonitrile

Using the procedure of Example 9, 1.17 g of the product of Example 12 and 5 ml of 2 N-hydrochloric acid were reacted to obtain, after chromatography on silica (methylene chloride-acetone (9-1)), 1.108 g of the expected product with a melting point of 184° C.-185° C. and having an Rf=0.23.

IR Spectrum CHCl₃:

=C-NH	3444 cm ⁻¹
C=N	2296 cm ⁻¹
C=O	1786, 1731 cm ⁻¹
Aromatics	1616-1505 cm ⁻¹

UV Spectrum (EtOH):

Max.	258 nm	ε = 15,600
Max.	286 nm	ε = 3,500

Example 14

4-(8-imino-6-oxo-5,7-diazaspiro(3,4))-octan-7-yl)-2-trifluoromethyl-benzonitrile

Using the procedure of Example 8, 3.1 ml of an isocyanate solution and 480 mg of 1-amino-cyclobutane-carbonitrile were reacted to obtain 990 mg of the expected product melting at 192° C.-193° C. and having an Rf=0.25.

IR Spectrum CHCl₃:

OH/NH	3380, 3315 cm ⁻¹
C=N	2240 cm ⁻¹
C=N	1754 cm ⁻¹
Aromatics	1612-1571-1510 cm ⁻¹

Using the procedure of Example 12, 7.4 ml of cyclobutanone were reacted to obtain 9.2 g of 1-amino-cyclobutane-carbonitrile.

Example 15

4-(6,8-dioxo-5,7-diazaspiro(3,4))-octan-7-yl)-2-trifluoromethyl-benzonitrile

Using the procedure of Example 9, a solution of 327 ml of the product of Example 14 in 1.5 ml of chloroform and 1.9 ml of 2N-hydrochloric acid were reacted to obtain, after chromatography on silica (methylene chlo-

ride-acetone (9-1)), 341 mg of the expected product melting at 210° C.-211° C. and having an Rf=0.32.

IR Spectrum CHCl₃:

OH/NH	3390 cm ⁻¹
C=N	2240 cm ⁻¹
C=N	1787, 1737 cm ⁻¹
Aromatics	1612-1577-1508 cm ⁻¹

UV Spectrum (EtOH):

Max.	259 nm	ε = 15,700
Inf.	277, 286, 301 nm	

Example 16

4-[1-(4-hydroxybutyl(2,4-dioxo-1,3-diazaspiro(4,4))-nonan-3-yl)-2-trifluoromethyl-benzonitrile

A solution of 808 mg of the product of Example 13, in 7 ml of dimethylformamide were added dropwise over 35 minutes to 142 mg of sodium hydride and, 10 minutes after the evolution of hydrogen disengagement, 650 mg of 4-chloro-tert.-butyldimethylsilyl ether and 408 mg of sodium iodide were added. The mixture was heated at 70° C. for 3 hours and, after returning to room temperature, 71 mg of sodium hydride were added. The mixture was stirred for 10 minutes and 330 mg of the silyl were reacted and 222 mg of sodium iodide were added thereto. The mixture was heated for 45 minutes at 70° C. and, after cooling to room temperature, 60 ml of water containing about 500 mg of monopotassium phosphate were added thereto. The mixture was extracted with ethyl ether and then with ethyl acetate and the combined organic phases were dried and evaporated to dryness under reduced pressure. The residue was chromatographed over silica (methylene chloride-acetone (98-2)) to obtain 560 mg of the silyl intermediate. 550 mg of the silylated intermediate were added to 6 mg of methanol and 1.5 ml of 2N hydrochloric acid, and the mixture was stirred for 30 minutes. 30 ml of water was added and the mixture was extracted with methylene chloride. The organic phase was dried and evaporated to dryness and the residue was chromatographed over silica (methylene chloride-acetone (9-1)) and then crystallized from isopropylether to obtain 381 mg of the expected product melting at 125°-126° C. and having an Rf=0.17.

IR Spectrum CHCl₃:

OH	3625 cm ⁻¹
C=N	2235 cm ⁻¹
C=N	1773, 1721 cm ⁻¹
Aromatics	1615-1580-1505 cm ⁻¹

UV Spectrum (EtOH):

Max.	239 nm	ε = 9,800
Max.	262 nm	ε = 14,600
Inf.	286 nm	

Example 17

4-[5-(4-hydroxybutyl(-6,8-dioxo-5,7-diazaspiro(3,4))-octan-7-yl)-2-trifluoromethyl-benzonitrile

ride-acetate (95-5) mixture to recover the fraction with an $R_f=0.32$. After crystallization from ether 610 mg of the expected product melting at 172°C .- 173°C . was obtained.

IR Spectrum CHCl_3 :

$\text{C}=\text{NH}$	3015-1673 cm^{-1}
$\text{C}=\text{N}$	2236 cm^{-1}
aromatics	1615-1580-1505 cm^{-1}

Using the procedure of Example 2, 7 g of cyclobutanone were reacted to obtain 10.6 g of 1-methylamino-cyclobutane-carbonitrile.

Example 7

4-(5-methyl-8-oxo-6-thioxo(5,7-diazaspiro(3,4))-octan-7-yl)-2-trifluoromethyl-benzonitrile

Using the procedure of Example 3, 514 mg of the product of Example 6 and 1.5 ml of 2N-hydrochloric acid were reacted to obtain, after chromatography over silica (cyclohexane-ethyl acetate (6-4)) to obtain a fraction with an $R_f=0.34$. Crystallization from ether yielded 499 mg of the expected product melting at 161°C .- 162°C .

IR Spectrum CHCl_3 :

$\text{C}=\text{O}$	1754 cm^{-1}
$\text{C}=\text{N}$	2236 cm^{-1}
aromatics	1615-1583-1504 cm^{-1}

UV Spectrum (EtOH):

Inf.	239 nm	$\epsilon = 17,400$
	257 nm	$\epsilon = 21,200$
	268 nm	$\epsilon = 19,000$

Example 8

4-(1-methyl-4-imino-2-oxo-1,3-diazaspiro (4,4))-nonan-3-yl)-2-trifluoromethyl-benzonitrile

1.5 ml of a solution of the isocyanate of 3-trifluoromethyl-4-benzonitrile of Example 1 starting from phosgene and 2-trifluoromethyl-4-benzonitrile (1.6M/l) in 1,2 dichloroethane were added at

-3°C . to a solution 300 mg of 1-methylamino-cyclopentanecarbonitrile in 3 ml of 1,2 dichloroethane in the presence of 0.5 ml of triethylamine; after stirring for 40 minutes, the solution was evaporated to dryness. The residue was chromatographed over silica and diluted with a methylene chloride-ethyl acetate (95-5) mixture to obtain 620 mg of the expected product.

Example 9

4-(1-methyl-2,4-dioxo-1,3-diazaspiro(4,4))-nonan-3-yl)-2-trifluoromethyl-benzonitrile

A mixture of 535 mg of the product of Example 8 in 10 ml of methanol and 2 ml of 2N-hydrochloric acid was heated at 50°C . with stirring for 1 hour and, after returning to the room temperature, 20 ml of water were added thereto. The mixture was extracted with methylene chloride and the organic phase was evaporated to dryness under the reduced pressure. The residue was dissolved in acetone and chromatographed over silica (methylene chloride-acetone (98-2)) to obtain 325 mg of the expected product.

IR Spectrum CHCl_3 :

$\text{C}=\text{O}$	1777(m), 1724 cm^{-1} (F)
$\text{C}=\text{N}$	2238 cm^{-1}
Aromatics	1616-1576-1505 cm^{-1}

UV Spectrum (EtOH):

Inf.	236 nm	$\epsilon = 10,000$
Max.	262 nm	$\epsilon = 13,900$
Inf.	277 nm	$\epsilon = 7,200$
Inf.	286 nm	$\epsilon = 3,700$

Example 10

4-(5-methyl-8-imino-6-oxo-5,7-diazaspiro(3,4))-octan-7-yl)-2-trifluoromethyl-benzonitrile

Using the procedure of Example 8, 2 ml of a solution of isocyanate and 352 mg of 1-methylamino-cyclobutane-carbonitrile were reacted to obtain, after chromatography over silica (eluant methylene-chloride ethyl acetate (85-15)), the product with $R_f=0.20$ and finally to obtain 301 mg of the expected product melting at 144°C .- 145°C .

IR Spectrum CHCl_3 :

OH/NH	3295 cm^{-1}
$\text{C}=\text{N}$	2240 cm^{-1}
$\text{C}=\text{O}$	1740 cm^{-1}
$\text{C}=\text{N}$	1664 cm^{-1}
aromatics	1611-1572-1508 cm^{-1}

Example 11

4-(6,8-dioxo-5-methyl-5,7-diazaspiro(3,4))-octan-7-yl)-2-trifluoromethyl-benzonitrile

Using the procedure of Example 9, 0.8 g of the product of Example 10 and 3 ml of 2N-hydrochloric acid were reacted and chloroform was used as the extraction solvent. After chromatography over silica (methylene chloride-ethyl acetate (95-5)), there were obtained 465 mg of the expected product melting at 165°C .- 166°C .

IR Spectrum CHCl_3 :

$\text{C}=\text{N}$	2236 cm^{-1}
$\text{C}=\text{O}$	1778, 1726 cm^{-1}
Aromatics	1616-1579-1505 cm^{-1}

UV Spectrum (EtOH):

Max.	238 nm	$\epsilon = 11,000$
Max.	262 nm	$\epsilon = 14,000$
Inf.	278-286 nm	

Example 12

4-(4-imino-2-oxo-1,3-diazaspiro(4,4))-nonan-3-yl)-2-trifluoromethyl-benzonitrile

Using the procedure of Example 8, 3.1 ml of 1.6M solution of isocyanate and 550 mg of 1-imino-cyclopentanecarbonitrile were reacted to obtain, after chromatography on silica (methylene chloride-acetone (90-10)), 1.24 g of the expected product melting at 212°C .- 213°C .

IR Spectrum CHCl_3 :

C=O	1788-1729 cm ⁻¹
C≡N	2235 cm ⁻¹
conjugated system + Aromatics	1614-1580-1515 cm ⁻¹

UV Spectrum (EtOH):

Max.	232 nm	ε = 17,300
Max.	254 nm	ε = 22,700

Example 2

4-[1-methyl-4-imino-2-thioxo-1,3-diazaspiro(4,4)nonan-3-yl]-2-trifluoromethyl-benzonitrile

A solution of 1.36 g of 1-methylamino-cyclopentane carbonitrile in 10 ml of tetrahydrofuran were added over about 2 minutes to 2.5 g of the isocyanate of step a of Example 1 and the mixture was stirred for 40 minutes. The solvent was evaporated and the residue was chromatographed over silica (elution with methylene chloride-ethyl acetate (87.5-12.5)) to obtain 3.32 g of the expected product melting at 165°-166° C. and having an Rf=0.3 (methylene chloride-ethyl-acetate (85-15)).

IR Spectrum CHCl₃:

=NH	3310-1672 cm ⁻¹
C=N	2230 cm ⁻¹
Aromatics	1614-1577-1505 cm ⁻¹

Preparation of 1-methylamino-cyclopentane-carbonitrile:

A solution of 6.5 of potassium cyanide in 13 ml of water was added at 15°-20° C. to a solution of 8.5 g of cyclopentanone and 7 g of methylamine hydrochloride in 7.5 ml of water and returned to room temperature; the mixture was stirred for 18 hours and extracted with methylene chloride. The organic phase was washed with aqueous sodium chloride, dried, and evaporated to dryness, and the residue was distilled to obtain 4.1 g of the expected product with a boiling point of 60° C.±0.3° C. at 7 mm of Hg.

Example 3

4-(1-methyl-4-oxo-2-thioxo-1,3-diazaspiro(4,4)nonan-3-yl)-2-trifluoromethyl-benzonitrile

52 ml of methanol were added to a solution of 5.2 ml of chloroform and to 259 mg of the product of Example 2 and then 7.5 ml of 2N-hydrochloric acid were added thereto. The mixture was refluxed for 1 hour and, after cooling to room temperature, was poured into 150 ml of iced water. The mixture was extracted with chloroform and the organic phase was washed aqueous sodium chloride dried and evaporated to dryness. The residue was chromatographed over silica, eluting with ethyl acetate-cyclohexane(3-7) to obtain the fractions with Rf equal to 0.35. After crystallization, from a mixture of methylene chloride and cyclohexane, 247 mg of the desired product, melting at 162° C.-163° C. and with an Rf=0.35 (cyclohexane-ethyl acetate (7-3)) were obtained.

IR Spectrum CHCl₃:

C=O	1765 cm ⁻¹
-----	-----------------------

-continued

CN	2235 cm ⁻¹
Aromatics	1609-1578-1505 cm ⁻¹

UV Spectrum (EtOH):

Max.	234 nm	ε = 17,600
Max.	256 nm	ε = 23,800
Inf.	266 nm	ε = 20,300

Example 4

4-(4,4-diethyl-3-methyl-5-imino-2-thioxo-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile

Using the procedure of Example 2, 2.5 g of the isothiocyanate of Step a) of Example 1 and 1.39 g of the appropriate amino nitrile were reacted to obtain 3.22 g of the expected product melting at 167° C.-168° C. and having an Rf=0.27 (methylene chloride-ethyl acetate (85-15)).

IR Spectrum CHCl₃:

=NH	1304-1673 cm ⁻¹
C=N	2230 cm ⁻¹
Aromatics	1614-1576-1505 cm ⁻¹

Preparation of 1-methyl amino-diethyl-carbonitrile
Using the procedure of Example 2, 8.6 g of diethyl ketone were reacted to obtain 4.8 g of the expected product with a boiling point of 77° C. at 40 mm of Hg.

Example 5

4-(4,4-diethyl-3-methyl-5-oxo-2-thioxo-imidazolidinyl)-2-trifluoromethyl-benzonitrile

Using the procedure of Example 3, 321 mg of the product of Example 4 and 65 ml of methanol and 14 ml of 2 N-hydrochloric acid were reacted to obtain 249 mg of the expected product melting at 126° C.-127° C. and having an Rf=0.45 (cyclohexane-ethyl acetate (4-6)).

IR Spectrum CHCl₃:

C=O	1753 cm ⁻¹
C≡N	2235 cm ⁻¹
Aromatics	1615-1580-1504 cm ⁻¹

UV Spectrum (EtOH):

Max.	234 nm	ε = 17,800
Max.	254 nm	ε = 24,100
Inf.	265 nm	

Example 6

4-(5-methyl-8-imino-6-thioxo(5,7-diazaspiro(3,4))-octan-7-yl)-2-trifluoromethyl-benzonitrile

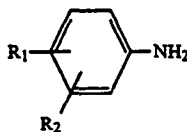
A solution of 221 mg of 1-methyl amino-cyclobutanecarbonitrile in 1 mg of 1,2-dichloroethane was added over 3 minutes to a solution of 456 mg of the isothiocyanate of Example 1 Step a) in 2 ml of 1,2-dichloroethane in the presence of 0.2 ml of triethylamine and, after stirring the mixture for 45 minutes, the solution was evaporated. The residue was chromatographed over silica and diluted with a methylene chlo-

nant tumors of cells containing androgen receptors. They are particularly useful for the treatment of breast, brain, skin and ovarian cancer and bladder, lymphatic system, liver, and kidney cancers. They are equally useful for the treatment of hirsutism, acne, seborrhea, androgenic alopecia, hyperpilosity, and in the veterinary field.

The compositions of the invention are useful in dermatology and can contain another ingredient such as an antibiotic, e.g. derivatives of azelaic acid, fusidic acid, erythromycin or with a derivative of retinoids for the treatment of acne. They can also be used with a 5 α -reductase inhibitor such as (5 α , 17 β)-1,1-dimethylethyl 3-oxo 4-aza Δ^1 -androstene-17 carboxamide (or Finasteride Merck, 11th ed.) or with azelaic acid or a blocking agent of androgen receptors for the treatment of acne, alopecia or hirsutism. In addition, they can be used with a product stimulating the growth of hair such as Minoxidil for the treatment of alopecia. The compositions can also be used in the veterinary domain and in the form of radioactive products, as well as in diagnostics as specific labels for the androgen receptors. As radioactive products they can be labeled with tritium, carbon 14, and/or iodine 125.

The novel method of the invention for inducing anti-androgenic activity in warm-blooded animals, including humans, comprises administering to the warm-blooded animals an anti-androgenically effective amount of at least one compound of Formula I and its non-toxic, pharmaceutically acceptable acid addition salts. The compounds may be administered parenterally, buccally, perlingually, rectally, or topically and the usual daily dose is 0.13 to 6.66 mg/kg depends on the condition treated, the specific compound, and the method of administration.

The starting compounds of Formula II may be prepared by reacting phosgene, when X is oxygen, or thio-phosgene, when X is sulfur, with an amine of the formula



A product of this type is described in French Patent No. 2,329,276. The amines of formula A are described in EP Patent No. 0,002,892 and French Patent No. 2,142,804.

The products of Formula III or III' are known or can be prepared from the corresponding cyanhydrins by the process described in J. Am. Chem. Soc., Vol. 75 (1953), p. 4841, or Beil. I, 4 526, or J. Org. Chem., Vol. 27 (1962), p. 2901. The compounds of Formula III wherein R₃ is other than hydrogen may be obtained by reacting a compound of the formula R'₃ Hal with 2-cyano-2-aminopropane under the conditions described above for reacting the said halide with the compounds of Formula IV. An example is described by Jilek et al, Collect. Czech. Chem. Comm., Vol. 54(8) 1989, p. 2248. The products of Formula IV' are described in French Patent No. 2,329,276.

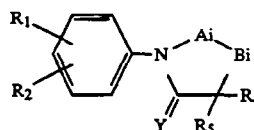
The compounds of formulae V and VI are commercially available known compounds and can be prepared by known methods.

The preparation of the compounds of Formula VI are described in the following publications: Zhur Preklad Khim., Vol. 28 (1955), p. 969-75 (CA, Vol. 50 (1956), p.

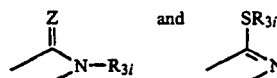
4881 a); Tetrahedron, Vol. 43 (1987), p. 1753; J. Org. Chem., Vol. 52 (1987), p. 2407; Zh. Org. Khim., Vol. 21 (1985), p.2006; J. Fluor. Chem., Vol. 17 (1981), p. 345; German Patent 637,318, European Patent 0,130,875, and Japanese Patent No. 81-121,524.

The products of Formula VI which are derivatives of hydantoin are largely used and are known in the literature such as J. Pharm. Pharmacol., 67, Vol. 19(4) (1967), p. 209-16; J. Chem. Soc., Vol. 74(2) (1972), p. 219-221; Khim. Farm. Zh., Vol 67(1)(5), p. 51-2; German Patent 2,217,914; European Patent 0,091,596 and J. Chem. Soc. Perkin. Trans. 1, Vol. 74(2), p. 48 and 219-221.

The novel intermediates of the invention are the compounds of the formula



wherein R₁, R₂ and Y have the above definitions and -Ai-Bi- is



wherein X is oxygen or sulfur and R_{3i} is R₃ with the reactive groups, among which are —OH and —NH₂, protected as above for R₃.

In the following examples, there are described several preferred embodiments to illustrate the invention. However, it should be understood that the invention is not intended to be limited to the specific embodiments.

EXAMPLES

Example 1

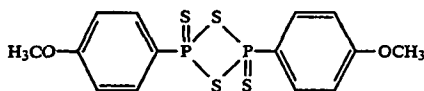
4-(3-methyl-5-oxo-2-thioxo-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile

a. A solution of 22 ml of distilled water and 1 ml of thiophosgene were slowly added to 2.23 g of 2-trifluoromethyl-4-amino-benzonitrile (prepared as in EP 000 2892) and the mixture was stirred for 1 hour and then extracted chloroform. The organic phase was washed with aqueous sodium chloride, dried, and evaporated to dryness under reduced pressure to obtain 3 grams of the desired product which was used as is.

b. A solution of 976 mg of N-methylglycine in 3.65 ml of 3 mol sodium hydroxide solution was added to 2.5 grams of the thioisocyanate of Step a) in solution in 5 ml of ethanol. The mixture was stirred for 30 minutes at room temperature and then refluxed for 1 hour. After returning to room temperature, the mixture was poured into a mixture of 20 ml of water and 10 ml of N-hydrochloric acid and extracted with chloroform. After chromatography over silica (elution with methylene chloride-acetone (95-5)), there was obtain 1.78 grams of product which was crystallized from a mixture of methylene chloride and cyclohexane to obtain 1.66 g. of the desired product melting at 220° to 221° C. and having an R_f=0.18 (cyclohexane-ethyl acetate 1-1).

IR Spectrum CHCl₃:

The transformation of $>C=O$ into $>C=S$ is effected with a Lawesson reagent of the formula



which is commercial product sold, for example, by Fluka and is described in Bull. Soc. Chim. Belg., Vol. 87 No. 3 (1987), p. 229. When two $>C=O$ groups are changed to $>C=S$, the reaction is effected with an excess of the Lawesson reagent. The same is also used when the molecule contains both $>C=O$ to $>C=S$.

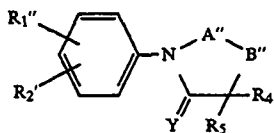
On the other hand, when part of the molecule contains two $>C=O$'s, and it is desired to obtain a product with only one $>C=S$, a deficiency of the Lawesson reagent is used to obtain a mixture of 3 products, each of two products with a $>C=O$ and $>C=S$ and one containing two $>C=S$'s. These products can be separated by known methods such as chromatography.

The reaction of the compounds of Formulas IV, IVA, or IV' with a compound of the formula R''_3 -Hal is effected in the presence of a strong base such as sodium hydride or potassium hydride in a phase transfer reaction in the presence of quaternary ammonium salts such as tert.-butyl ammonium. The protective groups of R''_3 may be those discussed above for R_3 . The reaction to eliminate the protective groups are as discussed above. For example, a tert-butyl dimethylsilyl group may be removed by hydrochloric acid as described in the examples herein.

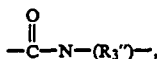
The optional esterification of the compounds of Formula I wherein R''_3 is free $-OH$ is effected under classical conditions using, for example, an acid or a functional derivative thereof such as its anhydride, e.g. acetic anhydride, in the presence of a base such as pyridine. The optional esterification or salification of the compounds of Formula I wherein R''_3 is $-COOH$ may be effected by known methods.

The optional amidification of the compounds of Formula I wherein R''_3 is $-COOH$ is also effected under classical conditions with primary or secondary amines with functional derivatives of $-COOH$, such as a symmetrical or mixed anhydride thereof.

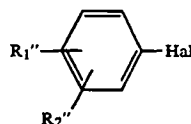
The process of the invention to prepare compounds of the Formula



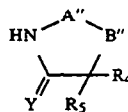
wherein R''_1 , R''_2 , and $-A''-B''-$ have the definitions of R_1 and R_2 , and $-A-B-$, except that, when $-A''-B''-$ is



and R''_3 is hydrogen or alkyl of 1 to 7 carbon atoms and Y is oxygen R''_1 is $-CN$, comprises reacting a compound of the Formula



wherein R''_1 and R''_2 have the above definitions and Hal is halogen with a compound of the formula



wherein $-A''-B''-$, R_4 , and Y have the above definitions, in the presence of a catalyst and optionally a solvent. In the compounds of formula V, the halogen is preferably chlorine but may be iodine or bromine.

The role of the catalyst is obviously to trap the hydrogen halide as it forms and to facilitate the condensation reaction of the compounds of Formulas V and VI to form the desired product. The catalyst is preferably a metal in its native form, its oxide, its salt, or it may be a base. When the catalyst is metal, it is preferably copper or nickel and the metallic salts are preferably the chloride or acetate. When the catalyst is a base, it is preferably sodium hydroxide or potassium hydroxide and dimethylsulfoxide may be added to the reaction medium.

The catalyst of the process may be selected from cuprous oxide, cupric oxide, metallic copper, or a base such as sodium hydroxide or potassium hydroxide, preferably cuprous oxide in powdered form. The solvent used preferably is a high boiling point ether such as phenyl oxide, diglyme, triglyme, or dimethylsulfoxide; also useful are high boiling point oils such as paraffin or petroleum jelly. Preferably, the process is effected in an ether solvent such as phenyl oxide, diglyme, triglyme or dimethylsulfoxide, most preferably in phenyl oxide or triglyme.

The process may be effected at atmospheric pressure or under pressure at temperatures above $100^\circ C.$, preferably above $150^\circ C.$, for more than two hours. The reaction is preferably effected with cuprous oxide in triglyme at temperatures of $200^\circ C.$ or higher for more than three hours.

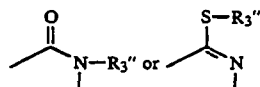
The novel anti-androgenic compositions of the invention are comprised of an anti-androgenically effective amount of at least one compound of Formula I or its non-toxic, pharmaceutically acceptable acid addition salts and an inert pharmaceutical carrier. The compositions may be in the form of tablets, dragees, capsules, syrups, suppositories, creams, pomades, lotions, or injectable solutions prepared in the usual manner.

Examples of suitable excipients are aqueous or non-aqueous vehicles, gum arabic, lactose, starch, magnesium stearate, cocoa butter, fatty bodies of animal or vegetable origin, paraffinic derivatives, glycols, wetting agents, dispersants, emulsifiers, and preservatives.

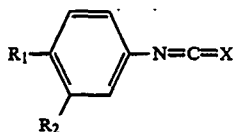
The compositions inhibit the effect of androgens on peripheral receptors and have an anti-androgenic activity useful for therapy in adults without the certain effects of a chemical castration. The compositions are useful for the treatment of adenoma and neoplasia of the prostate as well as benign hypertrophy of the prostate, they are also useful for the treatment of benign or malign-

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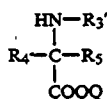
- b) hydrolysis of $C=NH$ to a ketone function and if appropriate of $>C=S$ to $>C=O$;
 c) transformation of $>C=O$ to $>C=S$, and
 d) reaction of the products of Formula IV wherein R'_3 is hydrogen after hydrolysis of $>C=NH$ to a ketone, with a compound of the formula R''_3-Hal , wherein Hal is halogen and R''_3 is R'_3 except hydrogen to obtain a compound of Formula I wherein -A-B- is



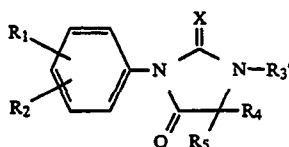
in which R'_3 has the above meaning; then if desired, the reaction of these products with an elimination agent for the optional protective groups that can be carried by R'_3 or, if appropriate, the reaction with an esterification, amidification, or salification agent, or reacting a compound of the Formula



wherein R_1 , R_2 and X have the above definitions, in the presence of a tertiary base, with a compound of the formula



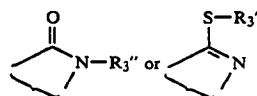
wherein R'_3 , R_4 and R_5 have the above definitions and Q is an alkali metal such as sodium or alkyl of 1 to 6 carbon atoms, to obtain a compound of the formula



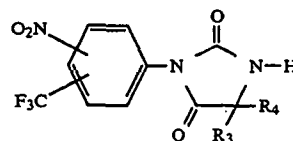
wherein R_1 , R_2 , X , R'_3 , R_4 and R_5 have the above definitions and optionally subjecting compound IVa to any one or more of the following reactions in any order:

- elimination of the optional protective groups that can be carried by R'_3 ;
- conversion of the $>C=O$ group or groups into $>C=S$ or, if appropriate conversion of $>C=S$ into $>C=O$;
- reaction of the products of Formula IVa, in which R'_3 is hydrogen, with a reagent of formula $Hal-R'_3$, wherein R'_3 is the same as R'_3 with the exception of hydrogen and Hal is halogen, to obtain the products of Formula I in which -A-B- is

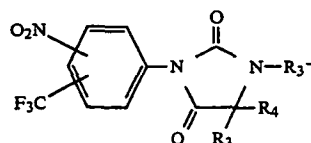
6



and optionally reaction of the latter to eliminate the protective group of R'_3 , or reaction of the same with an esterification, salification or amidification agent, or reaction of a compound of the formula R''_3-Hal as defined above with a compound of the formula



to obtain a compound of the formula



and optionally subjecting the latter to at least one of the following reactions:

- elimination of optional protective groups of R'_3 and then reaction with an esterification, salification or amidification agent; and
- transformation of $>C=O$ to $>C=S$.

The reaction of the products of Formula II with the products; of Formula III is preferably effected in an organic solvent such as tetrahydrofuran, dichloroethane, ethyl ether, or isopropyl ether in the presence of a tertiary base such as triethylamine, pyridine, methyl-ethyl pyridine.

The optional reactive functional groups of R_3 which are optionally protected in compounds of Formula III, IVa, or IV'' are $-OH$ or amino which are protected by the usual protective groups. Examples of such protective groups for $-NH_2$ are tert-butyl, tert-amyl, trichloroacetyl, chloroacetyl, benzhydryl, trityl, formyl and benzyloxycarbonyl. Examples of hydroxy protective groups are formyl, chloroacetyl, tetrahydropyranyl, trimethylsilyl, and tert-butyl dimethylsilyl.

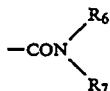
The above list of protective groups is not intended to be exhaustive and any protective group known, for example, in peptide chemistry may be used. Other known protective groups are described in French Patent 2,499,995 which is incorporated herein by reference. The optional reactions to eliminate groups are indicated in the said patent and the preferred method of elimination is acid hydrolysis with hydrochloric acid, benzene sulfonic acid, p-toluene sulfonic acid, formic acid, or trifluoroacetic acid, preferably hydrochloric acid.

The optional hydrolysis of $>C=NH$ to $>C=O$ is preferably effected by reaction with refluxing aqueous hydrochloric acid. When the hydrolysis of $>C=NH$ to $>C=O$ is effected with a molecule also containing $>C=S$, the latter may be transformed into a $>C=O$ group. The free hydroxy optionally contained in R_3 may also be transformed into $-SH$.

3

Examples of easily cleavable esters include methoxymethyl, ethoxymethyl, acyloxyalkyl such as pivaloyloxymethyl, pivaloyloxyethyl, acetoxyethyl, and acetoxyethyl; alkoxy-carbonyloxyalkyl such as methoxycarbonyloxymethyl, methoxycarbonyloxyethyl, isopropoxycarbonyloxymethyl, and isopropoxycarbonyloxyethyl. Other esters are described in European Patent No. 0,034,536.

The amidified carboxy are of the type



wherein R₆ and R₇ are individually selected from the group consisting of hydrogen and alkyl of 1 to 4 carbon atoms such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec.-butyl and tert.butyl.

Examples of mono and dialkylamino are methylamino, ethylamino, dimethylamino, diethylamino, and methylethylamino. The heterocyclic of 5 to 6 ring members optionally containing another heteroatom of



may be pyrrolyl, imidazolyl, pyridyl, pyrazinyl, pyrimidyl, indolyl, piperidino, morpholino, and piperazinyl, preferably piperidino or morpholino.

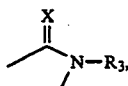
Examples of salts of salified carboxy are sodium, potassium, lithium, calcium, magnesium, ammonium, and organic bases such as methylamine, propylamine, trimethylamine, diethylamine, and triethylamine. Sodium salts are preferred.

The alkylamino and dialkylamino are preferably alkyl of 1 to 4 carbon atoms such as methylamino, ethylamino, propylamino, isopropylamino, butylamino, dimethylamino, diethylamino, and ethylmethylamino. Examples of the heterocyclics containing at least one heteroatom are saturated monocyclics such as oxiranyl, oxolanyl, dioxolanyl, imidazolidinyl, pyrazolidinyl, piperidyl, piperazinyl, and morpholinyl.

The alkyl, alkenyl, and alkynyl are optionally interrupted by one or more sulfur, oxygen, or nitrogen heteroatoms. Examples are alkoxyalkyl such as methoxymethyl, methoxyethyl, methoxypropyl, and methoxybutyl, as well as alkoxy alkoxyalkyl such as methoxyethoxymethyl.

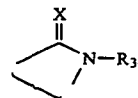
When the products of Formula I contain a salifiable amino group, the acid addition salts of non-toxic, pharmaceutically acceptable acids may be formed. Examples of said acids are inorganic acids such as nitric acid, hydrochloric acid, sulfuric acid, and phosphoric acid, as well as organic acids such as formic acid, acetic acid, propionic acid, benzoic acid, and methane sulfonic acid.

Among the preferred compounds of Formula I are those wherein Y is oxygen, -A-B- is



4

and X and R₃ are defined as above; those wherein -A-B- is

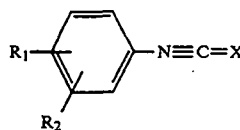


X has the above definition, and R₃ is hydrogen or alkyl of 1 to 6 carbon atoms optionally interrupted by at least one of —O—, —S—, and optionally substituted by —OH, —OH esterified with an acyl of an organic carboxylic acid of 1 to 7 carbon atoms, or free, esterified, or salified carboxy, are also worthy of special mention.

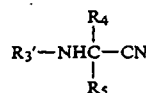
Among the preferred compounds of Formula I are those wherein R₃ is hydrogen or alkyl of 1 to 6, preferably 1 to 4, carbon atoms optionally substituted with —OH; those wherein R₂ is 3—CF₃ and R₁ is 4—CN; those wherein R₄ and R₅ are individually hydrogen, ethyl, or —CF₃; and those wherein R₄ and R₅ together with the carbon atoms form cyclobutyl or cyclopentyl.

Specific preferred compounds of Formula I are 4-(3-methyl-5-oxo-2-thioxo-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile, 4-[1-methyl-4-oxo-2-thioxo-1,3-diazaspiro (4,4)-nonan-3-yl]-2-(trifluoromethyl)-benzonitrile, and 4-(4,4-diethyl-3-methyl-5-oxo-2-thioxo-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile.

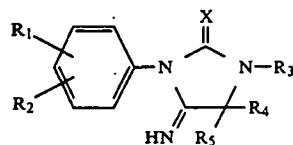
The inventive process for the preparation of a compound of Formula I comprises reacting the compound of the Formula



wherein R₁, R₂, and X have the above definitions, with a compound of the Formula



in the presence of a tertiary base, wherein R'₃ has the definition of R₃ with the active functions optionally protected, R₄ and R₅ have the above definitions with the proviso that R₄ and R₅ are not both methyl and, if R₁ is 4—NO₂, R₂ is 3—CF₃, X is —O—, and R'₃ is hydrogen; and if one of R₁ or R₅ is —CH₃, the other is —CH₂OH to obtain a compound of the formula



wherein R₁, R₂, X, R'₃, R₄ and R₅ have the above definitions and optionally subjecting the latter to at least one of the following reactions in any order:

a) elimination of the optional protective groups of R'₃;

PHENYLIMIDAZOLIDINES

STATE OF THE ART

Japanese application No. J 48087030 describes 3-phenyl-2-thiohydantoin useful for inhibiting the germination of certain plants. U.S. Pat. No. 4,907,518 describes imidazolidines different from Formula I having anti-androgenic activity. Other pertinent art includes U.S. Pat. Nos. 3,823,240; 4,873,256; 4,407,814; 4,482,739 and 4,234,736.

OBJECTS OF THE INVENTION

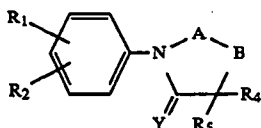
It is an object of the invention to provide novel compounds of Formula I and their non-toxic, pharmaceutically acceptable acid addition salts, novel intermediates, and a novel process for the preparation of the compounds.

It is another object of the invention to provide novel anti-androgenic compositions and a novel method of inducing anti-androgenic activity in warm-blooded animals.

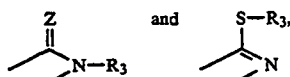
These and other objects and advantages of the invention will become obvious from the following detailed description.

THE INVENTION

The novel phenylimidazolidines of the invention have the Formula

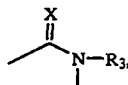


wherein R_1 and R_2 are individually selected from the group consisting of $-\text{CN}$, $-\text{NO}_2$, halogen, $-\text{CF}_3$, free carboxy, salified carboxy and carboxy esterified with lower alkyl, $-A-B-$ is selected from the group consisting of



X , is $-\text{O}-$ or $-\text{S}-$, R_3 is selected from the group consisting of hydrogen, alkyl, alkenyl and alkynyl all of up to 12 carbon atoms, aryl and aralkyl of up to 12 carbon atoms, all optionally substituted with at least one member of the group consisting of $-\text{OH}$, halogen, $-\text{SH}$, $-\text{CN}$, acyl of up to 7 carbon atoms, acyloxy of up to 7 carbon atoms, $-\text{S}-$ aryl of up to 12 carbon atoms optionally substituted with a member of the group consisting of $-\text{CF}_3$, alkyl, alkoxy, alkenyl, alkenyloxy, alkynyl and alkynyloxy, with the sulfur being optionally oxidized to sulfone or sulfoxide, free, esterified, amidified or salified carboxy, $-\text{NH}_2$, mono and dialkylamino and heterocyclic of 3 to 6 ring members and containing at least one heteroatom selected from the group consisting of oxygen, sulfur, and nitrogen, the alkyl, alkenyl, and alkynyl being optionally interrupted with at least one member of the group consisting of oxygen, nitrogen, and sulfur optionally oxidized to sulfide or sulfone, Y is $-\text{O}-$, $-\text{S}-$ or $=\text{NH}$, R_4 and R_5 are individually selected from the group consisting

of hydrogen and alkyl of up to 12 carbon atoms optionally substituted with at least one halogen or, taken together with the carbon atom to which they are attached, form cycloalkyl of 3 to 7 carbon atoms except the compounds wherein R_4 and R_5 are both methyl or one is hydroxymethyl, Y is $-\text{O}-$ or $=\text{NH}-$, $-A-B-$ is



X is oxygen, R_3 is hydrogen, R_1 is $4-\text{NO}_2$ and R_2 is $3-\text{CF}_3$; and their non-toxic, pharmaceutically acceptable acid addition salts.

The following examples of Alkyl of up to 12 carbon atoms includes methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec.-butyl, tert.-butyl, pentyl, isopentyl, sec.-pentyl, tert.-pentyl, neopentyl, hexyl, isohexyl, sec.-hexyl, tert.-hexyl, heptyl, octyl, decyl, undecyl, and dodecyl, whether branched or linear. Preferred are alkyl of 1 to 4 carbon atoms, especially methyl, ethyl, propyl, isopropyl.

Examples of alkenyl of up to 12 carbon atoms are vinyl, allyl, 1-propenyl, butenyl, pentenyl, hexenyl, preferably alkenyl of 2 to 4 carbon atoms, and especially butenyl or allyl. Examples of alkynyl of up to 12 carbon atoms are ethynyl, propargyl, butynyl, pentynyl and hexynyl, preferably 2 to 4 carbon atoms such as propargyl. Examples of cycloalkyl are cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl and cyclooctyl.

Examples of aryl are carbocyclic aryl such as phenyl and naphthyl, heterocyclic aryl of 5 to 6 ring members containing at least one heteroatom selected from the group consisting of oxygen, sulfur, and nitrogen. Examples of 5 membered ring heteroaryls are furyl, thienyl, pyrrolyl, thiazolyl, oxazolyl, imidazolyl, thiadiazolyl, pyrazolyl, and isoxazolyl. Examples of 6 membered ring heteroaryl are pyridyl, pyrimidinyl, pyridazinyl, and pyrazinyl. Examples of condensed aryls are indolyl, benzofuranyl, benzothienyl and quinoleinyl. The preferred aryl is phenyl.

Examples of aralkyl include the alkyls recited above substituted with the aryls cited above. The preferred aralkyls are phenethyl and benzyl. Examples of halogen are fluorine, chlorine, bromine, and iodine, but preferred are fluorine, chlorine, and bromine. Examples of alkyl substituted with at least one halogen are fluoromethyl, chloromethyl, bromomethyl, iodomethyl, difluoromethyl, dichloromethyl, dibromomethyl, and trifluoromethyl.

Examples of substituents for aryl and aralkyl are phenyl substituted by fluorine, $-\text{OCH}_3$, or $-\text{CF}_3$ in the p-position. Examples of acyl are preferably those of up to 7 carbon atoms, such as acetyl, propionyl, butyryl, and benzoyl, as well as valeryl, hexanoyl, acryloyl, crotonoyl, carbamoyl, and formyl. The acyloxy may be derived from the same acids, especially acetyloxy and propionyloxy.

The esterified carboxy may be alkoxy carbonyl such as methoxycarbonyl, ethoxycarbonyl, propoxycarbonyl, butoxycarbonyl, tert.-butoxycarbonyl, cyclobutyloxy carbonyl, cyclopentyloxy carbonyl and cyclohexyloxy carbonyl.



US005434176A

United States Patent [19][11] **Patent Number:** **5,434,176**

Claussner et al.

[45] **Date of Patent:** **Jul. 18, 1995**[54] **PHENYLIMIDAZOLIDINES**[75] **Inventors:** André Claussner, Villemomble;
Francois Goubet, Paris; Jean-Georges
Teutsch, Pantin, all of France[73] **Assignee:** Roussel UCLAF, France[21] **Appl. No.:** 68,736[22] **Filed:** May 28, 1993[30] **Foreign Application Priority Data**

Jul. 8, 1992 [FR] France 92 08432

[51] **Int. Cl.⁶** **A61K 31/415**[52] **U.S. Cl.** **514/391; 548/301.4**[58] **Field of Search** **548/301.4; 514/391**[56] **References Cited****U.S. PATENT DOCUMENTS**

379,038	1/1975	Magnani	514/391
4,473,393	9/1984	Nagpal	71/92
4,753,957	6/1988	Chan	514/391
4,944,791	7/1990	Schröder et al.	548/278
5,084,472	1/1992	Moguilewsky et al.	514/389

FOREIGN PATENT DOCUMENTS

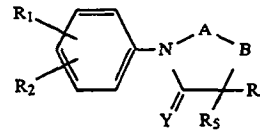
0001813	5/1979	European Pat. Off.
0091596	10/1983	European Pat. Off.
0436426	7/1991	European Pat. Off.
0494819	7/1992	European Pat. Off.
2075751	9/1971	France
2329276	10/1975	France

OTHER PUBLICATIONS

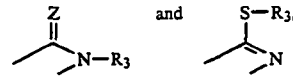
Chemical Abstracts vol. 114, No. 119, Abst. No. 185374 (1991).

Kruger et al., Arch. Pharm 311, pp. 39-47 (1978).
CA. 81, No. 7, Aug. 19, 1984.*Primary Examiner*—Joseph Paul Brust
Attorney, Agent, or Firm—Bierman and Muserlian[57] **ABSTRACT**

A compound selected from the group consisting of compounds of the formula



wherein R_1 and R_2 are individually selected from the group consisting of $-\text{CN}$, $-\text{NO}_2$, halogen, $-\text{CF}_3$, free carboxy, salified carboxy, and carboxy esterified with lower alkyl; $-\text{A}-\text{B}-$ is selected from the group consisting of



and X , R_3 , R_4 and R_5 are defined as in the specification and their non-toxic, pharmaceutically acceptable acid addition salts having anti-androgenic activity.

8 Claims, No Drawings

-continued

Products of example	Incubation 24 H
5	8

2) Determination of the androgen or anti-androgen activity by the dosage of ornithine carboxylase

Six week old male Swiss mice castrated 24 hours received oral dose of the test products as a 0.5% suspension in methyl cellulose or in ethanol by oral or percutaneous route simultaneously with a sub-cutaneous injection of 3 mg/kg of testosterone propionate in solution in corn oil to determine the anti-androgen activity. Active agonists were determined in the absence of testosterone propionate. The test compounds as well as testosterone were administered in a volume of 10 ml/kg. 20 hours after the treatments, the animals were killed, the kidneys were removed and then homogenized at 0° C. with a teflon-glass grinding apparatus in 10 volumes of buffer Tris-HCl 50 mM at a pH 7.4 containing 250 μM of pyridoxal phosphate, 0.1 mM EDTA and 5 mM of dithiothreitol. The homogenate was centrifuged at 209,000 g for 30 minutes.

At 37° C., renal ornithine decarboxylase transforms an isotopic mixture of cold ornithine and tritiated ornithine in cold putrescine and tritiated putrescine. The putrescine was then collected on selective ion-exchange papers, after drying, excess non-transformed cold and tritiated ornithine were eliminated by washing 3 times with 0.1M ammonium hydroxide. The papers were dried and the radioactivity was determined after addition of an Aqualite sample. The results expressed in fmoles (10⁻¹⁵M) of tritiated putrescine formed per hour/mg of protein are reported in the following table.

Products of Example	% Inhibition of ODL Test A
3	28
5	43

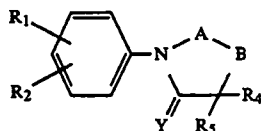
CONCLUSION

The tests show that the tested compounds of the invention possess a strong anti-androgen activity and are devoid of agonist activity.

Various modifications of the compounds and method of the invention may be made without departing from the spirit or scope thereof and it is to be understood that the invention is intended to be limited only as defined in the appended claims.

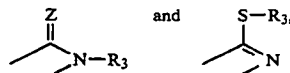
What is claimed is:

1. A method of inducing anti-androgenic activity in warm-blooded animals comprising administering to warm-blooded animals an anti-androgenically effective amount of at least one compound selected from the group consisting of compounds of the formula



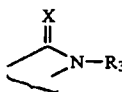
wherein R₁ and R₂ are individually selected from the group consisting of —CN, —NO₂, halogen, —CF₃, free

carboxy, salified carboxy and carboxy esterified with lower alkyl; -A-B- is selected from the group consisting of



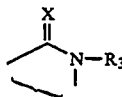
wherein X is =O or =S, R₃ is selected from the group consisting of hydrogen, alkyl, alkenyl, alkynyl, carbocyclic aryl and carbocyclic aralkyl each of up to 12 carbon atoms, all optionally substituted with at least one member of the group consisting of —OH, halogen, —SH, —CN, acyl of up to 7 carbon atoms acyloxy of up to 7 carbon atoms, —S— carbocyclic aryl of up to 12 carbon atoms optionally substituted with a member of the group consisting of —CF₃, alkyl, alkoxy, alkenyl, alkenyloxy, alkynyl, and alkynyloxy, with the sulfur being optionally oxidized to sulfone or sulfoxide, free, esterified, amidified or salified carboxy, —NH₂, mono and dialkylamino, and when the latter alkyl, alkenyl and alkynyl being optionally interrupted with at least one member of the group consisting of oxygen, nitrogen, and sulfur said sulfur being optionally oxidized to sulfide or sulfone, Y is =O, =S, or =NH; R₄ and R₅ taken together with the carbon atom to which they are attached, form cycloalkyl of 3 to 7 carbon atoms and their non-toxic, pharmaceutically acceptable acid addition salts.

2. A method of claim 1 wherein Y is —O—, -A-B- is



and X and R₃ are as defined in claim 1.

3. A method of claim 1 wherein -A-B- is



X is as defined in claim 1 and R₃ is selected from the group consisting of hydrogen and alkyl of 1 to 6 carbon atoms optionally interrupted with at least one —O— or —S— and optionally substituted with at least one member of the group consisting of —OH, OH esterified with an acyl of an organic carboxylic acid of 1 to 7 carbon atoms, and free, esterified or salified carboxy.

4. A method of claim 1 wherein R₃ is hydrogen or alkyl of 1 to 6 carbon atoms optionally substituted by —OH.

5. A method of claim 1 wherein R₃ is alkyl of 1 to 4 carbon atoms optionally substituted by —OH.

6. A method of claim 1 wherein R₂ is 3—CF₃ and R₁ is 4—CN.

7. A method of claim 1 wherein R₄ and R₅ taken together with the carbon atom to which they are attached, form cyclopentyl or cyclobutyl.

8. A method of claim 1 which is 4-[1-methyl-4-oxo-2-thioxo-1,3-diazaspiro-(4,4)-nonan-3-yl]-2-trifluoromethylbenzotrile.

* * * * *



US005750553A

United States Patent [19]

[11] Patent Number: 5,750,553

Claussner et al.

[45] Date of Patent: May 12, 1998

[54] **OPTIONALLY SUBSTITUTED
PHENYLIMIDAZOLIDINES, THEIR
PREPARATION PROCESS AND
INTERMEDIATES, THEIR USE AS
MEDICAMENTS AND THE
PHARMACEUTICAL COMPOSITIONS
CONTAINING THEM**

FOREIGN PATENT DOCUMENTS

0494819 7/1992 European Pat. Off. .
494819 7/1992 European Pat. Off. .
0578516 1/1994 European Pat. Off. .
48-8703 11/1973 Japan .

[75] Inventors: **Andre Claussner, Villemombe;**
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Primary Examiner—Mukund J. Shah
Assistant Examiner—Tamthom T. Ngo
Attorney, Agent, or Firm—Bierman, Muserlian and Lucas

[73] Assignee: **Roussel Uclaf, France**

[21] Appl. No.: **669,419**

[22] PCT Filed: **Jan. 4, 1995**

[86] PCT No.: **PCT/FR95/00004**

§ 371 Date: **Jun. 27, 1996**

§ 102(e) Date: **Jun. 27, 1996**

[87] PCT Pub. No.: **WO95/18794**

PCT Pub. Date: **Jul. 13, 1995**

[30] Foreign Application Priority Data

Jan. 5, 1994 [FR] France 94 00042
Sep. 6, 1994 [FR] France 94 10660

[51] Int. Cl.⁶ **A01N 43/50; C07D 233/40**

[52] U.S. Cl. **514/392; 548/318.5; 548/321.1**

[58] Field of Search **548/318.5, 321.1;**
514/392

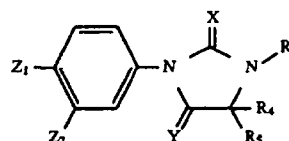
[56] References Cited

U.S. PATENT DOCUMENTS

5,411,981 5/1995 Gaillard-Kelly et al. 514/386

[57] ABSTRACT

A compound in all possible racemic, enantiomeric and diastereoisomer forms of the formula



wherein Y is —O—, Z₂ is —CF₃, Z₁ is —CN or —NO₂, X is —O— or —S—, R₃ is hydrogen or alkyl of 1 to 4 carbon atoms optionally substituted by at least one halogen or —CN and R₄ and R₅ are individually alkyl of 1 to 4 carbon atoms optionally substituted with a member of the group consisting of halogen, —OH, esterified or etherified or protected hydroxy and phenylthio optionally substituted by halogen or —OH, and wherein at least one of R₄ and R₅ is substituted having antiandrogenic activity.

6 Claims, No Drawings

1

**OPTIONALLY SUBSTITUTED
PHENYLIMIDAZOLIDINES, THEIR
PREPARATION PROCESS AND
INTERMEDIATES, THEIR USE AS
MEDICAMENTS AND THE
PHARMACEUTICAL COMPOSITIONS
CONTAINING THEM**

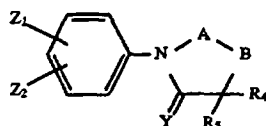
This is a 371 application of PCT/FR 95/00004 filed on Jan. 4, 1995, published as WO95/18794 Jul. 13, 1995.

The present invention relates to new optionally substituted phenylimidazolidines, their preparation process and intermediates, their use as medicaments and the pharmaceutical compositions containing them.

In the Japanese Application J 48087030 3-phenyl 2-thiohydantoin is described which are presented as inhibiting the germination of certain plants.

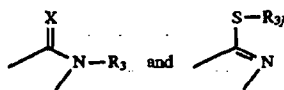
In European Patent Applications 0,494,819 and 0,578, 516 imidazolidines are described which are presented as possessing an anti-androgenic activity. The products of this Patent are however different from the products of the following Patent Application.

Therefore a subject of the present invention is the products of general formula (I):



in which:

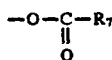
Z_1 and Z_2 , identical or different, represent a cyano, nitro radical, a halogen atom, a trifluoromethyl radical or an esterified, amidified or salified free carboxy radical, the group $-A-B-$ is chosen from the radicals



in which X represents an oxygen or sulphur atom, R_{3j} represents R_3 with the exception of the hydrogen value and R_3 is chosen from the following radicals:

a hydrogen atom,

alkyl, alkenyl, alkynyl, aryl or arylalkyl radicals having at most 12 carbon atoms, these radicals being optionally substituted by one or more substituents chosen from halogen atoms and the following radicals: optionally esterified, etherified or protected hydroxy, alkoxy, hydroxyalkyl, alkenyloxy, alkynyloxy, trifluoromethyl, mercapto, cyano, acyl, acyloxy, aryl, optionally substituted S-alkyl, S-aryl, in which the sulphur atom is optionally oxidized in the form of the sulfoxide or sulphone, free, esterified, amidified or salified carboxy, amino, mono- or dialkylamino, a cyclic radical containing 3 to 6 members and optionally containing one or more heteroatoms chosen from sulphur, oxygen or nitrogen atoms and the

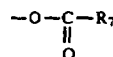


radical in which R_7 represents an alkyl, hydroxy, alkoxy, aryl or aryloxy radical, the alkyl, alkenyl or alkynyl radicals

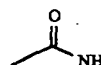
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moreover being optionally interrupted by one or more atoms of oxygen, nitrogen or sulphur optionally oxidized in the form of the sulfoxide or sulphone, the nitrogen atoms being optionally oxidized.

the aryl and aralkyl radicals moreover being optionally substituted by an alkyl, alkenyl or alkynyl, alkoxy, alkenyloxy, alkynyloxy or trifluoromethyl radical. Y represents an oxygen or sulphur atom or an NH radical. R_4 and R_5 , identical or different, represent a hydrogen atom or an alkyl radical having 1 to 12 carbon atoms optionally substituted by one or more substituents chosen from halogen atoms, the

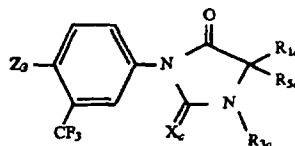


radical as defined above, the optionally esterified, etherified or protected hydroxyl radical, linear or branched phenylthio and alkylthio radicals containing at most 8 carbon atoms, phenylthio and alkylthio radicals, in which the sulphur atom can be oxidized in the form of the sulfoxide or sulphone, themselves being optionally substituted by one or more radicals chosen from halogen atoms, the optionally esterified, etherified or protected hydroxyl radical, the free, esterified, amidified or salified carboxy radical, amino, mono- or dialkylamino radicals, with the exception of the products in which R_4 and R_5 , identical or different, represent a hydrogen atom or an alkyl radical having 1 to 12 carbon atoms non-substituted or substituted by one or more halogen atoms, those in which one of R_4 or R_5 represents a methyl radical and the other represents a hydroxymethyl radical, Y represents an oxygen atom or an NH radical, the group $-AB-$ represents the radical



Z_1 in position 4 represents a nitro radical and Z_2 in position 3 represents a trifluoromethyl radical, those in which Z_1 and Z_2 both represent a halogen atom and those in which one of Z_1 and Z_2 represents a halogen atom, one of R_4 and R_5 represents an alkyl radical substituted by the group $-S-CH_3$ and R_3 represents a hydrogen atom or an alkyl radical.

b) the products of formula (I_a)



in which Z_3 represents a cyano or nitro radical.

R_{3a} represents a hydrogen atom or a linear or branched alkyl radical, containing at most 4 carbon atoms, optionally substituted by a fluorine atom, a cyano radical,

R_{4a} and R_{5a} are such that one represents a methyl radical and the other represents a methyl radical substituted by a fluorine atom or R_{4a} and R_{5a} are identical and represent a methyl radical substituted by a fluorine atom, or R_{4a} and R_{5a} form with the carbon atom to which they are linked a cyclopentyl radical, p1 X_c represents a sulphur or oxygen atom, with the exception of the product in which Z_3 represents a cyano

radical. X_a represents a sulphur atom. R_{3a} represents a methyl radical and R_{4a} and R_{5a} form with the carbon atom to which they are linked a cyclopentyl radical.

and c) the following products:

- 4-(4,4-dimethyl 2,5-dioxo 3-(2-fluoroethyl) 1-imidazol-idinyl) 2-(trifluoromethyl) benzonitrile. 5
 4-(4,4-dimethyl 2,5-dioxo 3-(2,2,2-trifluoroethyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.
 4-(4,4-dimethyl 3-(2-fluoroethyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile. 10
 4-(4,4-dimethyl 2,5-dioxo 3-(2-(2-hydroxyethoxy) ethyl) limidazolidinyl) 2-(trifluoromethyl) benzonitrile.

the said products of formula (I), (I_a) and the products mentioned being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with mineral and organic acids or with mineral and organic bases of the said products.

For the definition of the substituents indicated above and in what follows, the definitions used can have the following values:

The term alkyl designates a linear or branched radical having at most 12 carbon atoms such as for example methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl, tert-butyl, pentyl, isopentyl, sec-pentyl, tert-pentyl, neo-pentyl, hexyl, isohexyl, sec-hexyl, tert-hexyl, heptyl, octyl, decyl, undecyl, dodecyl.

Alkyl radicals having at most 4 carbon atoms are preferred and notably the methyl, ethyl, propyl, isopropyl and n-butyl radicals.

The term alkenyl designates a linear or branched radical having at most 12 carbon atoms such as for example vinyl, allyl, 1-propenyl, butenyl, pentenyl, hexenyl.

Among the alkenyl radicals, the radicals having at most 4 carbon atoms are preferred and notably the allyl or butenyl radical.

The term alkynyl designates a linear or branched radical having at most 12 carbon atoms such as for example ethynyl, propargyl, butynyl, pentynyl or hexynyl.

Among the alkynyl radicals, the radicals having at most 4 carbon atoms are preferred and notably the propargyl radical.

By aryl is meant the carbocyclic aryl radicals such as phenyl or naphthyl or the monocyclic heterocyclic aryl radicals with 5 or 6 members or the heterocyclic aryl radicals constituted by condensed rings, containing one or more heteroatoms preferably chosen from oxygen, sulphur and nitrogen. Among the heterocyclic aryls with 5 members, the following radicals can be mentioned: furyl, thienyl, pyrrolyl, thiazolyl, oxazolyl, imidazolyl, thiadiazolyl, pyrazolyl, isoxazolyl, tetrazolyl.

Among the heterocyclic aryl radicals with 6 members, the pyridyl, pyrimidinyl, pyridazinyl and pyrazinyl radicals can be mentioned.

Among the condensed aryl radicals, the indolyl, benzofuranyl, benzothienyl and quinolinyl radicals can be mentioned.

The phenyl, tetrazolyl and pyridyl radicals are preferred.

By arylalkyl is meant the radicals resulting from the combination of the alkyl radicals and the aryl radicals mentioned above.

The benzyl, phenylethyl, pyridylmethyl, pyridylethyl or tetrazolylmethyl radicals are preferred.

By halogen is meant, of course, the fluorine, chlorine, bromine or iodine atoms.

The fluorine, chlorine or bromine atoms are preferred.

As particular examples of alkyl radicals substituted by one or more halogens, the following radicals can be mentioned: monofluoro, chloro, bromo or iodomethyl, difluoro, dichloro or dibromomethyl and trifluoromethyl.

As particular examples of substituted aryl or aralkyl radicals, there can be mentioned those in which the phenyl radical is substituted in the para position by a fluorine atom or by a methoxy or trifluoromethyl radical.

By acyl radical is preferably meant a radical having at most 7 carbon atoms such as the acetyl, propionyl, butyryl or benzoyl radical, but it can also represent a valeryl, hexanoyl, acryloyl, crotonoyl or carbamoyl radical; the formyl radical can also be mentioned.

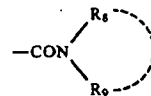
By acyloxy radical is meant the radicals in which the acyl radicals have the meaning indicated above and for example the acetoxy or propionyloxy radicals.

By esterified carboxy is meant for example the radicals such as the alkyloxycarbonyl radicals, for example methoxy-carbonyl, ethoxycarbonyl, propoxycarbonyl, butyl or tert-butyloxycarbonyl.

There can also be mentioned the radicals formed with the easily cleavable ester remainders such as the methoxymethyl, ethoxymethyl radicals; the acyloxyalkyl radicals such as pivaloyloxymethyl, pivaloyloxyethyl, acetoxymethyl or acetoxylethyl; the alkyloxycarbonyloxy alkyl radicals such as the methoxycarbonyloxy methyl or ethyl radicals, the isopropylloxycarbonyloxy methyl or ethyl radicals.

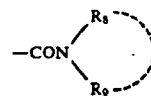
A list of such ester radicals can be found for example in the European Patent EP 0.034.536.

By amidified carboxy is meant the groups of

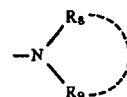


type in which the R_8 and R_9 radicals, identical or different, represent a hydrogen atom or an alkyl radical having 1 to 4 carbon atoms such as the methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl or tert-butyl radicals.

In the groups

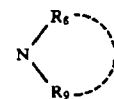


defined above, those in which the



radical represents the amino, mono- or dimethylamino radical are preferred.

The



radical can also represent a heterocycle which may contain an additional heteroatom. The pyrrolyl, imidazolyl, indolyl, piperidino, morpholino, piperazinyl radicals can be mentioned. The piperidino or morpholino radicals are preferred.

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By salified carboxy is meant the salts formed for example with an equivalent of sodium, potassium, lithium, calcium, magnesium or ammonium. The salts formed with the organic bases such as methylamine, propylamine, trimethylamine, diethylamine, triethylamine, can also be mentioned.

The sodium salt is preferred.

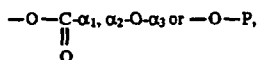
By alkylamino radical is preferably meant the radicals in which the alkyl contains at most 4 carbon atoms. The methylamino, ethylamino, propylamino or butyl (linear or branched) amino radicals can be mentioned.

Similarly, by dialkylamino radical is preferably meant the radicals in which the alkyl contains at most 4 carbon atoms. For example the dimethylamino, diethylamino, methylethylamino radicals can be mentioned.

By heterocyclic radical containing one or more heteroatoms is meant for example the saturated heterocyclic, monocyclic radicals such as the following radicals: oxiranyl, oxolanyl, dioxolanyl, pyrrolidinyl, imidazolidinyl, pyrazolidinyl, piperidyl, piperazinyl or morpholinyl.

By alkyl, alkenyl, or alkynyl radicals optionally interrupted by a heteroatom chosen from sulphur, oxygen or nitrogen atoms is meant the radicals containing one or more of these atoms, identical or different in their structure, these heteroatoms obviously not being able to be situated at the end of the radical. There can be mentioned for example the alkoxyalkyl radicals such as methoxymethyl or methoxyethyl or also the alkoxy alkoxyalkyl radicals such as methoxyethoxymethyl.

By esterified, etherified or protected hydroxyl radical is meant respectively the radicals



formed from a hydroxyl radical, according to the usual methods known to a man skilled in the art and in which P represents a protective group.

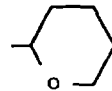
α_1 , α_2 and α_3 represent in particular an alkyl, alkenyl, alkynyl, aryl or arylalkyl radical, having at most 12 carbon atoms and optionally substituted as defined above in particular for R_3 .

Examples of the protective group P, as well as the formation of the protected hydroxyl radical, are given in particular in the common book for a man skilled in the art: Protective Groups in Organic Synthesis, Theodora W. Greene, Harvard University, printed in 1981 by Wiley-Interscience Publishers, John Wiley & Sons.

The protective group of the hydroxyl radical that can be represented by P, can be chosen from the following list: for example formyl, acetyl, chloroacetyl, bromoacetyl, dichloroacetyl, trichloroacetyl, trifluoroacetyl, methoxyacetyl, phenoxyacetyl, benzoyl, benzoylformyl, p-nitrobenzoyl. The following groups can also be mentioned: ethoxycarbonyl, methoxycarbonyl, propoxycarbonyl, $\beta\beta$ -trichloroethoxycarbonyl, benzyloxycarbonyl, tert-butoxycarbonyl, 1-cyclopropylethoxycarbonyl, tetrahydro-pyranyl, tetrahydrothiopyranyl, methoxytetrahydropyranyl, trityl, benzyl, 4-methoxybenzyl, benzhydryl, trichloroethyl, 1-methyl 1-methoxyethyl, phthaloyl, propionyl, butyryl, isobutyryl, valeryl, isovaleryl, oxalyl, succinyl and pivaloyl, phenylacetyl, phenylpropionyl, mesyl, chlorobenzoyl, para-nitrobenzoyl, para-tert-butylbenzoyl, caprylyl, acryloyl, methylcarbamoyl, phenylcarbamoyl, naphthylcarbamoyl.

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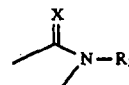
P can in particular represent the radical



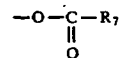
or also a silicon derivative such as trimethylsilyl.

When the products of formulae (I) and (I_a) as defined above contain an amino radical salifiable by an acid it is understood that these acid salts are also part of the invention. There can be mentioned the salts formed with hydrochloric or methanesulphonic acids for example.

A particular subject of the invention is the products of formula (I) as defined above, in which Z_1 and Z_2 represent a trifluoromethyl, nitro or cyano radical, Y represents an oxygen atom or an NH radical, the group —A—B— represents the radical:



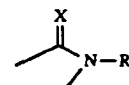
in which X represents an oxygen or sulphur atom, R_3 represents a hydrogen atom, a linear or branched alkyl radical containing at most 6 carbon atoms, optionally interrupted by one or more oxygen or sulphur atoms, a phenyl or pyridyl radical, these radicals being optionally substituted by one or more radicals chosen from halogen atoms, the following radicals: phenyl, optionally esterified, etherified or protected hydroxyl, alkoxy, cyano, trifluoromethyl, hydroxyalkyl, free, esterified, amidified or salified carboxy, amino, mono- or dialkylamino, the nitrogen atom of the pyridyl radical being optionally oxidized, R_4 and R_5 represent a linear or branched alkyl radical containing at most 6 carbon atoms, optionally substituted by one or more radicals chosen from optionally esterified, etherified or protected hydroxyl radicals, halogen atoms, the



radical in which R_7 represents a linear or branched alkyl or alkoxy radical and alkylthio and phenylthio radicals, themselves optionally substituted by one or more radicals chosen from halogen atoms and the free, esterified, etherified or protected hydroxyl radical.

the said products of formula (I) being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with mineral and organic acids or with mineral and organic bases of the said products of formula (I).

Among these products, a particular subject of the invention is the products of formula (I) as defined above, in which Z_1 and Z_2 represent a trifluoromethyl, nitro or cyano radical, Y represents an oxygen atom or an NH radical, the group —A—B— represents the group:



in which X represents an oxygen or sulphur atom, R_3 represents a hydrogen atom or an alkyl radical having 1 to

6 carbon atoms optionally substituted by one or more radicals chosen from halogen atoms and the optionally esterified, etherified or protected hydroxyl radical, the free, esterified, amidified or salfied carboxy radical and the cyano radical, the alkyl radical being optionally interrupted by one or more oxygen or sulphur atoms.

R_4 and R_5 represent an alkyl radical containing at most 6 carbon atoms optionally substituted by one or more radicals chosen from the optionally esterified, etherified or protected hydroxyl radical, halogen atoms and alkylthio and phenylthio radicals themselves optionally substituted by one or more radicals chosen from halogen atoms and the hydroxyl radical, the said products of formula (I) being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with mineral and organic acids or with mineral and organic bases of the said products of formula (I).

Among these products, a more particular subject of the invention is the products of formula (I) as defined above, in which Y represents an oxygen atom or an NH radical, Z_2 in position 3 represents a trifluoromethyl radical and Z_1 in position 4 represents a cyano or nitro radical, X represents an oxygen or sulphur atom.

R_3 represents a hydrogen atom or an alkyl radical having at most 4 carbon atoms, optionally substituted by one or more radicals chosen from halogen atoms or the cyano radical, R_4 and R_5 , identical or different, represent a linear or branched alkyl radical containing at most 4 carbon atoms optionally substituted by a free, esterified, etherified or protected hydroxyl radical, a halogen atom, or a phenylthio radical optionally substituted by a halogen atom or a free, esterified, etherified or protected hydroxyl radical.

the said products of formula (I) being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with mineral and organic acids or with mineral and organic bases of the said products of formula (I).

Among the preferred products of the invention, there can be mentioned more particularly the products of formula (I) as defined above the names of which follow:

2-(trifluoromethyl) 4-(4-(hydroxymethyl) 4-methyl 2,5-dioxo 1-imidazolidinyl) benzonitrile.

4-(3,4-dimethyl) 4-(hydroxymethyl) 5-oxo 2-thioxo 1-imidazolidinyl 2-(trifluoromethyl) benzonitrile.

2-(trifluoromethyl) 4-(4-(hydroxymethyl) 3,4-dimethyl 2,5-dioxo 1-imidazolidinyl) benzonitrile.

4-(2,5-dioxo 3-(2-fluoroethyl) 4-(hydroxymethyl) 4-methyl 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.

1,5-dimethyl 5-(hydroxymethyl) 3-(4-nitro 3-(trifluoromethyl) phenyl) 2,4-imidazolidinedione.

4-(4,4-bis(hydroxymethyl)-2,5-dioxo-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile the said products of formula (I) being in all their racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with mineral and organic acids or with mineral and organic bases of the said products of formula (I).

Also a quite particular subject of the invention is the products of formula (I_a) as defined above, the names of which follow:

4-(4-(fluoromethyl) 3,4-dimethyl) 2,5-dioxo 1-imidazolidinyl 2-(trifluoromethyl) benzonitrile.

4-(3,4-dimethyl) 4-(fluoromethyl) 5-oxo 2-thioxo 1-imidazolidinyl 2-(trifluoromethyl) benzonitrile.

4-(2,5-dioxo 3-(2-fluoroethyl) 4-(fluoromethyl) 4-methyl 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.

4-(2,4-dioxo 1,3-diazaspiro(4.4)nonan-3-yl) 2-(trifluoromethyl) benzonitrile.

4-(2,4-dioxo 1-(2-fluoroethyl) 1,3-diazaspiro(4.4)nonan-3-yl) 2-(trifluoromethyl) benzonitrile.

1,5-dimethyl 5-(fluoromethyl) 3-(4-nitro 3-(trifluoromethyl) phenyl) 2,4-imidazolidinedione.

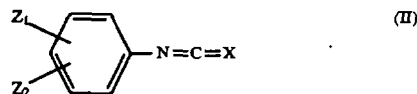
3-(4-cyano 3-(trifluoromethyl) phenyl) 2,4-dioxo 5-(fluoro-methyl) 5-methyl 1-imidazolidinacetonitrile.

4-(4,4-bis-(fluoromethyl) 3-methyl 5-oxo 2-thioxo 1-imidazolidinyl imidazolidinyl 2-(trifluoromethyl) benzonitrile.

the said products of formula (I_a) being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms, as well as the addition salts with mineral and organic acids or with mineral and organic bases of the said products of formula (I_a)

Also a subject of the invention is a preparation process for the products of formula (I), (I_a) and cited products as defined above characterized in that:

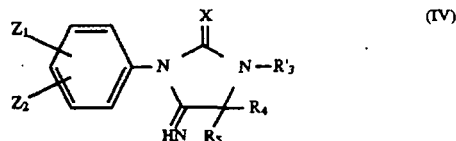
either a product of formula (II):



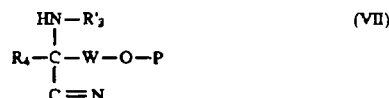
in which Z_1 , Z_2 and X have the meaning indicated above, is reacted in the presence of a tertiary base with a product of formula (III):



in which R_4 and R_5 have the meaning indicated above and R_3 has the values indicated above for R_3 in which: the optional reactive functions are optionally protected and it being understood that R_4 and R_5 do not represent simultaneously a methyl radical and that if Z_1 represents an NO_2 radical in position 4, Z_2 represents a CF_3 radical in position 3, X represents an oxygen atom and R_3 represents a hydrogen atom, then one of R_4 or R_5 does not represent a CH_3 radical and the other a CH_2OH radical, in order to obtain a product of formula (IV):

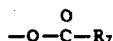


in which Z_1 , Z_2 , X, R_3 , R_4 and R_5 have the previous meaning, or the product of formula (II), as defined above, is reacted in the presence of a tertiary base with a product of formula (VII):

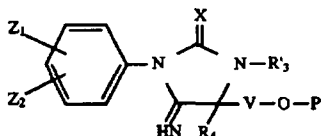


in which W has the meaning indicated above for R_3 with the exception of the hydrogen atom, the alkyl radical substituted by a free, esterified, etherified or protected hydroxyl radical and the value

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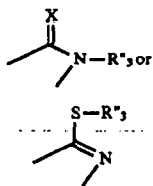
as defined above, and P represents a protective group of OH or a radical such that —O—P represents an etherified hydroxyl radical and R₃ and R₄ have the meaning indicated above, in order to obtain a product of formula (VIII):



in which X, Z₁, Z₂, R₃, R₄, W and P have the meaning indicated above, of which product of formula (VIII) if necessary and if desired, the OH radical can be released from OP which can then if necessary and if desired, be esterified or converted into a halogen radical,

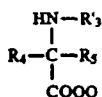
which products of formulae (IV) and (VIII), if necessary or if desired, are subjected to any one or more of the following reactions, in any order:

- elimination reaction of the optional protective groups that can be carried by R₃;
- hydrolysis reaction of the >C=NH group into a carbonyl function and if appropriate conversion of the >C=S group into the >C=O group;
- conversion reaction of the >C=O group or groups into the >C=S group;
- action on the products of formula (IV) or (VIII) in which R₃ represents a hydrogen atom, and after hydrolysis of the >C=NH group into a carbonyl function, of a reagent of formula Hal—R³ in which R³ has the values of R₃ with the exception of the hydrogen value and Hal represents a halogen atom, in order to obtain products of formulae (I), (I_a) and cited products as defined above, in which the —A—B— group represents the group



in which R³ has the meaning indicated previously then, if desired, action on these products, of an elimination agent of the optional protective groups that can be carried by R³ or if appropriate, action of an esterification, amidification or salification agent,

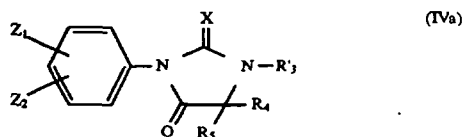
or a product of formula (II) as defined above is reacted in the presence of a tertiary base with a product of formula (III)



in which R₃, R₄ and R₅ have the meaning indicated above and Q represents either an alkali metal atom or an alkyl radical containing 1 to 6 carbon atoms, in order to obtain a

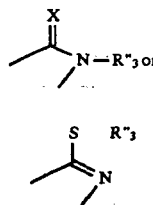
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product of formula (IVa):



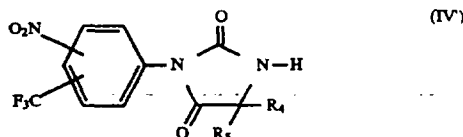
in which X, Z₁, Z₂, R₃, R₄ and R₅ have the meaning indicated above, which if desired is subjected to any one or more of the following reactions, in any order:

- elimination reaction of the optional protective groups that can be carried by R₃;
- conversion reaction of the >C=O group or groups into the >C=S group or if appropriate of the >C=S group into the >C=O group;
- action on the products of formula (IVa) in which R₃ represents a hydrogen atom of a reagent of formula Hal—R³ in which R³ has the values of R₃ with the exception of the hydrogen value and Hal represents a halogen atom, in order to obtain products of formulae (I), (I_a) and cited products as defined above, in which the A—B— group represents the group

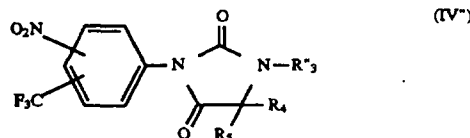


in which R³ has the meaning indicated previously then, if desired, action on these products of an elimination agent of the optional protective groups that can be carried by R³ or if appropriate, action of an esterification, amidification or salification agent,

or a reagent of formula Hal—R³ in which Hal and R³ have the values indicated previously is reacted on a product of formula (IV')



in order to obtain a product of formula (IV'')



which product of formula (IV), (IVa), (IV'), (IV'') or (VIII) represents or does not represent a product of formula (I) and which, in order to obtain if necessary or if desired a product of formula (I), is subjected to any one or more of the following reactions in any order:

- elimination reaction of the optional protective groups that can be carried by R₃ then if appropriate action of an esterification, amidification or salification agent;
- conversion reaction of the >C=O group or groups into >C=S groups, the said products of formula (I) thus

obtained being in all the possible racemic, enantiomeric and diastereoisomeric isomer forms.

The action of the products of formula (II) with the products of formula (III) is preferably carried out in an organic solvent such as tetrahydrofuran or dichloroethane but ethyl ether or isopropyl ether can also be used.

The operation is carried out in the presence of a tertiary base such as triethylamine or also pyridine or methylethylpyridine.

The optional reactive functions that can be contained by R_3 and that are optionally protected, are hydroxy or amino functions. The usual protective groups are used to protect these functions. There can be mentioned for example the following protective groups of the amino radical: tert-butyl, tert-amyl, trichloroacetyl, chloroacetyl, benzhydryl, trityl, formyl, benzyloxycarbonyl.

As protective group of the hydroxy radical there can be mentioned the radicals such as formyl, chloroacetyl, tetrahydropyranyl, trimethylsilyl, tert-butyl dimethylsilyl.

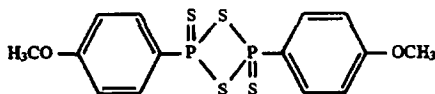
It is well understood that the above list is not limitative and that other protective groups, for example known in the chemistry of the peptides, can be used. A list of such protective groups is found for example in the French Patent BF 2.499.995 whose content is incorporated here by way of reference.

The optional elimination reactions of the protective groups are carried out as indicated in the said Patent BF 2.499.995. The preferred method of elimination is acid hydrolysis using acids chosen from hydrochloric, benzene sulphonic or paratoluene sulphonic, formic or trifluoroacetic acids. Hydrochloric acid is preferred.

The optional hydrolysis reaction of the $>C=NH$ group into the ketone group is also preferably carried out using an acid such as aqueous hydrochloric acid for example under reflux.

When the hydrolysis of the $>C=NH$ group into the carbonyl group is carried out on a molecule also containing a $>C=S$ group, this can be converted into the $>C=O$ group. The free OH radical that can optionally be contained by R_3 can then be converted into the SH radical.

The conversion reaction of the $>C=O$ group or groups into the $>C=S$ group is carried out using the so-called Lawesson reagent of formula:



which is a product marketed for example by the FLUKA company and whose use is described for example in the publication: Bull. Soc. Chim. Belg. Vol. 87, No. 3, (1987) p. 229.

When it is desired to convert two $>C=O$ functions into two $>C=S$ functions the operation is carried out in the presence of an excess of Lawesson reagent. The same goes when one starts with a molecule containing a $>C=S$ function and a $>C=O$ function and it is desired to convert the said $>C=O$ function into a $>C=S$ function.

On the other hand when one starts with a molecule containing two $>C=O$ functions and it is desired to obtain a product containing only a single $>C=S$ function. The operation is carried out in the presence of a deficit of Lawesson reagent. Then in general a mixture of three products is obtained: each of the two products containing a $>C=O$ function and a $>C=S$ function and the product containing two $>C=S$ functions. These products can then be separated by the usual methods such as chromatography.

The action on the product of formula (IV), (IVa), (IV') or (VIII) of the reagent of formula $Hal-R''_3$ is carried out in the presence of a strong base such as sodium or potassium hydride. The operation can be carried out by phase transfer reaction in the presence of quaternary ammonium salts such as tert-butyl ammonium.

The protective groups that can be carried by the R''_3 substituent can be for example one of those previously mentioned for R_3 . The elimination reactions of the protective groups are carried out under the conditions indicated above.

An example of the elimination of the tertbutyldimethylsilyl group by means of hydrochloric acid is given hereafter in the examples.

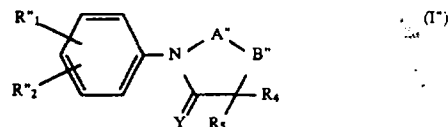
The optional esterification of the products of formulae (I), (Ia) and as defined above, in which R''_3 contains a free OH radical is carried out under standard conditions. There can be used an acid or a functional derivative, for example an anhydride such as acetic anhydride in the presence of a base such as pyridine.

The optional esterification or salification of the products of formulae (I), (Ia) and the cited products as defined above, in which R''_3 represents a COOH group, is carried out under standard conditions known to a man skilled in the art.

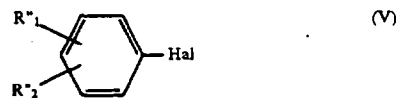
The optional amidification of the products of formulae (I), (Ia) and as defined above, in which R''_3 contains a COOH radical, is carried out under standard conditions. A primary or secondary amine can be used on a functional derivative of the acid for example a symmetrical or mixed anhydride.

The reaction of the product of formula (II) as defined above with the product of formula (VII) as defined above to give the product of formula (VIII) as defined above, can be carried out notably in the presence of methylene chloride at a temperature of approximately $-30^\circ C$.

Also a subject of the present invention is a preparation process for the products of formula (I''):



in which R_4 and R_5 are defined as above and R''_1 , R''_2 , $-A''-B''$ have the meanings indicated above for Z_1 , Z_2 and $-A-B-$, it being understood that when $-A''-B''$ represents a $-CO-N(R''_3)-$ group in which R''_3 represents a hydrogen atom or a linear or branched alkyl radical having at most 7 carbon atoms and Y represents an oxygen atom, R''_1 represents a cyano radical, process characterized in that a product of formula (V):



in which R''_1 and R''_2 have the previous meanings and Hal represents a halogen atom, is reacted with a product of formula (VI):



in which $-A''-B''$, R_4 , R_5 and Y have the meaning indicated above, the reaction being carried out in the presence of a catalyst and optionally of a solvent.

With regard to the products of formula (V), the term Hal preferably designates the chlorine atom, but can also represent a bromine or iodine atom.

The rôle of the catalyst is probably to trap the hydrogen halide which is released and thus to facilitate the condensation reaction of the product of formula (V) with the product of formula (VI) to give the desired product.

A more particular subject of the invention is a process as defined above in which the catalyst is a metal in native or oxidized form or a base.

When the catalyst used is a metal, this metal can be copper or nickel. It can be in native form, in the form of the metal oxide or also in the form of the metal salts.

The metal salts can be a chloride or an acetate.

When the catalyst is a base, this base can be for example soda or potash and, if desired, dimethylsulphoxide can be added to the reaction medium.

A more particular subject of the invention is a process as defined above in which the catalyst is chosen from cuprous oxide, cupric oxide, copper in the native form and a base such as soda or potash.

The copper in native form used as catalyst is preferably in powdered form.

A particular subject of the invention is a process as defined above in which the catalyst is cuprous oxide.

The solvent used is preferably chosen from ethers with a high boiling point such as, for example, phenyl oxide, diglyme, triglyme and dimethylsulphoxide but can also be, for example, an oil with a high boiling point such as paraffin or vaseline.

A more particular subject of the invention is a process as defined above characterized in that the operation is carried out in the presence of a solvent of ether type such as phenyl oxide, diglyme, triglyme or dimethylsulphoxide.

A quite particular subject of the invention is a process as defined above in which the solvent used is phenyl oxide or triglyme.

The preparation process of the desired product defined above can be carried out under pressure or at atmospheric pressure, preferably at a high temperature.

Therefore a subject of the invention is a process as defined above characterized in that the reaction is carried out at a temperature higher than 100° C. and preferably higher than 150° C.

A more particular subject of the invention is a process as defined above characterized in that the reaction is carried out for more than 2 hours.

A very particular subject of the invention is a process as defined above characterized in that the reaction is carried out in the presence of cuprous oxide, in triglyme, at a temperature higher than or equal to 200° C. and for more than 3 hours.

The products which are a subject of the present invention possess useful pharmacological properties, in particular they fix on the androgen receptor and they present an anti-androgenic activity.

Tests given in the experimental part illustrate these properties.

These properties make the products of formula (I) as defined above of the present invention of use as medicaments mainly for:

the treatment of adenomas and neoplasias of the prostate as well as benign hypertrophy of the prostate, alone or in combination with analogues of LHRH. They can also be used in the treatment of benign or malignant tumours possessing androgen receptors and more particularly cancers of the breast, skin, ovaries, bladder, lymphatic system, kidney and liver.

the treatment of cutaneous affections such as acne, hyperseborrhea, alopecia or hirsutism. These products can therefore be used in dermatology alone or in combination with antibiotics such as derivatives of azelaic and fusidic acids, erythromycin, as well as derivatives of retinoic acid or an inhibitor of 5 α -reductase such as (5 α , 17 β)-1,1-dimethylethyl 3-oxo 4-aza-androst-1-ene 17-carboxamide (or Finasteride, Merck 11th Ed.) for the treatment of acne, alopecia or hirsutism. They can also be combined with a product stimulating hair growth such as Minoxidil for the treatment of alopecia.

The products of formulae (I), (I_a) and the cited products as defined above, in radioactive form (tritium, carbon 14, iodine 125 or fluorine 18) can also be used as a specific marker for the androgen receptors. They can also be used in diagnostics in medical imaging.

The products of formulae (I), (I_a) and the cited products as defined above can also be used in the veterinary domain for the treatment of behavioural disorders such as aggressiveness, androgen-dependent affections, such as circum anulum in dogs, and tumours having androgen receptors. They can also be used to bring about a chemical castration in animals.

Therefore a subject of the invention is the use, as medicaments, of the pharmaceutically acceptable products of formulae (I) and (I_a) as defined above.

Also a subject of the invention is the use, as medicaments, of the following products:

4-(4,4-dimethyl 2,5-dioxo 3-(2-fluoroethyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.
4-(4,4-dimethyl 2,5-dioxo 3-(2,2,2-trifluoroethyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.
4-(4,4-dimethyl 3-(2-fluoroethyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.
4-(4,4-dimethyl 2,5-dioxo 3-(2-(2-hydroxyethoxy) ethyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.

A particular subject of the invention is the use, as medicaments, of the following products:

2-(trifluoromethyl) 4-(4-(hydroxymethyl) 4-methyl 2,5-dioxo 1-imidazolidinyl) benzonitrile.
4-(3,4-dimethyl) 4-(hydroxymethyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.
2-(trifluoromethyl) 4-(4-(hydroxymethyl) 3,4-dimethyl 2,5-dioxo 1-imidazolidinyl) benzonitrile.
4-(2,5-dioxo 3-(2-fluoroethyl) 4-(hydroxymethyl) 4-methyl 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.
1,5-dimethyl 5-(hydroxymethyl) 3-(4-nitro 3-(trifluoromethyl) phenyl) 2,4-imidazolidinedione.
4-(4,4-bis (hydroxymethyl) 2,5-dioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.

Also a particular subject of the invention is the use, as medicaments, of the following products of formula (I_a):

4-(4-(fluoromethyl) 3,4-dimethyl) 2,5-dioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.
4-(3,4-dimethyl) 4-(fluoromethyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile:
4-(2,5-dioxo 3-(2-fluoroethyl) 4-(4-(fluoromethyl) 4-methyl 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.
4-(2,4-dioxo 1,3-diazaspiro(4.4)nonan-3-yl) 2-(trifluoromethyl) benzonitrile.
4-(2,4-dioxo 1-(2-fluoroethyl) 1,3-diazaspiro(4.4)nonan-3-yl) 2-(trifluoromethyl) benzonitrile.
1,5-dimethyl 5-(fluoromethyl) 3-(4-nitro 3-(trifluoromethyl) phenyl) 2,4-imidazolidinedione.
3-(4-cyano 3-(trifluoromethyl) phenyl) 2,4-dioxo 5-(fluoromethyl) 5-methyl 1-imidazolidinacetoneitrile.

4-(4,4-bis-(fluoromethyl) 3-methyl 5-oxo 2-thioxo 1-imidazolidinyl 2-(trifluoromethyl) benzonitrile.

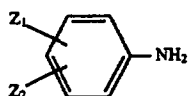
The products can be administered by parenteral, buccal, perlingual, rectal or topical route.

Also a subject of the invention is the pharmaceutical compositions, characterized in that they contain, as active ingredient, at least one of the medicaments of formulae (I), (Ia) and the cited products as defined above.

These compositions can be presented in the form of injectable solutions or suspensions, tablets, sugar-coated tablets, capsules, syrups, suppositories, creams, ointments and lotions. These pharmaceutical forms are prepared according to the usual methods. The active ingredient can be incorporated with excipients usually employed in these compositions, such as aqueous or non-aqueous vehicles, talc, gum arabic, lactose, starch, magnesium stearate, cocoa butter, fatty substances of animal or vegetable origin, paraffin derivatives, glycols, various wetting, dispersing or emulsifying agents, preservatives.

The usual dose, variable according to the patient being treated and the affection in question, can be, for example, from 10 mg to 500 mg per day for man, by oral route.

The products of formula (II) used at the start of the invention can be obtained by the action of phosgene when X represents an oxygen atom or thiophosgene when X represents a sulphur atom on the corresponding amine of formula (A):



(A)

An example of such a preparation is given hereafter in the experimental part. A product of this type is also described in the French Patent BF 2,329,276.

The amines of formula (A) are described in the European Patent EP 0,002,892 or the French Patent BF 2,142,804.

The products of formula (III) or (III') are known or can be prepared from the corresponding cyanhydrin according to the process described in the publications: *J. Am. Chem. Soc.* (1953), 75, 4841. BEIL I 4 526 or *J. Org. Chem.* 27 2901 (1962).

The products of formula (III) in which R₃ is different from a hydrogen atom can be obtained by the action of a product of formula R₃ Hal on 2-cyano 2-amino propane under the conditions set out above for the action of R₃ Hal on the products of formula (IV). An example of this type of preparation is described in the reference: Jilek et al. *Collect. Czech. Chem. Comm.* 54(8) 2248 (1989).

The products of formula (IV') are described in the French Patent BF 2,329,276.

The products of formulae (V) and (VI), used at the start of a process which is a subject of the invention, for obtaining the products of formulae (I), (I_a) as defined above, are known and commercially available or can be prepared according to methods known to a man skilled in the art.

The preparation of products of formula (VI) is described in particular in the following publications:

Zhur. Preklad. *Khim.* 28, 969-75 (1955) (CA 50, 4881a, 1956)

Tetrahedron 43, 1753 (1987)

J. Org. 52, 2407 (1987)

Zh. Org. Khim. 21, 2006 (1985)

J. Fluor. Chem. 17, 345 (1981) or in the Patents:

German Patent DRP 637,318 (1935)

European Patent EP 0,130,875

Japanese Patent JP 81,121,524.

The products of formula (VI) which are derivatives of hydantoin are widely used and mentioned in the literature such as for example in the following articles:

J. Pharm. Pharmacol., 67, Vol. 19(4), p. 209-16 (1967)

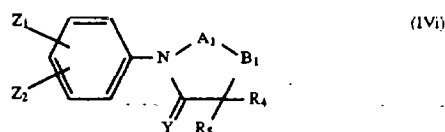
Khim. Farm. Zh., 67, Vol. 1(5) p. 51-2

German Patent 2,217,914

European Patent 0,091,596

J. Chem. Soc. Perkin. Trans. 1, p. 219-21 (1974).

Also a subject of the invention is, as new industrial products and notably as new industrial products which can be used as intermediates for the preparation of the products of formulae (I), (I_a) and the cited products as defined above, the products of formula (IVi):

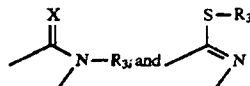


(IVi)

in which Z₁, Z₂, R₄, R₅ and Y have the meanings indicated above and the group:



is chosen from the radicals:



in which X represents an oxygen or sulphur atom and R₃ is chosen from the values of R₃ containing a protected reactive function, with the exception of the products in which R₄ and R₅, identical or different, represent a hydrogen atom or an alkyl radical having 1 to 12 carbon atoms optionally substituted by one or more halogen atoms.

Among the reactive functions which can be protected the hydroxyl and amino functions can be mentioned. These functions can be protected as indicated above for the substituent R₃.

The following examples illustrate the invention without however limiting it.

EXAMPLE 1

2-(trifluoromethyl) 4-(4-(hydroxymethyl) 4-methyl 2,5-dioxo 1-imidazolidinyl) benzonitrile

Stage A: 1-(tetrahydro 2H-pyran-2-yl) oxy 2-propanone
50 g of hydroxyacetone, 100 cm³ of methylene chloride and 0.5 g of 1% monohydrated paratoluene sulphonic acid are introduced together. Then over 5 hours at 20° C., 62.44 g of 3,4-dihydro 2-pyran is added. After 2 hours 30 minutes of introduction, 0.5 g of paratoluene sulphonic acid is added then the mixture is agitated for one hour 30 minutes and 100 cm³ of water saturated with sodium bicarbonate is added. Agitation is carried out for 5 minutes at an alkaline pH then the reaction medium is decanted and extraction is carried out with methylene chloride, then the extracts are washed with water, the organic phases are dried, filtered and brought to dryness. 101.8 g of expected product is obtained (pale yellow oil).

Stage B: 2-amino 2-methyl 3-((tetrahydro 2H-pyran 2-yl) oxy) propanenitrile

77.3 g of potassium cyanide, 178.1 g of alumina and 70 g of ammonium chloride in 1 litre of acetonitrile are agitated under ultrasound for one hour while maintaining the temperature at 40° C.

Next 95 g of the product obtained in Stage A) above is added, then the mixture is rinsed with 0.2 l of acetonitrile and agitated for about 21 hours.

Filtration is carried out, followed by rinsing with acetonitrile and drying. Purification is carried out on silica (eluant: cyclohexane—ethyl acetate 1—1) and 65.5 g of expected product (yellow oil) is collected.

IR Spectrum: CHCl_3

$\text{C}\equiv\text{N}$ 2230

NH 3393

3330

Stage C: 2-(trifluoromethyl) 4-(4-(hydroxymethyl) 4-methyl 2,5-dioxo 1-imidazolidinyl) benzonitrile

a) Condensation 2-(trifluoromethyl) 4-[5-imino 4-methyl 2-oxo 4-[(tetrahydro 2-H-pyran 2-yl) oxy] methyl] 1-imidazolidinyl] benzonitrile

12.77 g of the product obtained in Stage B above is introduced at 20° C. \pm 2° C. into 127.7 ml of methylene chloride. Then over about 1 hour 30 minutes, under agitation at -30° C. \pm 3° C., a previously-filtered solution of 11.4 g of the product obtained in the preparation of Example 7 of the European Patent Application EP 0,494,819 in 171 ml of methylene chloride is added and agitation is carried out for about one hour at -30° C. \pm 3° C. then the solvent is evaporated off under reduced pressure at 40° C. 24.7 g of the expected condensation product is obtained, used as it is for the methanolic hydrolysis.

b) Hydrolysis 2-(trifluoromethyl) 4-(4-(hydroxymethyl) 4-methyl 2,5-dioxo 1-imidazolidinyl) benzonitrile

21.3 g of the product obtained above in a is introduced under agitation at 20° C. \pm 2° C., into 213 ml of methanol. Then 67 ml of 2N hydrochloric acid is added over 2 minutes. The mixture is taken to reflux for one hour then left to cool under agitation. After concentration by distilling off about 100 ml of methanol, the reaction medium is placed under magnetic stirring for about one hour at a temperature of 0°/45° C. then separated.

The crystals obtained are purified, 3 volumes of methanol are added, followed by taking to reflux for 15 minutes, then leaving to cool down under agitation at 20°-25° C. and separating, 10.7 g of expected product (white crystals) is obtained. M.p.=218° C.

Microanalysis:

	Theoretical	Product dried at 60° C.
C %	49.85	49.7
N %	13.4	13.4
F %	18.19	18.0
H %	3.22	3.2

IR complex absorption OH/NH region

$\text{C}\equiv\text{N}$ approx. 2230 cm^{-1}

$\text{C}=\text{O}$ 1780-1735 cm^{-1}

Aromatics 1604-1575-1503 cm^{-1}

EXAMPLE 2

2-(trifluoromethyl) 4-(4-(hydroxymethyl) 3,

4dimethyl 2,5-dioxo 1-imidazolidinyl) benzonitrile

1) Formation of tetrahydropyranic ether

626 mg of the product of Example 1, 10 ml of tetrahydrofuran, 30 mg of paratoluene sulphonic acid, H_2O and 2 ml of dihydropyran are introduced together. At end of about 30 minutes, the mixture is poured into 10 ml of sodium bicarbonate and 1 ml of triethylamine and extracted with chloroform. The organic phase is washed with salt water, dried and the solvent is evaporated off. Purification is carried out on silica (eluant: CH_2Cl_2 , MeOH). 830 mg of the expected ether is obtained.

2) Methylation of the nitrogen

103 mg of 50% sodium hydride is introduced and 830 mg of the ether obtained above in 1) in 7 ml dimethylformamide is added over about 40 minutes, then 10 minutes after the cessation of the release of hydrogen, the reaction medium is placed in a water bath and 0.18 ml of methyl iodide and 0.5 ml of dimethylformamide are added. After reaction for 30 minutes, the medium is poured into 40 ml of water containing about 0.5 g of monopotassium phosphate and extracted with ether, then the organic phase is washed with salt water, dried and the solvent is evaporated off under reduced pressure. Purification is carried out on silica (eluant: CH_2Cl_2 — Me_2CO (95-5)). 770 mg of product is obtained, used as it is for the following stage.

3) Hydrolysis of the tetrahydropyranic ether

770 mg of the N-methylated ether obtained above in 2) is introduced into 10 ml of methanol, 1.5 ml of 2N hydrochloric acid and heated at about 40° C.

After 30 minutes, the mixture is taken to ambient temperature, poured into 20 ml of sodium bicarbonate, extraction is carried out with chloroform, the extracts are washed with salt water, dried and the solvent is evaporated off under reduced pressure. Purification is carried out on silica (eluant: CH_2Cl_2 — Me_2CO (3-1)). 111 mg of the crude product obtained above is recrystallized from 5 ml of hot isopropanol, concentrated to about 1 ml and ice-cooled for 16 hours. After filtration and drying, 90 mg of the expected product is, obtained (white crystals) M.p.=178°-179° C.

Microanalysis

	C	H	F	N
% calculated	51.38	3.70	17.41	12.84
% found	51.5	3.7	17.5	12.8

IR CHCl_3

OH 3620 cm^{-1}

$\text{C}=\text{O}$ 1781-1728 cm^{-1}

$\text{C}\equiv\text{N}$ 2235 cm^{-1}

Aromatics 1615-1576-1505 cm^{-1}

UV EtOH

Max 262 nm $\epsilon=13900$

Inf 278 nm $\epsilon=7200$

Inf 286 nm $\epsilon=3800$

EXAMPLE 3

4-(2,5-dioxo 3-(2-fluoroethyl) 4-(hydroxymethyl) 4-methyl 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

a) Alkylation with 1,2-bromofluoroethane

830 mg of the ether obtained in Stage 1) of Example 2 and 7.5 ml of dimethylsulphoxide are added drop by drop to 104

mg of 5% sodium hydride. About 20 minutes after cessation of the release of hydrogen, 0.22 ml of 1,2-bromofluoroethane is added in one lot. After about one hour of reaction, the medium is poured into 5 ml of water containing 500 mg of monopotassium phosphate and extracted with ether. The organic phase is washed with water then with salt water, dried and the solvent is evaporated off under reduced pressure. Purification is carried out on silica (eluant: CH_2Cl_2 - Me_2CO (97.5-2.5)) and 743 mg of expected 25 product is obtained.

b) Hydrolysis of the tetrahydropyranic ether

743 mg of the product obtained above in 1) is introduced into 10 ml of methanol, 1.5 ml of 2N hydrochloric acid and the mixture is taken to 40° C. then after 45 minutes is poured into 20 ml of sodium bicarbonate and extracted with chloroform. The organic phase is washed with salt water, dried and the solvent is evaporated off under reduced pressure. Purification is carried out on silica (eluant: CH_2Cl_2 - Me_2CO (9-1)) then the crystals obtained are dissolved in 20 ml of isopropanol at 60° C., followed by filtration, rinsing and concentrating to about 5 ml, ice-cooling for about one hour and separating, 466 mg of the expected product (white crystals) is obtained. M.p.=146°-147° C.

Microanalysis

	C	H	F	N
% calculated	50.15	3.65	21.15	11.70
% found	50.1	3.50	20.9	11.7

IR CHCl_3

OH 3612 cm^{-1}

>=O 1782 (m)-1727 (f) cm^{-1}

C=N 2235 cm^{-1}

UV EtOH

Max 260 nm $\epsilon=15500$

Inf 278 nm $\epsilon=6700$

Inf 286 nm $\epsilon=3300$

EXAMPLE 4

4-(3,4-dimethyl) 4-(hydroxymethyl) 5-oxo 2-thioxo 1-imidazolidiny] 2-(trifluoromethyl) benzonitrile
Stage A: (\pm) 2-methyl 2-(methylamino) 3-[(tetrahydro 2-H pyran 2-yl) oxy] propanenitrile

1.54 g of methylamine hydrochloride in solution in 10 cm^3 of water and 3.35 g of the ketone obtained in Stage A of Example 1 are introduced together and the suspension obtained is agitated for about 10 minutes.

1.06 g of NaCN in solution in 5 cm^3 of water is poured into the mixture and agitation is carried out overnight at ambient temperature.

Extraction is carried out with methylene chloride, the extracts are washed with a saturated solution of sodium chloride, dried and the solvent is evaporated off under reduced pressure. 3.72 g of expected product (yellow oil) is obtained, used as it is for the following stage.

IR CHCl_3

NH approx. 3345 cm^{-1}

C=N approx. 2230 cm^{-1} .

Stage B: 2-(trifluoromethyl) 4-[5-imino 3,4-dimethyl 4-[(tetrahydro 2-H-pyran 2-yl) oxy] methyl] 2-thioxo 1-imidazolidiny] benzonitrile

2.38 g of aminonitrile in solution in 8 cm^3 of 1,2-dichloroethane is introduced and 0.5 cm^3 of triethylamine is added, the mixture is cooled down to a temperature of -5° to 0° C. and 2.75 g of the isothiocyanate obtained in the

preparation of Example 11 of the European Patent Application EP 0.494.819 in solution in 17 cm^3 of 1,2-dichloroethane is poured in over 20 minutes at a temperature below 0° C. The reaction medium is left to return to ambient temperature and agitation is maintained for about 2 hours, followed by drying and evaporating the solvent under reduced pressure.

After purification on silica (eluant: CH_2Cl_2 -acetone (92-8)), 3.31 g of expected product is obtained.

10 IR CHCl_3

>C=NH 3308 cm^{-1}

-C=N 2236 cm^{-1}

>C=N 1679 cm^{-1}

>C=S 1614 cm^{-1}

15 Aromatic 1575 cm^{-1}

1505 cm^{-1}

1496 cm^{-1}

Stage C: 4-(3,4-dimethyl) 4-(hydroxymethyl) 5-oxo 2-thioxo 1-imidazolidiny] 2-(trifluoromethyl) benzonitrile

19 cm^3 of 2N hydrochloric acid is added drop by drop to 3.25 g of the product obtained in Stage B above, in solution in 35 cm^3 of methanol, and the whole is taken to reflux for 35 minutes.

Neutralization is carried out with a solution of sodium bicarbonate, followed by extraction with chloroform, the organic phase is washed with a saturated solution of sodium chloride, dried and the solvent is evaporated off under pressure. After purification on silica (eluant: CH_2Cl_2 -ACOEt (85-15)), then recrystallization from 10 cm^3 of isopropanol, 1.90 g of expected product (white crystals) is obtained. M.p.=167°-168° C.

Microanalysis

	C	H	N	F	S
% calculated	48.98	3.52	12.24	16.60	9.34
% found	48.9	3.6	12.1	16.7	9.1

40 IR CHCl_3

Absence of C=NH

-OH 3620 cm^{-1}

>C=O 1759 cm^{-1}

>C=N 2238 cm^{-1}

45 Aromatics 1610 cm^{-1}

+1576 cm^{-1}

conjugated 1505 cm^{-1}

system 1494 cm^{-1}

EXAMPLE 5

1,5-dimethyl 5-(hydroxymethyl) 3-(4-nitro 3-(trifluoromethyl) phenyl) 2,4-imidazolidinedione

Stage A: Formation of tetrahydropyranic ether

The operation is carried out as in 1) of Example 2 above, replacing in this preparation the product of Example 1 by 870 mg of the product obtained as in Example 2 of the European Patent Application EP 0.305.270 in 13 ml of tetrahydrofuran, 40 mg of paratoluene sulphonic acid, H_2O . 2.6 ml of dihydropyran. After about 15 minutes, the reaction mixture is poured into 10 ml of a saturated solution of sodium bicarbonate and 1 ml of triethylamine and extracted with chloroform. The organic phase is washed with salt water, dried, the solvent is evaporated off under reduced pressure and purification is carried out on silica (eluant: CH_2Cl_2 -MeOH (95-5)) and the expected product is obtained.

Stage B: Methylation of the nitrogen

The operation is carried out as in 2) of Example 2 above, starting from the product obtained in 1) above, and the expected product is obtained.

Stage C: Hydrolysis of the tetrahydropyranic ether.

The operation is carried out as in 3) of Example 2 above, starting from 955 mg of the product obtained in 2) above and 698 mg of expected product (white crystals) is obtained. M.p.=153°-154° C.

Microanalysis

	C	H	F	N
% calculated	44.96	3.48	16.41	12.10
% found	45.0	3.50	16.3	12.1

IR CHCl₃OH 3620 cm⁻¹>C=O 1782-1727 cm⁻¹Aromatic 1618-1596-1545-1498 cm⁻¹and NO₂ band

UV EtOH

Infl 214 nm ε=13000

Max 271 nm ε=6100

Infl 320 nm

EXAMPLE 6

4-(4-(fluoromethyl) 3,4-dimethyl) 2,5-dioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

0.2 ml of diethylaminosulphide trifluoride, then drop by drop the solution cooled down to -60° C. of 0.2 g of the product of Example 2 and 6.5 ml of tetrahydrofuran, are added to 1 ml of tetrahydrofuran, cooled down to about -60° C.

The resultant mixture is left to return to ambient temperature, then heated at 30° C. After one hour, the reaction medium is poured into 18 ml of sodium bicarbonate and extracted with ether. The organic phase is washed with salt water, dried and the solvents are evaporated off under reduced pressure. Purification is carried out on silica (eluant: CH₂Cl₂-cyclohexane (9-1)) then the crystals obtained are dissolved in 30 ml of isopropanol at 60° C., followed by filtration, rinsing with 2 ml of isopropanol, concentration to about 5 ml and ice-cooling overnight. After separation and drying, 136 mg of expected product (white crystals) is obtained. M.p.=153°-154° C.

Microanalysis

	C	H	F	N
% calculated	51.07	3.37	23.08	12.76
% found	50.8	3.2	25.0	12.7

IR CHCl₃C≡N 2240 cm⁻¹>C=O 1785-1733 cm⁻¹Aromatic 1616-1575-1505 cm⁻¹

UV EtOH

Max 259 nm ε=15200

Infl 278 nm ε=5800

Infl 286 nm ε=2900

EXAMPLE 7

4-(3,4-dimethyl) 4-(fluoromethyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

2 cm³ of anhydrous tetrahydrofuran is cooled down to -60° C. and drop by drop over about 15 minutes at a

temperature comprised between -60° C. and -53° C., 0.88 cm³ of diethylamino-sulphide trifluoride then 0.930 g of the product of Example 4 in solution in 7 cm³ of anhydrous tetrahydrofuran are poured in drop by drop. After the reaction medium has returned to ambient temperature, it is maintained for about 30 minutes at about 30° C., it is then poured into 25 cm³ of a solution of sodium bicarbonate and ice. Extraction is carried out with ether, the ethereal phase is washed with a saturated solution of sodium chloride, dried and the solvents are evaporated off under reduced pressure. After purification on silica (eluant: CH₂Cl₂-cyclohexane (9-1)) and recrystallization from isopropanol, 1.010 g of expected product (white crystals) is obtained after drying. M.p.=163° C.

Microanalysis

	C	H	F	N	S
20 % calculated	48.69	3.21	22.01	12.17	9.28
% found	48.6	3.10	22.2	12.1	9.5

IR CHCl₃

Absence of OH

25 N≡C approx. 2238 cm⁻¹>C=O 1762 cm⁻¹Conjugated syst.+1615-1580 cm⁻¹Aromatic 1505-1491 cm⁻¹

UV EtOH

30 Max 235 nm ε=19200

Max 253 nm ε=23000

Infl 265 nm ε=18300

EXAMPLE 8

4-(2,5-dioxo 3-(2-fluoroethyl) 4-(fluoromethyl) 4-methyl 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

1 ml of tetrahydrofuran is cooled down to -50° C. and 0.33 ml of diethylaminosulphide trifluoride is added then a solution cooled down beforehand to about -50° C. of 360 mg of the product of Example 3 and 4 ml of tetrahydrofuran is added drop by drop. Rinsing is carried out with 0.5 ml of tetrahydrofuran and the medium is left to return to ambient temperature then taken to about 30° C. It is then poured into 30 ml of sodium bicarbonate and 10 g of ice and extracted with chloroform then the organic phase is washed with salt water and dried. Purification is carried out on silica (eluant: CH₂Cl₂—ethyl acetate (99-1)) then the crystals obtained are dissolved in 30 ml of isopropanol under reflux, followed by filtration, rinsing with 1 ml of isopropanol, concentration to about 7 ml and leaving at rest for 16 hours at 0° C. After separation and drying, 307 mg of expected product (white crystals) is obtained. M.p.=137°-138° C.

Microanalysis

	C	H	F	N
60 % calculated	49.87	3.35	26.29	11.63
% found	49.8	3.4	26.2	11.6

IR CHCl₃C≡N 2235 cm⁻¹65 >C=O 1786-1730 cm⁻¹Aromatic 1616-1575-1505 cm⁻¹

UV EtOH

Max 258 nm $\epsilon=16000$
 Infl 277 nm $\epsilon=5300$
 Infl 285 nm $\epsilon=2700$

EXAMPLE 9

4-(2,4-dioxo 1,3-diazaspiro(4,4)nonan 3-yl) 2-(trifluoromethyl) benzonitrile

Stage A: 1-amino cyclopentanecarbonitrile

40 ml of ammonium hydroxide, 7.9 g of ammonium chloride and 6.14 g of sodium cyanide are introduced together then agitation is carried out until total dissolution in an ice bath at a temperature of -8°C . Then 8.8 ml of cyclopentanone is added drop by drop at a temperature of about -9°C . the reaction medium is left to return to ambient temperature and agitated overnight.

The organic phase is then decanted, the aqueous phase is extracted with methylene chloride, then the organic phases are washed with salt water and dried. After distillation at $55^{\circ}\text{C} \pm 2^{\circ}\text{C}$, 1.2 g of expected product is obtained.

IR CHCl_3

NH_2 3381-3330 cm^{-1}

$\text{C}\equiv\text{N}$ 2226 cm^{-1}

Absence $\text{C}=\text{O}$

Stage B: 2-(trifluoromethyl) 4-[4-imino 2-oxo 1,3-diazaspiro[4.4]nonan 3-yl] benzonitrile

550 mL of the product obtained in Stage A above, 4 ml of 1,2-dichloroethane and 0.2 ml of triethylamine are introduced together then the mixture is brought to 0°C , and 3.1 ml of the product obtained in the preparation of Example 7 of the European Patent Application EP 0,494,819 is added over 5 minutes at a temperature of -4°C , and the whole is left to return to ambient temperature.

After about 40 minutes of reaction the medium is concentrated to dryness, the residue is dissolved in 40 ml of acetone, the solvent is evaporated off and purification is carried out on silica (eluant: CH_2Cl_2 - Me_2CO (90-10)), 1.24 g of expected product (white crystals) is obtained. M.p.= 212° - 213°C .

IR nujol

OH/NH 3350-3280 cm^{-1}

$\text{CH}\equiv$ 2240 cm^{-1}

$\text{C}=\text{O}$ 1744 cm^{-1}

>N 1670 cm^{-1}

Aromatic 1610-1574-1510 cm^{-1}

Stage C: 4-(2,4-dioxo 1,3-diazaspiro(4,4)nonan 3-yl) 2-(trifluoromethyl) benzonitrile

1.17 g of the product obtained in Stage B above in 20 ml of methanol, 3 ml of chloroform and 5 ml of 2N hydrochloric acid are introduced together then taken to about 50°C , for about 2 hours.

The mixture is taken to ambient temperature, poured into 40 ml of water and extracted 3 times with methylene chloride. The organic phase is washed with salt water, dried and the solvent is evaporated off under reduced pressure, then purification is carried out on silica (eluant: CH_2Cl_2 - Me_2CO (9-1)), 1.108 g of expected product (white crystals) is obtained. M.p.= 184° - 185°C .

Microanalysis

	C	H	F	N
% calculated	55.73	3.74	17.63	13.00
% found	55.6	3.7	17.4	12.8

IR CHCl_3

$=\text{C}-\text{NH}$ 3444 cm^{-1}

$\text{C}\equiv\text{N}$ 2236 cm^{-1}

>O 1786-1731 cm^{-1}

Aromatic 1616-1505 cm^{-1}

UV EtOH

5 Max 258 nm $\epsilon=15600$

Infl 286 nm $\epsilon=3500$

EXAMPLE 10

4-(2,4-dioxo 1-(2-fluoroethyl) 1,3-diazaspiro(4,4)nonan-3-yl) 2-(trifluoromethyl) benzonitrile

0.323 g of the product of Example 9 in solution in 2.5 cm^3 of dimethylsulphoxide is poured drop by drop over about 20 minutes into 0.050 g of sodium hydride at 50°C in oil. Agitation is carried out for about one hour 20 minutes then 0.09 cm^3 of 1-bromo 2-fluoroethane in solution in 0.2 cm^3 of dimethylsulphoxide is poured in drop by drop. After about 2 hours at ambient temperature, the reaction medium is heated for 10 minutes at between 30° and 35°C , then poured into 12 cm^3 of water containing 0.2 g of monosodium phosphate and extracted with ether. The ethereal phase is washed with a saturated solution of sodium chloride, dried and evaporated to dryness. Purification is carried out on silica (eluant: CH_2Cl_2 -ethyl acetate (99-1)) and 0.249 g of expected product is obtained. M.p.= 115°C .

Microanalysis

	C	H	F	N
% calculated	55.29	4.09	20.58	11.38
% found	55.3	4.1	20.25	11.3

IR CHCl_3

$-\text{C}=\text{N}$ 2238 cm^{-1}

$-\text{C}=\text{O}$ 1776-1723 cm^{-1}

Aromatic 1616-1575-1505 cm^{-1}

UV EtOH

Max 260 nm $\epsilon=15600$

Infl 287 nm

EXAMPLE 11

1,5-dimethyl 5-(fluoromethyl) 3-(4-nitro 3-(trifluoromethyl) phenyl) 2,4-imidazolidinedione

0.09 ml of diethylaminosulphide trifluoride is added to 1 ml of tetrahydrofuran, cooled down to about -60°C , and drop by drop a solution cooled down to about -60°C , of 210 mg of the product of Example 5 and 2.5 ml of tetrahydrofuran is added. The mixture is rinsed with 0.5 ml of tetrahydrofuran, left to return to ambient temperature, taken to a temperature of about -65°C , and 0.1 ml of diethylaminosulphide trifluoride is added.

After one hour 20 minutes, the reaction medium is poured into 8 ml of sodium bicarbonate and extracted with ether. The organic phase is washed with salt water, dried, the solvent is evaporated off under reduced pressure and purification is carried out on silica with CH_2Cl_2 - Me_2CO (99-1) as eluant. 152 mg of expected product (white crystals) is obtained. M.p.= 118° - 119°C .

Microanalysis				
	C	H	F	N
% calculated	44.71	3.17	21.76	12.03
% found	44.9	3.1	21.42	11.9

IR CHCl_3
 —C=O 1786–1732 cm^{-1}
 Aromatic and 1618–1597–1546–1498 cm^{-1}
 UV EtOH
 Infl 214 nm $\epsilon=13400$
 Max 267 nm $\epsilon=6200$
 Infl 320 nm

EXAMPLE 12

4-(4.4-dimethyl-2.5-dioxo-3-(2-(2-hydroxyethoxy) ethyl)-1-imidazolidinyl-2-(trifluoromethyl)-benzoxonitrile

Stage 1: 4-(4.4-dimethyl-2.5-dioxo-3-(2-(2-(((1.1-dimethyl-ethyl) dimethylsilyl) oxy) ethoxy) ethyl)-1-imidazolidinyl-2-(trifluoromethyl)-benzoxonitrile

The solution of 0.594 g of the product of Example 8 of EP 0.494.819 and 4.5 ml of dimethylsulphoxide are poured drop by drop over 15 minutes into 0.101 g of sodium hydride at 50% in oil and agitation is carried out for 30 minutes after cessation of the release of hydrogen.

0.594 g of (2-(2-bromoethoxy) ethoxy)-dimethyl-(1.1-dimethylethyl) silane in solution in 1 ml of dimethylsulphoxide is poured in drop by drop over 5 minutes.

Agitation is carried out for one hour at ambient temperature then 0.056 g of (2-(2-bromoethoxy) ethoxy)-dimethyl-(1.1-dimethyl-ethyl) silane is added and the whole is maintained for 2 hours 30 minutes between 30° C. and 40° C.

The mixture is poured into 0.6 g of monosodium phosphate and 30 ml of ice-cooled water and extracted with ether.

The ethereal phase is washed with a solution of NaCl, dried and the solvent is evaporated off.

After chromatography on silica (eluant: methylene chloride/ethyl acetate 99/1), 0.776 g of expected product (oil) is isolated.

IR CHCl_3 , cm^{-1}
 Absence of =C—NH
 —CN approx. 2235



Aromatic 1616–1577–1505

Stage 2: 4-(4.4-dimethyl-2.5-dioxo-3-(2-(2-hydroxyethoxy) ethyl)-1-imidazolidinyl)-2-(trifluoromethyl)-benzoxonitrile

0.759 g of the product obtained in Stage 1 above is introduced into 7.5 ml of methanol and 2 ml of 2N hydrochloric acid is added drop by drop.

The mixture is agitated for 40 minutes at ambient temperature, poured into ice-cooled water and extracted with chloroform.

The organic phase is washed with a saturated solution of sodium chloride and dried; the solvent is evaporated off then chromatography is carried out on silica (eluant: methylene

chloride/acetone 9/1), and 0.549 g of expected product is isolated. M.p.<60° C.

ANALYSES

5 IR CHCl_3 , (cm^{-1})
 Complex —OH towards 3610–3620
 —CN approx. 2235
 C=O 1779–1725
 Aromatics 1616–1575–1505

EXAMPLE 13

4-(4.4-dimethyl 2.5-dioxo 3-(2-fluoroethyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

15 The solution of 600 mg of the product of Example 8 of the European Patent Application EP 0.494.819 and 5 ml of dimethylformamide is added drop by drop over about 20 minutes to 104 ml of 50% sodium hydride. The mixture is rinsed with 0.5 ml of dimethylformamide and after cessation of the release of hydrogen, 0.16 ml of 1-bromo 2-fluoroethane is added. A further 98 ml of 50% sodium hydride is then added and after approximately 10 minutes, 0.1 ml of 1-bromo 2-fluoroethane is added and the whole is heated to 50° C. It is returned to ambient temperature. 25 poured into 20 ml of water containing 200 mg of monopotassium phosphate and extracted with ether. The extracts are washed with water then with salt water, dried, the solvent is evaporated off under reduced pressure, followed by dissolution in 20 ml of Me_2CO and purification on silica (eluant CH_2Cl_2 —ethyl acetate (99–1)). 200 mg of expected product (white crystals) is obtained. M.p.=10° C. 8–109° C.

Microanalysis

	C	H	F	N
% calculated	52.48	3.82	22.14	12.24
% found	52.4	3.7	22.00	12.4

40 IR CHCl_3
 $\text{C}\equiv\text{N}$ 2236 cm^{-1}
 C=O 1780–1728 cm^{-1}
 Aromatic 1618–1580–1504 cm^{-1}

EXAMPLE 14

4-(4.4-dimethyl 2.5-dioxo 3-(2.2.2-trifluoro-ethyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

50 0.742 g of the product of Example 8 of the European Patent Application EP 0.494.819 in solution in 7.5 cm^3 of dimethylsulphoxide is poured over 20 minutes into 0.125 g of sodium hydride at 50% in oil and the resultant mixture is rinsed with 1 cm^3 of dimethylsulphoxide. After the release of hydrogen is complete, agitation is maintained for about 20 minutes and 0.5 cm^3 of iodotrifluoromethane is poured in and 0.5 cm^3 of 15-crown-5 ether is added. The mixture is maintained at 60° C. for 16 hours. After addition of 0.25 cm^3 of iodotrifluoromethane, heating is continued for about 19 hours at 80° C. The reaction medium is then poured into 30 cm^3 of water +0.5 g of monosodium phosphate and extracted with ether.

The ethereal phase is washed with a saturated solution of sodium chloride, dried and evaporated to dryness. Purification is carried out on silica (eluant: CH_2Cl_2 —ethyl acetate (99–1)), followed by recrystallization from 10 cm^3 of isopropanol and 0.262 g of expected product (white crystals) is obtained. M.p.=110° C.

	Microanalysis			
	C	H	F	N
% calculated	47.5	2.92	30.05	11.08
% found	47.3	2.8	30.0	11.0

IR CHCl_3
Absence of N—H
>C=O 1790–1732 cm^{-1}
Aromatic 1616–1578–1505 cm^{-1}
C≡N 2235 cm^{-1}
UV EtOH
Max 255 nm $\epsilon=16400$
Infr 276 nm $\epsilon=4400$
Infr 285 nm $\epsilon=2400$

EXAMPLE 15

4-(4,4-dimethyl 3-(2-fluoroethyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile
Stage A: 4-[3-(2-fluoroethyl) 5-imino 4,4-dimethyl 2-thioxo 1-imidazolidinyl] 2-(trifluoromethyl) benzonitrile

1.86 g of 2-[(2-fluoroethyl) amino] 2-methyl propanenitrile and 0.55 cm^3 of triethylamine are introduced into 12 cm^3 of 1,2-dichloroethane. The mixture is cooled down to 0° C. and 2.97 g of the isothiocyanate obtained as in the preparation of Example 11 of the European Patent Application EP 0,494,819 in solution in 22 cm^3 of 1,2-dichloroethane is poured in over about 25 minutes at a temperature of about 0° C. Agitation is continued for 7 hours at ambient temperature then the solvent is evaporated off under reduced pressure. Purification is carried out on silica (eluant: CH_2Cl_2 —acetone (95–5) then (97–3)), the residue is taken up in a few cm^3 of ether, followed by separating, drying, and 1.84 g of expected product is obtained. M.p.=160° C.

IR CHCl_3
C=NH 3308 cm^{-1}
C≡N 2236 cm^{-1}
Conjugated system 1677 cm^{-1}
+1604 cm^{-1}
aromatics 1576–1504 cm^{-1}

Stage B: 4-(4,4-dimethyl 3-(2-fluoroethyl) 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

6.9 cm^3 of 2N hydrochloric acid is added drop by drop to 1.65 g of the product obtained in Stage A above, in solution in 60 cm^3 of methanol. After about one hour 30 minutes at 50° C., the mixture is neutralized with sodium bicarbonate and the methanol is distilled off. The residue is taken up in water, followed by extraction with ether, the ethereal phase is washed with a solution of sodium chloride and dried. After purification on silica with CH_2Cl_2 as eluant and recrystallization from 15 cm^3 of isopropanol, 2.99 g of expected product (white crystals) is obtained. M.p.=135° C.

	Microanalysis				
	C	H	F	N	S
% calculated	50.14	3.65	21.15	11.69	8.92
% found	50.1	3.60	21.0	11.7	9.2

IR CHCl_3
Absence of =C—NH
>C=O 1758 cm^{-1}

N≡C 2236 cm^{-1}
Conjugated syst. 1610 cm^{-1}
+1576 cm^{-1}
Aromatic 1504 cm^{-1}
UV EtOH
Max 233 nm $\epsilon=17700$
Max 253 nm $\epsilon=21600$

EXAMPLE 16

3-(4-cyano 3-(trifluoromethyl) phenyl) 2,4-dioxo 5-(hydroxymethyl) 5-methyl 1-imidazolidinacetoneitrile

The operation is carried out as in Example 2, starting with the product of Example 1, replacing the iodomethyl in 2) of Example 2 with bromocyanomethyl and in this way the expected product is obtained. M.p.=171° C.

EXAMPLE 17

3-(4-cyano 3-(trifluoromethyl) phenyl) 2,4-dioxo 5-(fluoromethyl) 5-methyl 1-imidazolidinacetoneitrile

The operation is carried out as for the preparation of Example 6, replacing the product of Example 2 with the product of Example 16 and in this way the expected product is obtained. M.p.=175° C.

EXAMPLE 18

(±) 4-[2-oxo 5-imino [(4-fluorophenyl) thio] methyl] 4-methyl 1-imidazolidinyl 2-trifluoromethyl benzonitrile

Stage A: (±) 3-[(4-fluorophenyl) thio] 2-amino 2-methyl propane nitrile

A solution of 4.81 g of ammonium chloride in 12 ml of water and 24.6 ml of 25° C. Be ammonium hydroxide then a solution of 8.32 g of 1-[4-fluorophenylthio] 2-propanone prepared as indicated in the Patent 82 46399 E. in 21 ml of ethanol at 96.2%, are added successively to 2.21 g of sodium cyanide in 45 ml of water. The resultant mixture is maintained at 60° C. and agitated for about 22 hours. It is cooled down to 0° C./+4° C., rinsed with ethanol, distilled, decanted, the aqueous phase is extracted with CH_2Cl_2 , washed with a saturated solution of sodium chloride, dried and the solvent is evaporated off under reduced pressure. Purification is carried out by chromatography on silica with cyclohexane—ethyl acetate (50—50) as eluant and 14.50 g of expected product is obtained.

IR CHCl_3
Absorption NM 3382–3327 cm^{-1}
Aromatic 1591–1491 cm^{-1}
—C≡N 2228 cm^{-1}

Stage B: (±) 4-[2-oxo 5-imino [(4-fluorophenyl) thio] methyl] 4-methyl 1-imidazolidinyl 2-trifluoromethyl benzonitrile

9.8 g of the product obtained in Stage A above and 35 ml of 1,2-dichloroethane are mixed together, and 0.2 ml of triethylamine is added. The mixture is cooled down to 5° C. and the solution of 7.7 g of the product obtained in the preparation of Example 7 of the European Patent Application EP 0,494,819 and 40 ml of 1,2-dichloroethane is introduced over 12 minutes at a temperature between 5° C. and 10° C.

The reaction medium is rinsed with 5 ml of dichloroethane and left for 16 hours at ambient temperature. The solvent is evaporated off under reduced pressure, the residue is purified on silica (eluant: CH_2Cl_2 — Me_2CO (93–7)), then

dissolved in 100 ml of isopropanol at about 60° C., followed by filtration, rinsing with 20 ml of hot isopropanol, concentration, ice-cooling for about 3 hours, separation, rinsing with ice-cooled isopropanol and drying. 2.815 g of expected product (white crystals) is obtained. M.p.=150° C.

Microanalysis					
	C	H	F	N	S
% calculated	54.03	3.34	17.99	13.26	7.59
% found	54.1	3.30	17.9	13.1	7.7

IR CHCl_3
 C=O 1756 cm^{-1}
 C=N 1669 cm^{-1}
 NH 3444 cm^{-1}
 Aromatic 1614–1591–1505–1491 cm^{-1}
 C=N exists

EXAMPLE 19

4-[2,5-dioxo 4-[(4-fluorophenyl) thio] methyl] 4-methyl 1-imidazolidinyl 2-trifluoromethyl benzonitrile

3.95 g of the product obtained in Example 18. 14 ml of 22° C. Be hydrochloric acid and 14 ml of water are introduced together and the suspension is heated under reflux.

After about one hour 30 minutes, the suspension is returned to ambient temperature, poured over ice+water 100 g (1—1) and extracted with ethyl acetate. The organic phase is washed with water, then with a saturated solution of sodium bicarbonate, and finally with a saturated solution of sodium chloride and the solvent is evaporated off. Purification is carried out by chromatography on silica (eluant: CH_2Cl_2 — Me_2CO (95—5)). The residue is taken up in 50 ml of ethanol 100 at 50° C., followed by filtration, rinsing with 5 ml of hot ethanol, concentration and leaving for 16 hours in a refrigerator at about 0° C. to +4° C.

After separation, the product is rinsed with ice-cooled ethanol and dried. 3.55 g of expected product (white crystals) is obtained. M.p.=153° C.

Microanalysis					
	C	H	F	N	S
% calculated	53.9	3.09	17.95	9.92	7.57
% found	53.9	3.1	18.3	9.8	7.8

IR CHCl_3
 C=O 1791–1734 cm^{-1}
 NH 3439 cm^{-1}
 C=N 2236 cm^{-1}
 Aromatics 1615–1591–1505–1492 cm^{-1}

EXAMPLE 20

4-(2,5-dioxo 3-(2-fluoroethyl) 4-((4-fluoro-phenyl) thiomethyl) 4-methyl 1-imidazolidinyl 2-(trifluoromethyl) benzonitrile

The solution of 0.254 g of the product obtained as in Example 19 in 2.2 cm^3 of dimethylsulphoxide is poured drop by drop over about 10 minutes onto 0.031 g of sodium hydride at 50% in oil. Agitation is maintained for about 40 minutes. Then the solution of 0.54 cm^3 of 1-bromo

2-fluoroethane in 0.7 cm^3 of dimethyl-sulphoxide is added drop by drop over about 5 minutes.

After agitation for 30 minutes, the reaction medium is poured onto 0.4 g of monosodium phosphate, water+ice. Extraction is carried out with ether, the organic phase is washed with a saturated solution of sodium chloride, dried and the solvent is evaporated off under reduced pressure. After purification on silica (eluant: CH_2Cl_2 —ethyl acetate (100—0.5)) then recrystallization from 15 cm^3 of isopropanol. 0.175 g of expected product (white crystals) is obtained. M.p.=155° C.

Microanalysis					
	C	H	F	N	S
% calculated	53.73	3.43	20.23	8.95	6.83
% found	53.5	3.2	20.5	9.0	7.1

IR CHCl_3
 Absence of =C—NH
 C=N approx. 2235 cm^{-1}
 >C=O 1780–1727 cm^{-1}
 Aromatics 1616–1591–1505–1492 cm^{-1}
 UV EtOH
 Max 255 nm $\epsilon=18600$
 Inf 278 nm $\epsilon=8000$
 Inf 287 nm $\epsilon=4400$

EXAMPLE 21

4-(4,4-bis(hydroxymethyl) 3-methyl 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile
 Stage A: 2-(methylamino) 2-[[[(tetrahydro 2-H-pyran 2-yl) oxy] methyl] 3-(tetrahydro 2H-pyran 2-yl) oxy] propanenitrile

2.7 g of 1,3-bis[(tetrahydro 2H-pyran-2-yl) oxy] 2-propanone obtained as below. 5 ml of water and 0.77 g of methylamine hydrochloride are introduced together then 503 mg of sodium cyanide and 3 ml of water are added over 5 minutes. After 2 hours 30 minutes of reaction, extraction is carried out with methylene chloride, the organic phase is washed with salt water, the solvent is evaporated off under reduced pressure. 3.3 g of expected product is obtained, used as it is for the following stage.

IR CHCl_3
 NH 3346 cm^{-1}
 C=N 2230 cm^{-1}
 C=O 1732 cm^{-1}
 Preparation of 1,3-bis-[(tetrahydro 2H-pyran 2-yl) oxy] 2-propanone used at the start of Example 21

9 g of 1,3-dihydroxyacetone dimer in suspension in 60 ml of dioxane is heated to 70° C. and returned to ambient temperature. 20 ml of 3,4-dihydro 2,4-pyran then 300 mg of paratoluene sulphonic acid and H_2O are added, while maintaining the temperature below 40° C. The reaction medium is maintained for 16 hours under agitation, poured into 300 ml of a saturated solution of sodium bicarbonate, the organic phase is washed with salt water, dried and the solvent is evaporated off under reduced pressure. After chromatographing the residue on silica (eluant: cyclohexane—ethyl acetate—triethylamine (8—2—0.5)), 17 g of expected product is obtained. RF=0.2.

Stage B: 2-(trifluoromethyl) 4-[4,4-bis-[(tetrahydro 2-H-pyran 2-yl) oxy] methyl] 5-imino 3-methyl 2-thioxo 1-imidazolidinyl] benzonitrile

2.39 cl of the isothiocyanate obtained in the preparation of Example 11 of the European Patent Application EP 0.494.

819 and 10 ml of 1,2-dichloroethane are mixed together. Then 3.2 g of the product obtained in Stage A above, 0.4 ml of triethylamine and 10 ml of 1,2-dichloroethane are added drop by drop to the solution cooled down to +5° C. After heating for about one hour 20 minutes, the solvent is evaporated off and purification is carried out on silica (eluant: ethyl acetate 7 cyclohexane 3). 3.82 g of the expected product is obtained.

IR CHCl_3
 $>\text{C}=\text{NH}$ 3314 cm^{-1}
 $\text{C}\equiv\text{N}$ 2230 cm^{-1}
 $\text{C}=\text{N}$ 1678-1670-1876
 $\text{C}=\text{S}$ 1505-1495 cm^{-1}

Aromatics

Stage C: 4-(4,4-bis(hydroxymethyl) 3-methyl 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

3.8 g of the product obtained in Stage B above is introduced into 38 ml of methanol and 19 ml of 2N hydrochloric acid, then the mixture is heated under reflux. After about 2 hours, it is poured into 200 ml of water and extracted with ether then with ethyl acetate. The organic phases are united and washed with salt water then the solvent is evaporated off. Purification is carried out on silica (eluant: CH_2Cl_2 -MeOH (95-5)) then the product is dissolved in 30 ml of isopropyl ether under reflux, followed by filtration and partial concentration. After ice-cooling for about one hour and separating, 282 mg of expected product (yellow crystals) is obtained. M.p.=169° C.-170° C.

Microanalysis

	C	H	F	N	S
% calculated	46.80	3.37	15.86	11.69	8.92
% found	46.8	3.3	15.9	11.5	9.0

IR Nujol
 OH/NH 3410-3385 cm^{-1}
 $\text{C}\equiv\text{N}$ 2240 cm^{-1}
 $\text{C}=\text{O}$ 1720 cm^{-1}
 Aromatics 1608-1580-1568 cm^{-1}
 UV EtOH
 Max 234 nm $\epsilon=17600$
 Max 256 nm $\epsilon=23200$

EXAMPLE 22

4-(4,4-bis(1-oxopropoxy) methyl) 3-methyl 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

200 mg of the product of Example 21 is introduced into 2 ml of pyridine and 25 mg of 4-dimethylamino pyridine then 16 ml of propionic anhydride is added. After reaction for 25 minutes, the mixture is poured into 20 ml of sodium bicarbonate and extracted with methylene chloride, the organic phase is washed with salt water, dried, the solvent is evaporated off and distillation is carried out 3 times with 30 ml of toluene. Purification is carried out on silica (eluant: Cl_2Cl_2), then crystallization from ether and 239 mg of expected product (white crystals) is obtained. M.p.=117° C.-118° C.

Microanalysis

	C	H	F	N	S
% calculated	50.95	4.27	12.09	8.91	6.80
% found	51.2	4.5	12.1	8.8	6.9

IR CHCl_3
 $>\text{C}=\text{O}$ 1755-1762 cm^{-1}
 $\text{C}\equiv\text{N}$ 2235 cm^{-1}
 Aromatics and $\text{C}=\text{S}$ 1615-1580-1504-1488 cm^{-1}
 UV EtOH
 Infl 236 nm $\epsilon=18800$
 Max 253 nm $\epsilon=22600$
 Infl 265 nm $\epsilon=18200$

EXAMPLE 23

4-(4,4-bis(fluoromethyl) 3-methyl 5-oxo 2-thioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

1 ml of tetrahydrofuran is cooled down to -50° C. and 0.66 ml of diethylaminosulphide trifluoride is added then the solution, cooled down to a temperature of about -50° C., of 360 mg of the product of Example 21 in 4 ml of tetrahydrofuran is added drop by drop. The resultant mixture is heated to a temperature of about 30° C.

After about 30 minutes, the resultant mixture is poured drop by drop into 50 ml of a saturated aqueous solution of sodium bicarbonate, extraction is carried out with chloroform, the organic phase is washed with salt water and the solvent is evaporated off. Purification is carried out on silica with CH_2Cl_2 -cyclohexane (9-1) as eluant then the product is dissolved in 20 ml of isopropanol at a temperature of about 60° C. Filtration is carried out, followed by rinsing with 1 ml of isopropanol and concentration, ice-cooling for one hour and separating, 314 mg of expected product (white crystals) is obtained. M.p.=122° C.-123° C.

Microanalysis

	C	H	F	N	S
% calculated	46.28	2.77	26.15	11.56	8.82
% found	46.3	2.7	25.7	11.25	8.8

IR CHCl_3
 $\text{C}\equiv\text{N}$ 2236 cm^{-1}
 $\text{C}=\text{O}$ 1763 cm^{-1}
 Aromatics 1615-1580-1504-1487 cm^{-1}
 UV EtOH
 Infl 237 nm $\epsilon=20300$
 Max 250 nm $\epsilon=22000$
 Infl 266 nm $\epsilon=17100$

EXAMPLE 24

4-(4,4-bis(hydroxymethyl) 3-methyl 2,5-dioxo 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

1) Formation of the tetrahydropyranic ethers
 360 mg of the product of Example 21 is introduced into 5 ml of tetrahydrofuran, 1 ml of 3,4-dihydro 2H pyran, and 15 mg of paratoluene sulphonic acid, H_2O .

After about 40 minutes, 10 ml of a saturated aqueous solution of sodium bicarbonate and 1 ml of triethylamine are poured into the mixture, extraction is carried out with chloroform, the extracts are washed with salt water, dried and the solvent is evaporated off.

Purification is carried out on silica (eluant: CH_2Cl_2 — MeOH (93-7)) and 600 mg of expected product is obtained, used as it is for the following stage.

2) Transition to hydantoin

600 mg of the diether obtained in 1) is introduced into 4 ml of dimethylformamide and 55 mg of 50% sodium hydride is added, then after cessation of the release of hydrogen, 0.09 ml of methyl iodide is added. About 40 minutes afterwards, 110 mg of 50% sodium hydride, then 10 minutes afterwards 0.18 ml of methyl iodide are successively added. The reaction mixture is poured into 10 ml of ice-cooled water containing 1.3 g of monopotassium phosphate and extraction is carried out with ether. The organic phase is washed with salt water, dried and the solvent is evaporated off. Purification is carried out on silica (eluant: CH_2Cl_2 —ethyl acetate (92.5-7.5)) and 370 mg of expected product is obtained, used as it is for the following stage.

3) Deprotection of the pyranic ethers

370 mg of the product obtained above in 2) is introduced into 4 ml of methanol and 2 ml of 2N hydrochloric acid then the solution is taken to 60° C. for about 2 hours.

The solution is then poured into 15 ml of salt water, drying is carried out, the solvent is evaporated off then the residue is dissolved in 20 ml of acetone and evaporated to dryness. Purification is carried out on silica (eluant: CH_2Cl_2 — MeOH (92.5-7.5)) followed by recrystallization from acetone and 197 mg of expected product (white crystals) is obtained, M.p.=217° C.—218° C.

UV EtOH

Max 263 nm $\epsilon=14600$

Infl 237, 278, 287 nm

EXAMPLE 25

4-(4,4-bis(hydroxymethyl) 3-methyl 5-imino
2thioxo 1-imidazolidinyl) 2-(trifluoromethyl)
benzonitrile

The operation is carried out as in Example 21 and at Stage C of the preparation of Example 21, 421 mg of expected product is obtained.

IR

$\text{C}\equiv\text{N}$ 2230 cm^{-1}

$\text{C}=\text{N}$, $\text{C}=\text{S}$ 1680-1614-1580-1510 cm^{-1}

Aromatics

EXAMPLE 26

4-(4,4-bis(hydroxymethyl)-2,5-dioxo-1-
imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

Stage 1: 1.3 bis [(tetrahydro 2H pyran-2 yl) oxy] 2 propanone

9 g of 2,5-dihydroxy 1,4-dioxane 2,5-dimethanol is introduced into 60 ml of dioxane and the suspension is taken to about 70° C. for 15 minutes then returned to ambient temperature. 20 ml of 3,4-dihydro 2H-pyran and 300 mg of monohydrated paratoluene sulphonic acid are then added and the temperature is maintained at about 40° C. then the medium is left for 16 hours at ambient temperature.

The medium is then poured into a mixture of 300 ml of a saturated solution of sodium bicarbonate +10 ml of triethylamine and extraction is carried out with methylene chloride. The organic phase is washed with salt water, dried and the solvent is evaporated off.

After chromatography on silica (eluant: ethyl cycloacetate/triethylamine 8/2), 17 g of expected product is obtained (pale yellow syrup).

ANALYSES:

IR CHCl_3 (cm^{-1})

Absence of OH

$\text{O}=\text{C}$ 1736

Stage 2: 2-amino 3-((tetrahydro-2H-pyran-2-yl) oxy) 2((
tetrahydro-2H-pyran-2-yl) oxy) methyl) propanenitrile

5.6 g of the product obtained in Stage 1 above is introduced into 8 ml of ammonium hydroxide, the mixture is taken to about -5° C. and 1.58 g of ammonium chloride and 1.23 g of sodium cyanide are added successively and the resultant mixture is left to rise to ambient temperature over about 40 minutes then heated at 40° C.±5° C. under agitation for 16 hours. It is returned to ambient temperature and extracted with chloroform, the organic phase is washed with salt water, dried and the solvent is evaporated off.

After chromatography on silica (eluant: ethyl cycloacetate/triethylamine 3/7), 4.41 g of expected product (pale yellow syrup) is obtained.

ANALYSES:

IR CHCl_3 (cm^{-1})

—CN 2235

NH2 3390-3317

Stage 3: 4-(5-imino-2-oxo-4,4-bis((tetrahydro-2H-pyran-2-yl) oxy) methyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

570 mg of the product obtained in Stage 2 above is introduced into 5 ml of isopropyl ether and 0.28 ml of triethylamine and the mixture is taken to -30° C. then 2.32 g of a solution of the product obtained in the preparation of Example 7 of the European Patent Application EP 0,494,819 at 18.4% in 1,2-dichloroethane is added over one hour.

4 ml of methylene chloride is added then the reaction medium is left to return to ambient temperature, left for about 2 hours and the solvent is evaporated off. After purification on silica (eluant: methylene chloride/acetone 9/1), 700 mg of expected product is obtained.

ANALYSES:

IR CHCl_3 (cm^{-1})

NH 3442-3317

—CN 2235

$\text{C}=\text{O}$ 1757

$\text{C}=\text{N}$ 1670

Aromatic 1614-1575-1505

Stage 4: 4-(4,4-bis(hydroxymethyl)-2,5-dioxo-1-
imidazolidinyl)-2-(trifluoromethyl)-benzonitrile

300 mg of the product obtained in Stage 3 above is introduced into 3 ml of methanol and 1.5 ml of 2N hydrochloric acid and the mixture is taken to reflux for one hour 30 minutes.

It is returned to ambient temperature, poured into 5 ml of an aqueous solution of sodium bicarbonate, extracted with ethyl acetate then the extracts are washed with a saturated aqueous solution of sodium chloride, dried and the solvent is evaporated off.

5 ml of methanol is added and purification is carried out on silica (eluant: methylene chloride—methanol 9/1).

The product is taken up in 20 ml of isopropanol under reflux then concentration is carried out and 225 mg of expected product (white crystals) is obtained, M.p.=207° C. 208° C.

ANALYSES:

IR NUJOL

OH/NH 3525-3365-3250 cm^{-1}

CN 2240 cm^{-1}

$\text{C}=\text{O}$ 1778-1738 cm^{-1}

Aromatic 1618-1578-1506 cm^{-1}

EXAMPLE 27

4-(4,4-bis(fluoromethyl) 2,5-dioxo 3-methyl 1-
imidazolidinyl) 2-(trifluoromethyl) benzonitrile

The operation is carried out as indicated in Example 23 using 120 mg of the product obtained in Example 24. After

chromatography on silica (eluant: CH_2Cl_2 —ethyl acetate 99-1), 111 mg of expected product is obtained. M.p.=137° C.—138° C.

ANALYSES:

IR CHCl_3 $\text{C}\equiv\text{N}$ 2235 cm^{-1} $\text{C}=\text{O}$ 1790–1735 cm^{-1} Aromatic 1617–1580–1505 cm^{-1}

EXAMPLE 28

4-(2,5-dioxo 3-ethyl 4-(hydroxymethyl 4-methyl 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

The operation is carried out as indicated in Example 2, Stages 2 and 3, using 1.033 g of the tetrahydropyranic ether prepared in Stage 1 and 0.24 ml of ethyl iodide. After chromatography on silica (eluant: methylene chloride—ethyl acetate), 0.796 g of expected product is obtained which is recrystallized from isopropanol. M.p.=138° C.

ANALYSES:

IR CHCl_3 OH 3616 cm^{-1} $\text{C}\equiv\text{N}$ 2236 cm^{-1} $\text{C}=\text{O}$ 1779 (m)—1725 (F) cm^{-1} Aromatic 1617–1506 cm^{-1}

EXAMPLE 29

3-(2,5-dioxo 3-ethyl 4-methyl 4-(2-methyl 1-oxopropoxy) methyl 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile

The operation is carried out as in Example 22 using 280 mg of product obtained in Example 28. 2.5 ml of pyridine, 23 mg of dimethylaminopyridine and 0.16 ml of isobutyric anhydride. After extraction with ether, elimination of the solvents and chromatography on silica (eluant: methylene chloride—ethyl acetate 100-1), 321 mg of expected product is obtained. M.p.=85° C.

ANALYSES:

IR CHCl_3 $\text{C}\equiv\text{N}$ 2235 cm^{-1} $\text{C}=\text{O}$ 1781 (m)—1728 (F) cm^{-1} Aromatic 1615–1575–1505 cm^{-1}

EXAMPLE 30

Carbonate of (1-(4-cyano 3-(trifluoromethyl) phenyl) 2,5-dioxo 3-ethyl 4-methyl 4-imidazolidinyl) methyl and of 1-methylethyl

The operation is carried out as in Example 22 using 376 mg of the product obtained in Example 28, 3.8 ml of pyridine, 25 mg of dimethylaminopyridine, and 2.2 ml of a toluene solution of isopropyl chloroformate (1M/l) is added at 0° C. After agitation for 30 minutes at 0° C. then for 3 hours at ambient temperature, 0.4 ml of the isopropyl chloroformate solution is added and agitation is continued at ambient temperature for 2 hours 30 minutes. The reaction medium is poured into 20 g of ice-cooled water, extracted with ether, the organic solution is washed with salt water, dried and the solvents are eliminated under reduced pressure. The residue is taken up in toluene, concentrated to dryness, the oil formed is left to crystallize and 422 mg of crude product is obtained which is chromatographed on silica (eluant: methylene chloride—ethyl acetate 100-2), 270 mg of expected product is obtained. M.p.=123° C.

ANALYSES:

IR CHCl_3 $\text{C}\equiv\text{N}$ 2235 cm^{-1} $\text{C}=\text{O}$ 1782–1744–1729 cm^{-1} 5 Aromatic 1616–1578–1505 cm^{-1}

EXAMPLE 31

4-(4,4-bis(hydroxymethyl) 2,5-dioxo 3-(4-hydroxybutyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.

1) Formation of the tetrahydropyranic ethers.

The operation is carried out as in Example 24 Stage 1, using 331 mg of the product obtained in Example 26. Extraction is carried out with methylene chloride, the extracts are washed with salt water, dried, the solvent is evaporated off and, after chromatography on silica (eluant: CH_2Cl_2 —MeOH 9-1), 500 mg of expected product is obtained, used as it is for the following stage.

2) Hydroxyalkylation.

456 mg of the diether obtained above in 3 ml of dimethylsulphoxide is added drop by drop over 20 minutes to 52 mg of sodium hydride then 20 minutes after cessation of the release of hydrogen, 374 mg of trimethylsilyl-4-iodobutanol is added. After 40 minutes of reaction, the medium is poured into 20 ml of water, extracted with ether, the organic phase is washed with salt water, dried and the solvent is evaporated off. 650 mg of crude product is obtained which is used as it is for the following stage.

3) Hydrolysis of the protective groups.

650 mg of the product obtained above is introduced into 7 ml of methanol and 3 ml of 2N hydrochloric acid then the solution is taken to 40° C. for about 40 minutes. It is poured into 20 ml of an aqueous solution of sodium bicarbonate, extracted with ethyl acetate, the extracts are washed with salt water, dried, the solvent is evaporated off. Purification is carried out on silica (eluant: CH_2Cl_2 —MeOH 9-1) and 950 mg of expected product is obtained.

ANALYSES:

UV EtOH

Max. 237 nm ϵ =8600Max. 263 nm ϵ =14000Infl. 278 nm ϵ =8400Infl. 287 nm ϵ =4200

EXAMPLE 32

4-(4,4-bis(hydroxymethyl) 2,5-dioxo 3-(2-fluoroethyl) 1-imidazolidinyl) 2-(trifluoromethyl) benzonitrile.

50 1) Fluoroalkylation.

The operation is carried out as in Example 3 Stage a, starting with 5 g of the tetrahydropyranic diether prepared as indicated in Example 31 Stage 1, and 1.1 ml of 2-bromo-1-fluoroethane. 5.31 g of expected product is obtained.

2) Hydrolysis of the tetrahydropyranic ether.

The operation is carried out as in Example 3 Stage b starting with 550 mg of the product obtained above, 6 ml of methanol and 2 ml of 2N hydrochloric acid. After chromatography on silica (eluant: CH_2Cl_2 —Me₂CO 8-2), 351 mg of expected product is obtained. M.p.=138° C.—139° C.

ANALYSES:

IR NUJOL

OH/NH 3580–3505 cm^{-1} $\text{C}\equiv\text{N}$ 2245 cm^{-1} 65 $\text{C}=\text{O}$ 1778–1716 cm^{-1} Aromatic 1616–1580–1512 cm^{-1}

UV EtOH

37

Max. 260 nm $\epsilon=15300$
 Infl. 280 nm $\epsilon=3400$

EXAMPLE 33

4-(4,4-bis(fluoromethyl) 2,5-dioxo 3-(2-fluoroethyl) 1-imidazolidinyl) 2-trifluoromethyl) benzonitrile.

1 ml of tetrahydrofuran is cooled down to -50°C . under an inert atmosphere and 0.66 ml of diethylamino sulphide trifluoride is added drop by drop, then 375 mg of the product obtained in Example 32 in 4 ml of tetrahydrofuran is added over 5 minutes. The reaction medium is left to return to ambient temperature, maintained under agitation for one hour, poured into an ice-cooled aqueous solution of sodium bicarbonate, extraction is carried out with chloroform, the organic phase is washed with salt water, dried, the solvent is evaporated off, the residue is chromatographed on silica (eluant: CH_2Cl_2 -cyclohexane 9-1) and 337 mg of expected product is obtained. M.p.= 136 - 137°C .

ANALYSES:

IR CHCl_3
 $\text{C}\equiv\text{N}$ 2235 cm^{-1}
 $\text{C}=\text{O}$ 1787-1736 cm^{-1}
 Aromatic 1617-1577-1505 cm^{-1}

EXAMPLE 34

4-(4,4-bis(2-methyl 1-oxopropoxy) methyl) 2,5-dioxo 3-(2-fluoroethyl) 1-imidazolidinyl) 2-trifluoromethyl) benzonitrile.

0.5 ml of isobutyric anhydride is added under an inert atmosphere to a solution containing 375 mg of the product obtained in Example 32, 4 ml of pyridine and 122 mg of dimethylamino-pyridine. Agitation is carried out for 30 minutes, the reaction medium is poured into 20 ml of a 50% aqueous solution of sodium bicarbonate, followed by drying and evaporating the solvent under reduced pressure. The residue is chromatographed on silica (eluant: CH_2Cl_2 -AcOEt 95-5) and 457 mg of expected product is obtained. M.p.= 71°C - 72°C .

ANALYSES:

IR CHCl_3
 $\text{C}\equiv\text{N}$ 2236 cm^{-1}
 $\text{C}=\text{O}$ 1789-1733 cm^{-1}
 Aromatic 1616-150 cm^{-1}
 UV EtOH
 Max. 257 nm $\epsilon=17000$
 Infl. 285 nm $\epsilon=2600$

EXAMPLE 35

Carbonate of bis (1-methylethyl) and (3-(4-cyano 3-(trifluoromethyl) phenyl) 2,4-dioxo 1-(2-fluoroethyl) 5-imidazolidinyl) bis (methylene) and \pm racemic of carbonate of (3-(4-cyano 3-(trifluoromethyl) phenyl) 2,4-dioxo 1-(2-fluoroethyl) 5-(hydroxymethyl) 5-imidazolidinyl) methyl and of 1-methylethyl

375 mg of the product obtained in Example 32 in 4 ml of pyridine and 122 mg of 4-dimethylaminopyridine is cooled down to -4°C . under an argon atmosphere. 550 mg of isopropyl chloroformate is added drop by drop at -4°C . The reaction medium is left to return to ambient temperature, agitation is continued for 2 hours. The reaction being incomplete, 122 mg of dimethylaminopyridine and 2 ml of isopropyl chloroformate are added and the mixture is heated

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for 18 hours at 50°C . It is returned to ambient temperature, poured into salt water, extraction is carried out with ethyl acetate, the extracts are dried, the solvents are eliminated, and 570 mg of crude product is obtained which is purified by chromatography on silica (eluant: CH_2Cl_2 -AcOEt 95-5), in order to obtain 275 mg of dicarbonate (M.p.= 122°C - 123°C .) then (eluant: CH_2Cl_2 - Me_2CO 9-1), in order to obtain 156 mg of monocarbonate (M.p.= 154°C - 155°C).

ANALYSES:

10 . Dicarbonate
 IR CHCl_3
 $\text{C}\equiv\text{N}$ 2238 cm^{-1}
 $\text{C}=\text{O}$ 1789-1749-1734 cm^{-1}
 Aromatic 1615-1578-1505 cm^{-1}
 15 UV EtOH
 Max. 256 nm $\epsilon=15400$
 Infl. 285 nm $\epsilon=2500$
 . Monocarbonate
 IR (Nujol)
 20 OH/NH 3450 cm^{-1}
 $\text{C}\equiv\text{N}$ 2250 cm^{-1}
 $\text{C}=\text{O}$ 1789-1736 cm^{-1}
 Aromatic 1616-1576-1506 cm^{-1}

EXAMPLE 36

Carbonate of bis (2-methylpropyl) and of (3-(4-cyano 3-(trifluoromethyl) phenyl) 2,4-dioxo 1-(2-fluoroethyl) 5-imidazolidinyl) bis (methylene)

375 mg of the product obtained in Example 32 in 4 ml of pyridine and 122 mg of 4-dimethylaminopyridine is cooled down to -4°C . under an argon atmosphere. 550 mg of isobutyl chloroformate is added drop by drop at -4°C . The medium is left to return to ambient temperature. After 40 minutes, the reaction medium is poured into water, washed with salt water, dried and the solvents are eliminated, the residue is chromatographed on silica (eluant: CH_2Cl_2 -AcOEt 92.5-7.5) and 476 mg of expected product is obtained. M.p.= 109°C - 110°C .

ANALYSES:

40 IR CHCl_3
 $\text{C}\equiv\text{N}$ 2236 cm^{-1}
 $\text{C}=\text{O}$ 1790-1754-1734 cm^{-1}
 Aromatic 1615-1578-1505 cm^{-1}
 45 UV EtOH
 Max. 256 nm $\epsilon=15500$
 Infl. 285 nm $\epsilon=2700$

EXAMPLE 37

50 Tablets were prepared having the following composition:

Product of Example 3	100 mg
Excipient s.q. for a tablet made up to	300 mg

55 (Detail of the excipient: lactose, starch, talc, magnesium stearate).

EXAMPLE 38

Tablets were prepared having the following composition:

Product of Example 26	100 mg
Excipient s.q. for a tablet made up to	300 mg

(Detail of the excipient: lactose, starch, talc, magnesium stearate).

PHARMACOLOGICAL STUDY OF THE
PRODUCTS OF THE INVENTION

1) Study of the affinity of the products of the invention for the androgen receptor

Male Sprague Dawley EOPS rats weighing 180-200 g, castrated 24 hours previously, are sacrificed, the prostates are removed, weighed and homogenized at 0° C. using a Potter glass, in a buffered solution (10 mM Tris, 0.25M saccharose, 0.1 mM PMSF (phenylmethanesulphonyl fluoride), 20 mM sodium molybdate, HCl pH 7.4) to which is added extemporaneously 2 mM of DTT (DL dithiothreitol), at the rate of 1 g of tissue per 8 ml of buffer.

The homogenate is then ultracentrifuged at 0° C., for 30 minutes at 209,000 g. Aliquots of the supernatant obtained (=cytosol), are incubated for 30 minutes and 24 hours at 0° C., with a constant concentration (I) of tritiated testosterone and in the presence of increasing concentrations (0 to 2500.10⁻⁹M), either of unlabelled testosterone, or of the products under test. The concentration of bound tritiated testosterone (B) is then measured in each incubate by the method of adsorption with carbon-dextran. Calculation of the relative bond affinity (RBA).

The following 2 curves are drawn: the percentage of the bound tritiated hormone B/T as a function of the logarithm of the concentration of the unlabelled reference hormone and B/T as a function of the logarithm of the concentration of the unlabelled product tested. The straight line of the equation $I_{50} = (B/T_{max} + B/T_{min})/2$ is determined. B/T_{max} = % of the bound tritiated hormone for an incubation of this tritiated hormone at the concentration (T). B/T_{min} = % of the bound tritiated hormone for an incubation of this tritiated hormone at the concentration (T) in the presence of a large excess of unlabelled hormone (2500.10⁻⁹M).

The intersections of the straight line I₅₀ and the curves enable the concentrations of the unlabelled reference hormone (CH) and of the unlabelled tested product (CX) which inhibit by 50% the binding of the tritiated hormone on the receptor to be evaluated. The relative bond affinity (RBA) of the tested product is determined by the equation $RBA = 100 (CH)/(CX)$.

The following results, expressed in RBA, are obtained. Reference product (Testosterone): 100

Product of Examples	Incubation: 24 hours
3	6
6	16
26	4

2) Determination of the androgen or anti-androgen activity of the products of the invention using dosage of ornithine decarboxylase (ODC).

Treatment protocol

6-week old male SWISS mice, castrated 24 hours previously, receive, by oral or percutaneous route, the products being studied (suspension in methyl cellulose at 0.5% or in solution in ethanol), simultaneously with a subcutaneous injection of testosterone propionate 3 mg/kg (solution in corn oil) in order to determine the antiandrogen activity. The agonistic activity is determined in the absence of testosterone propionate.

Testosterone propionate is administered in a volume of 10 ml/kg.

20 hours after the treatments, the animals are sacrificed, the kidneys are removed, then homogenized at 0° C., using a teflon glass grinder in 10 volumes of Tris-HCl 50 mM (pH

7.4) buffer containing 250 μM of pyridoxal phosphate, 0.1 mM EDTA, and 5 mM of dithiothreitol. The homogenate is then centrifuged at 209,000 g for 30 minutes.

Dosage principle

At 37° C., renal ornithine decarboxylase converts an isotopic mixture of unlabelled ornithine and tritiated ornithine into unlabelled putrescine and tritiated putrescine.

The putrescine is then collected on selective ion-exchange papers. After drying, the excess unconverted tritiated and unlabelled ornithine is eliminated by washing 3 times with 0.1M of ammonium hydroxide. The papers are dried, then the radioactivity is counted after addition of scintillating Aqualite.

The results are expressed as fmoles (10⁻¹⁵M) of tritiated putrescine formed/hour/mg of proteins.

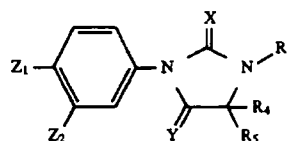
The results are expressed as % of inhibition of the ODC of the controls receiving only testosterone propionate. Test: the products are administered by percutaneous route at 1.5 mg/kg in a volume of 10 μl.

Products of Examples	Test
3	47
6	84

Conclusion: The tests indicated above show that the tested products of the invention possess a strong anti-androgen activity.

We claim:

1. A compound in all possible racemic, enantiomeric and diastereoisomer forms of the formula



wherein Y is —O—, Z₂ is —CF₃, Z₁ is —CN or —NO₂, X is —O— or —S—, R₃ is hydrogen or alkyl of 1 to 4 carbon atoms optionally substituted by at least one halogen or —CN and R₄ and R₅ are individually alkyl of 1 to 4 carbon atoms optionally substituted with a member of the group consisting of halogen, —OH, esterified or esterified or protected hydroxy and phenylthio optionally substituted by halogen or —OH, and wherein at least one of R₄ and R₅ is substituted.

2. A compound of a) of claim 1 selected from the group consisting of 2-(trifluoromethyl)-4-(4-(hydroxymethyl)-4-methyl-2,5-dioxo-1-imidazolidinyl)-benzoxazole, 4-(3,4-dimethyl)-4-(hydroxymethyl)-5-oxo-2-thioxo-1-imidazolidinyl-2-(trifluoromethyl)-benzoxazole, 2-(trifluoromethyl)-4-(4-(hydroxymethyl)-3,4-dimethyl-2,5-dioxo-1-imidazolidinyl)-benzoxazole, 4-(2,5-dioxo-3-(2-fluoroethyl)-4-(hydroxymethyl)-4-methyl-1-imidazolidinyl)-2-(trifluoromethyl)-benzoxazole, 1,5-dimethyl-5-(hydroxymethyl)-3-(4-nitro-3-(trifluoromethyl)phenyl)-2,4-imidazolidinedione and 4-(4,4-bis(hydroxymethyl)-2,5-dioxo-1-imidazolidinyl)-2-(trifluoromethyl)-benzoxazole.

3. A compound of b) of claim 1 selected from the group consisting of 4-(4-(fluoromethyl)-3,4-dimethyl)-2,5-dioxo-1-imidazolidinyl-2-(trifluoromethyl)-benzoxazole, 4-(3,4-dimethyl)-4-(fluoromethyl)-5-oxo-2-thioxo-1-imidazolidinyl-2-(trifluoromethyl)-benzoxazole, 4-(2,5-dioxo-3-(2-fluoroethyl)-4-(fluoromethyl)-4-methyl-1-imidazolidinyl)-2-(trifluoromethyl)-benzoxazole, 4-(2,4-

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dioxo-1,3-diazaspiro(4.4)nonan-3-yl)-2-(trifluoromethyl)-benzonitrile, 4-(2,4-dioxo-1-(2-fluoroethyl)-1,3-diazaspiro(4.4) nonan-3-yl)-2-(trifluoromethyl)-benzonitrile, 1,5-dimethyl-5-(fluoromethyl)-3-(4-nitro-3-(trifluoromethyl)-phenyl)-2,4-imidazolidinedione, 3-(4-cyano-3-(trifluoromethyl)-phenyl)-2,4-dioxo-5-(fluoromethyl)-5-methyl-1-imidazolidinacetonitrile and 4-(4,4-bis-(fluoromethyl)-3-methyl-5-oxo-2-thioxo-1-imidazolidinyl)-2-(trifluoromethyl)-benzonitrile.

4. A method of inducing antiandrogenic activity in warm-blooded animals comprising administering to warm-blooded

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animals an antiandrogenically effective amount of a compound of claim 1.

5. A method of inducing antiandrogenic activity in warm-blooded animals comprising administering to warm-blooded animals an antiandrogenically effective amount of a compound of claim 2.

6. A method of inducing antiandrogenic activity in warm-blooded animals comprising administering to warm-blooded animals an antiandrogenically effective amount of a compound of claim 3.

* * * * *

Brief Articles

Design, Synthesis, and Pharmacological Characterization of 4-[4,4-Dimethyl-3-(4-hydroxybutyl)-5-oxo-2-thioxo-1-imidazolidinyl]-2-iodobenzonitrile as a High-Affinity Nonsteroidal Androgen Receptor Ligand[#]

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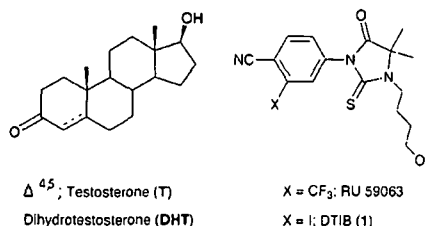
4-[4,4-Dimethyl-3-(4-hydroxybutyl)-5-oxo-2-thioxo-1-imidazolidinyl]-2-trifluoromethylbenzonitrile (RU 59063) is a prototype of a new class of high-affinity nonsteroidal androgen receptor (AR) ligands. The search for a radioiodinated AR ligand prompted us to synthesize 4-[4,4-dimethyl-3-(4-hydroxybutyl)-5-oxo-2-thioxo-1-imidazolidinyl]-2-iodobenzonitrile (DTIB) wherein the trifluoromethyl group of RU 59063 was substituted with the similarly hydrophobic iodine atom. DTIB displayed subnanomolar binding affinity ($K_i = 0.71 \pm 0.22$ nM) for the rat AR in competitive binding assays. Additionally, DTIB demonstrated potent agonist activity, comparable to that of the natural androgen 5 α -dihydrotestosterone (DHT), in a cell-based functional assay (cotransfection assay). DTIB represents a new lead for the development of high-affinity radioiodinated AR radioligands.

Introduction

The androgen receptor (AR) is an important member of the superfamily of nuclear hormone receptors that function as ligand-dependent regulators of transcription.¹ ARs respond to signaling by the endogenous steroidal androgens testosterone and 5 α -dihydrotestosterone (DHT) and play a critical role in sexual development and function in males.²

The development and progression of prostate cancer is known to be androgen-dependent, and AR expression is frequently observed in primary prostate tumors and metastases.³ Consequently, AR-targeted radioligands are under investigation for the noninvasive imaging of tumor sites in prostate cancer using positron emission tomography (PET) and single-photon emission computed tomography (SPECT).⁴ The majority of these studies to date have focused on steroid-based ligands, including the naturally occurring steroid ligands (testosterone, DHT) and synthetic steroids [mibolerone, metribolone (R 1881)].⁵ Successful PET imaging of prostate and prostate tumor metastases has been recently reported in a preliminary clinical study with a fluorine-18-labeled derivative of DHT.⁶ In contrast, the development of high-affinity radioiodinated steroid-based AR ligands for SPECT imaging has had limited success. This is attributed, in part, to the sensitivity of

Chart 1. Structures of AR Ligands



the AR ligand binding domain to the bulky nature of iodinated steroids.⁷

Nonsteroidal antiandrogens offer an alternative approach to the design of high-affinity radioiodinated AR ligands. Since nonsteroidal AR ligands have greater conformational flexibility and are more amenable to structural modification than steroid-based ligands, they could serve as templates for the design of iodinated ligands that are better accommodated at the AR ligand binding domain. RU 59063 (Chart 1) is a prototype member of a novel class of nonsteroidal antiandrogens which displays high AR affinity ($K_a = 5.4$ nM for human AR) and selectivity (>1000-fold selectivity for AR over progestin, glucocorticoid, mineralocorticoid, and estrogen receptors).⁸ In addition, [³H]RU 59063 demonstrated high specific binding and 3–8-fold greater AR binding affinity as compared to [³H]testosterone in radioligand binding assays conducted with rat, mouse, hamster, and human AR tissue preparations.⁸

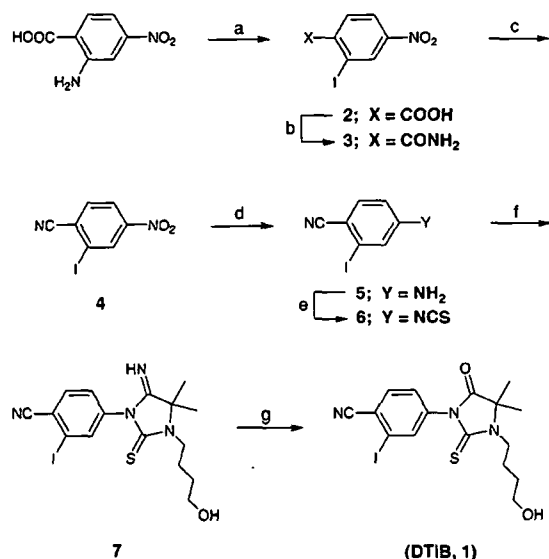
The high AR selectivity, reduced lipophilicity, and ease of synthesis of RU 59063 and its derivatives as compared to steroid analogues make these compounds attractive candidates for investigation as radioiodinated AR radioligands. As a first step toward this goal, we

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Scheme 1^a

selected the iodinated derivative 4-[4,4-dimethyl-3-(4-hydroxybutyl)-5-oxo-2-thioxo-1-imidazolidinyl]-2-iodobenzonitrile (DTIB) (Chart 1) for initial evaluation based on the similar hydrophobic properties of the trifluoromethyl ($\pi = 0.88$) and iodine ($\pi = 1.12$) groups.⁹ We report here the synthesis of DTIB and its initial in vitro pharmacological characterization (AR binding, AR-mediated transcriptional activation) as a novel high-affinity, nonsteroidal AR ligand.

Chemistry

DTIB (**1**) was prepared by a seven-step synthetic route as outlined in Scheme 1. Commercially available 2-amino-4-nitrobenzoic acid was converted to 2-iodo-4-nitrobenzoic acid (**2**) in moderate yield (42%) via the Sandmeyer diazotization reaction. Sequential reaction of **2** with thionyl chloride and ammonia provided 2-iodo-4-nitrobenzamide (**3**). Dehydration of **3** was accomplished with thionyl chloride to give the nitrile **4**, which was subsequently reduced to the aniline derivative **5** with stannous chloride in EtOH. Synthesis of the isothiocyanate derivative **6** and subsequent steps were conducted according to the general procedure of Teutsch et al.⁸ Thus, treatment of **5** with thiophosgene gave the isothiocyanate **6** which was condensed with 2-(4-hydroxybutylamino)-2-cyanopropane in the presence of triethylamine to afford the imine intermediate **7**. Acid hydrolysis of **7** provided DTIB (**1**) as a white amorphous solid in 16% overall yield. DTIB and intermediate compounds were purified by silica gel flash chromatography and fully characterized by ¹H NMR, mass spectral, and elemental analysis.

Results and Discussion

The binding affinity of DTIB and RU 59063 to the rat prostate cytosolic AR was determined using a competitive binding assay in the presence of the high-affinity AR radioligand, [³H]mibolerone.¹⁰ These data

Table 1. Inhibition Constants for DTIB (**1**) and Selected Ligands at the Rat AR^a

compd	K _i ± SEM (nM) ^b
DTIB (1)	0.71 ± 0.22
RU 59063	2.23 ± 0.50
DHT	0.69 ± 0.20
testosterone	1.4 ± 0.4
mibolerone	0.75 ± 0.08

^a Binding studies were conducted using rat prostate cytosol AR and [³H]mibolerone as radioligand. ^b Data are presented as mean ± SEM and are the average of at least three determinations each conducted in duplicate.

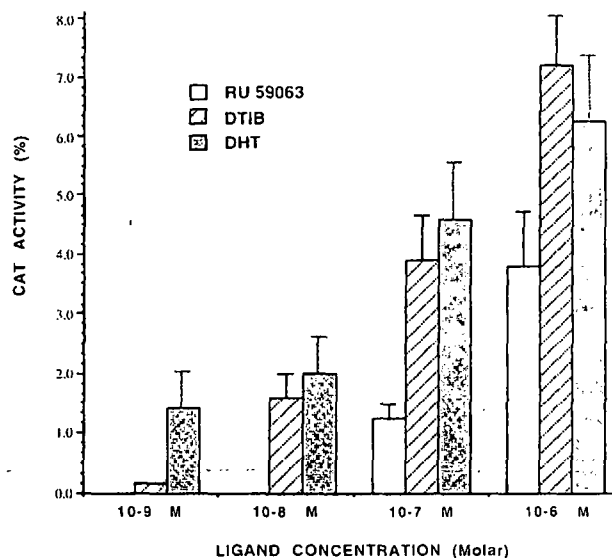


Figure 1. Comparison of AR agonist dose response of RU 59063, DTIB, and DHT in cotransfected CV-1 cells. Data points represent the mean ± SEM of triplicate determinations; chloramphenicol acetyltransferase (CAT) activity is reported as % conversion/ μ g of protein; RU 59063 was tested at two dose levels: 10⁻⁷ and 10⁻⁶ M.

and the binding affinity values obtained for other routinely used AR ligands such as DHT, mibolerone, and testosterone are presented in Table 1. As seen from these data, DTIB ($K_i = 0.71 \pm 0.22$ nM) is one of the highest-affinity AR ligands reported to date. DTIB inhibited [³H]mibolerone binding to the rat AR as efficiently as mibolerone and DHT, two tritiated AR ligands currently in use. Furthermore, DTIB showed a 3-fold improvement in AR affinity over RU 59063 indicating that iodine substitution was well-tolerated in this region of the molecule. Hill coefficients (data not shown) were close to unity in all cases indicating that the binding interaction was to a single site.

The AR functional activity of DTIB, RU 59063, and DHT was evaluated using an in vitro cell-based assay (cotransfection assay) system.¹¹ DTIB and RU 59063 displayed a dose-dependent stimulation of AR-mediated transcriptional activity in these assays with a potency comparable to that of the natural hormone agonist DHT (Figure 1). Similar agonistic effects (data not shown) were observed for these ligands using a more complex natural gene fragment (C' Δ 9, a transcriptional enhancer of the mouse sex-limited protein gene¹²). The agonistic behavior of RU 59063 seen in transfection experiments is in contrast to the potent in vivo AR antagonist activity reported for this analogue in rodent models.⁸ A similar discrepancy between in vitro and in vivo functional

activity has been observed for the structurally similar antiandrogen RU 56187.¹³

Although our initial design strategy of DTIB was based on the similar hydrophobicity of iodine and trifluoromethyl, it must be noted that these functional groups do not share similar electronic and steric properties. In particular, iodine is less electronegative (σ_m , σ_p values are 0.35 and 0.18, respectively, for iodine versus 0.43 and 0.54, respectively, for CF₃) and has a 3-fold larger steric size than trifluoromethyl (molar refractivity values for iodine and CF₃ are 13.9 and 5.0, respectively).⁹ Our binding affinity data therefore indicate that the AR binding domain that interacts with the trifluoromethyl substituent in RU 59063 is sufficiently large to accommodate the increased steric bulk of an iodine atom. Taken together, these results suggest that steric and hydrophobic interactions may be more important than electronic interactions at this binding region for high AR binding in this class of nonsteroidal ligands.

Conclusions

In summary, the present study has identified a new iodinated nonsteroidal AR ligand, DTIB, which demonstrates subnanomolar binding affinity and potent agonist activity in *in vitro* AR binding and functional assays, respectively. These studies also suggest the existence of a large hydrophobic pocket in the AR binding domain that interacts with the *meta* substituent of this pharmacophore. Studies are currently underway to evaluate the utility of radioiodinated analogues of DTIB as radioligands for *in vitro* and *in vivo* studies of AR.

Experimental Section

General Methods. Melting points were determined with a Thomas-Hoover melting point apparatus and are uncorrected. ¹H NMR spectra were obtained in either CDCl₃ or CD₃OD with a Bruker WM-360 (360 MHz) instrument using tetramethylsilane (TMS) as internal standard. Chemical shifts (δ) are reported in parts per million (ppm) relative to TMS, and coupling constants (J) are reported in hertz (Hz). Elemental analyses were performed by the Department of Chemistry, University of Michigan, and were within $\pm 0.4\%$ of the calculated values. RU 59063 and 2-(4-hydroxybutylamino)-2-cyanopropane were synthesized as previously described.⁸ All other chemical reagents were obtained from Aldrich Chemical Co., Milwaukee, WI, and were used without further purification. Organic extracts were dried over anhydrous Na₂SO₄ and concentrated to dryness by rotoevaporation under reduced pressure.

2-Iodo-4-nitrobenzoic Acid (2). A vigorously stirred slurry of 2-amino-4-nitrobenzoic acid (10 g, 55 mmol) and aqueous 9 N H₂SO₄ (64 mL) was diazotized at 0 °C (ice-salt bath) by dropwise addition of a solution of NaNO₂ (4.17 g, 60.4 mmol) in water (50 mL). The mixture was stirred at 0 °C for 1 h and treated with urea (1.1 g, 18.3 mmol) to destroy excess HNO₂. A solution of NaI (9.9 g, 66 mmol) in H₂O (50 mL) was then added dropwise at 0–5 °C. The reaction was allowed to warm to ambient temperature, stirred a further 2 h, diluted with 1% aqueous NaHSO₃ solution (200 mL) and filtered. The crude product was rinsed with hot EtOAc (3 \times 100 mL) to remove a dark orange side product and the residue was purified by flash chromatography [gradient elution with EtOAc:hexane:glacial acetic acid (30:70:1 to 80:20:1)] to give 6.8 g (42%) of 2 as light yellow needles: mp 143.5–145 °C [benzene:hexane (1:1)]; ¹H NMR (CD₃OD) δ 8.77 (d, 1H, J = 2.2, H-3), 8.30 (dd, 1H, J = 8.5, 2.2, H-5), 7.92 (d, 1H, J = 8.5, H-6). Anal. (C₇H₄NO₄I) C, H, N.

2-Iodo-4-nitrobenzamide (3). A mixture of 2 (6.60 g, 22.5 mmol) and SOCl₂ (100 mL) was heated at 60 °C for 3 h under argon and concentrated by rotoevaporation. Residual SOCl₂ was removed from the crude product mixture by coevaporation with dry CHCl₃ (3 \times 50 mL). Concentrated aqueous NH₃ (100 mL) was then added and the mixture stirred overnight at ambient temperature. The precipitate was filtered, rinsed with H₂O and dried in an oven at 60 °C. The crude product was purified by flash chromatography (75% EtOAc in hexane) to give 5.6 g (85%) of 3 as cream-colored crystals: mp 208–209 °C (EtOAc); ¹H NMR (CD₃OD) δ 8.70 (d, 1H, J = 2.2, H-3), 8.29 (dd, 1H, J = 8.5, 2.2, H-5), 7.59 (d, 1H, J = 8.4, H-6). Anal. (C₇H₅N₂O₃I) C, H, N.

2-Iodo-4-nitrobenzotrile (4). A mixture of 3 (5.5 g, 18.8 mmol) and SOCl₂ (35 mL) was refluxed for 3 h under argon and concentrated under reduced pressure. The residue was purified by flash chromatography (20% EtOAc in hexane) to give 4.12 g (80%) of 4 as cream-colored crystals: mp 154–155.5 °C [EtOAc:hexane (1:4)]; ¹H NMR (CDCl₃) δ 8.76 (d, 1H, J = 2.2, H-3), 8.32 (dd, 1H, J = 8.5, 2.2, H-5), 7.82 (d, 1H, J = 8.6, H-6). Anal. (C₇H₃N₂O₂I) C, H, N.

4-Cyano-3-iodoaniline (5). A mixture of 4 (2.3 g, 8.4 mmol) and SnCl₂·2H₂O (9.3 g, 41.4 mmol) in EtOH (35 mL) was refluxed for 2 h. The mixture was concentrated under reduced pressure and treated with H₂O (100 mL) and the pH was adjusted to 9 by treatment with 5% aqueous NaHCO₃. The mixture was extracted with EtOAc; the organic layers were washed with saturated brine and H₂O and dried. The residue obtained after removal of solvent was purified by flash chromatography (40% EtOAc in hexane) to afford 2 g (98%) of 5 as white fluffy crystals: mp 146–148 °C [EtOH:H₂O (1:5)]; ¹H NMR (CDCl₃) δ 7.33 (d, 1H, J = 8.5, H-5), 7.14 (d, 1H, J = 2.3, H-2), 6.62 (dd, 1H, J = 8.5, 2.2, H-6), 4.19 (br s, 2H, exchangeable with D₂O, NH₂). Anal. (C₇H₅N₂I) C, H, N.

2-Iodo-4-isothiocyanatobenzotrile (6). A solution of 5 (0.54 g, 2.2 mmol) in THF (10 mL) was added dropwise at ambient temperature to a well-stirred suspension of thiophosgene (0.30 g, 2.6 mmol) in H₂O (5 mL). TLC analysis [silica; hexane:EtOAc (6:1)] at 3 h indicated completeness of reaction. The mixture was diluted with H₂O (20 mL), extracted with CHCl₃ (2 \times 20 mL) and dried. The residue obtained after removal of solvent was dried under high vacuum to afford a quantitative yield of a yellow-brown solid which was used directly in the next step. A small portion of the crude product was purified by flash chromatography (15% EtOAc in hexane) to give an analytical sample of 6 as a white solid: mp 116–118 °C; ¹H NMR (CDCl₃) δ 7.76 (d, 1H, J = 2.1, H-3), 7.58 (d, 1H, J = 8.3, H-6), 7.26 (dd, 1H, J = 8.3, 2.1, H-5); HRMS (EI) calcd for C₈H₃N₂IS (M⁺) 285.9062, found 285.9058.

4-[4,4-Dimethyl-3-(4-hydroxybutyl)-5-imino-2-thioxo-1-imidazolidinyl]-2-iodobenzotrile (7). A solution of 2-(4-hydroxybutylamino)-2-cyanopropane⁸ (0.30 g, 1.94 mmol) in anhydrous THF (2 mL) was added dropwise at ambient temperature to a stirred solution of the crude isothiocyanate 6 (0.55 g, 1.94 mmol) and Et₃N (25 mg, 0.25 mmol) in anhydrous THF (2 mL). The reaction mixture was refluxed for 1 h, at which point, TLC analysis [silica; hexanes:EtOAc (6:1)] indicated completeness of reaction. The reaction mixture was concentrated under reduced pressure and the residue purified by flash chromatography (35% acetone in chloroform) to provide 0.55 g (64%) of the title compound 7 as a straw-colored viscous liquid: ¹H NMR (CDCl₃) δ 7.93 (s, 1H, Ar-H), 7.75 (d, 1H, J = 8.3, Ar-H), 7.49 (d, 1H, J = 8.2, Ar-H), 3.75–3.66 (m, 4H, –NCH₂– and –CH₂O–), 1.92 (m, 2H, CH₂), 1.66 (m, 2H, CH₂), 1.58 (s, 6H, C(CH₃)₂); HRMS (EI) calcd for C₁₆H₁₉N₄ISO (M⁺) 442.0324, found 442.0320.

4-[4,4-Dimethyl-3-(4-hydroxybutyl)-5-oxo-2-thioxo-1-imidazolidinyl]-2-iodobenzotrile (DTIB, 1). A solution of the imine derivative 7 (0.48 g, 1.1 mmol) in CH₃OH (8 mL) was treated with aqueous 2 N HCl (1 mL) and refluxed for 1 h. The cooled reaction mixture was poured into H₂O (80 mL) and extracted with EtOAc (2 \times 50 mL), and the organic layers dried and concentrated under reduced pressure. The crude product was purified by flash chromatography (70% EtOAc in

hexane) to give 0.44 g (90%) of DTIB (1) as a white amorphous solid: $^1\text{H NMR}$ (CDCl_3) δ 7.96 (d, 1H, $J = 1.9$, Ar-H3), 7.71 (d, 1H, $J = 8.3$, Ar-H6), 7.52 (dd, 1H, $J = 8.3, 1.9$, Ar-H5), 3.73 (m, 4H, $-\text{NCH}_2-$ and $-\text{CH}_2\text{O}-$), 1.94 (m, 2H, CH_2), 1.66 (m, 2H, CH_2), 1.57 (s, 6H, $\text{C}(\text{CH}_3)_2$). Anal. ($\text{C}_{16}\text{H}_{18}\text{N}_3\text{O}_2$) S, H, N.

Androgen Receptor Binding Assays. Androgen receptor binding assays were conducted by a commercial laboratory (MDS Panlabs, Bothell, WA) using [^3H]mibolerone as radioligand and rat prostate cytosol as a source of AR as previously described.¹⁰ In brief, cytosol from the ventral prostate of castrated male Wistar rats (175 \pm 25 g) was prepared in modified phosphate buffer (pH 7.2) containing protease inhibitors and triamcinolone acetonide (5 mM). Competitive binding of test ligands were determined by incubation of increasing concentrations (0.3–100 nM) of each ligand and a saturating concentration of [^3H]mibolerone (2 nM) with cytosol (0.4 mg) at 4 $^\circ\text{C}$ for 18 h. Separation of bound and free radioligand was achieved by further incubation with a hydroxylapatite slurry for 15 min and filtration. Radioactivity bound to the washed filters was quantified by liquid scintillation counting. Binding affinity data are reported as mean \pm standard error of the mean (SEM) and represent the average of at least three separate experiments each conducted in duplicate.

Cotransfection Assays. AR-mediated transcriptional activation by ligands was studied in a cell-based bioassay system (cotransfection assays) as previously reported.¹¹ In brief, mammalian CV-1 cells were transfected with the mouse AR and an androgen-responsive reporter gene, consisting of three tandem androgen response elements upstream of the thymidine kinase promoter and the bacterial chloramphenicol acetyltransferase (CAT) gene. Media containing the reference compound (DHT) or test compound in concentrations ranging from 1 nM to 1 μM were added to the cells. Three independent transfection assays were performed. Following incubation, cells were washed with phosphate buffered saline and lysed by several cycles of freeze-thaw. Cell extracts were assayed for CAT activity and the data reported as CAT conversion rates (%).¹¹

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References

- (1) (a) Evans, R. M. The steroid and thyroid hormone receptor superfamily. *Science* **1988**, *240*, 889–895. (b) Tsai, M. J.; O'Malley, B. W. Molecular mechanisms of action of steroid/thyroid receptor superfamily members. *Annu. Rev. Biochem.* **1994**, *63*, 451–486. (c) Katzenellenbogen, J. A.; Katzenellenbogen, B. S. Nuclear hormone receptors: ligand-activated regulators of transcription and diverse cell responses. *Chem. Biol.* **1996**, *3*, 529–536.
- (2) (a) Willy, P. J.; Mangelsdorf, D. J. Nuclear orphan receptors: the search for novel ligands and signaling pathways. In *Hormones and Signaling*; O'Malley, B. W., Ed.; Academic Press: San Diego, 1998; Vol. 1, pp 307–358. (b) Wilson, E. M.; Simental, J. A.; French, F. S.; Sar, M. Molecular analysis of the androgen receptor. *Ann. N. Y. Acad. Sci.* **1991**, *637*, 56–63.
- (3) Hobisch, A.; Culig, Z.; Radmayr, C.; Bartsch, G.; Klocker, H.; Hittmair, A. Androgen receptor status of lymph node metastases from prostate cancer. *Prostate* **1996**, *28*, 129–135.
- (4) (a) Katzenellenbogen, J. A. Designing steroid receptor-based radiotracers to image breast and prostate tumors. *J. Nucl. Med.* **1995**, *36* (Suppl.), 8S–13S. (b) Labaree, D. C.; Brown, T. J.; Hoyte, R. M.; Hochberg, R. B. 7 α -Iodine-125-iodo-5 α -dihydrotestosterone: a radiolabeled ligand for the androgen receptor. *J. Nucl. Med.* **1997**, *38*, 402–409. (c) Ali, H.; Rousseau, J.; van Lier, J. E. Synthesis of (17 α ,20E/Z)iodovinyltestosterone and 19-nortestosterone derivatives as potential radioligands for androgen and progesterone receptors. *J. Steroid Biochem. Mol. Biol.* **1994**, *49*, 15–29.
- (5) (a) Bonasera, T. A.; O'Neil, J. P.; Xu, M.; Dobkin, J. A.; Cutler, P. D.; Lich, L. L.; Choe, Y. S.; Katzenellenbogen, J. A.; Welch, M. J. Preclinical evaluation of fluorine-18-labeled androgen receptor ligands in baboons. *J. Nucl. Med.* **1996**, *37*, 1009–1015. (b) Liu, A. J.; Katzenellenbogen, J. A.; Van Brocklin, H. F.; Mathias, C. J.; Welch, M. J. 2-[F-18]Fluoromibolerone, a positron-emitting radiotracer for androgen receptors: synthesis and tissue distribution studies. *J. Nucl. Med.* **1991**, *32*, 81–88.
- (6) Cutler, P. D.; Dehdashti, F.; Siegel, B. A.; Downer, J. B.; Welch, M. J. Investigation of a prostate ligand 16 β -[F-18]Fluoro-5 α -dihydrotestosterone for staging prostate carcinoma. *J. Nucl. Med.* **1996**, *37* (Suppl.), 87P.
- (7) Welch, M. J.; Downer, J. B.; Katzenellenbogen, J. A. Steroid hormone receptors as targets for diagnostic imaging. In *Current Directions in Radiopharmaceutical Research and Development*; Mather, S. J., Ed.; Kluwer Academic Publishers: Dordrecht, The Netherlands, 1996; pp 137–156.
- (8) Teutsch, G.; Goubet, F.; Battmann, T.; Bonfils, A.; Bouchoux, F.; Cerede, E.; Gofflo, D.; Gaillard-Kelly, M.; Philibert, D. Nonsteroidal antiandrogens: synthesis and biological profile of high-affinity ligands for the androgen receptor. *J. Steroid Biochem. Mol. Biol.* **1994**, *48*, 111–119.
- (9) Hansch, C.; Leo, A. The Hydrophobic Parameter: Measurement and Calculation. In *Exploring QSAR: Fundamentals and Applications in Chemistry and Biology*; Heller, S. R., Ed.; American Chemical Society: Washington, DC, 1995; Chapter 1, p 7, Chapter 3, p 73, Chapter 4, pp 108–109.
- (10) (a) Schilling, K.; Liao, S. The use of radioactive 7 α ,17 α -dimethyl-19-nortestosterone (mibolerone) in the assay of androgen receptors. *Prostate* **1984**, *5*, 581–588. (b) Liao, S.; Witte, D.; Schilling, K.; Chang, C. The use of a hydroxylapatite-filter steroid receptor assay method in the study of the modulation of androgen receptor interaction. *J. Steroid Biochem.* **1984**, *20*, 11–17.
- (11) (a) Adler, A. J.; Scheller, A.; Hoffman, Y.; Robins, D. M. Multiple components of a complex androgen-dependent enhancer. *Mol. Endocrinol.* **1991**, *5*, 1587–1596. (b) Gorman, C. M.; Mofatt, L. F.; Howard, B. H. Recombinant genomes which express chloramphenicol acetyltransferase in mammalian cells. *Mol. Cell. Biol.* **1982**, *2*, 1044–1051. (c) Seed, B.; Sheen, J.-Y. A simple phase-assay for chloramphenicol acyltransferase activity. *Gene* **1988**, *67*, 271–277.
- (12) Robins, D. M.; Scheller, A.; Adler, A. J. Specific steroid response from a nonspecific DNA element. *J. Steroid Biochem. Mol. Biol.* **1994**, *49*, 251–255.
- (13) Kempainen, J. A.; Langley, E.; Wong, C.; Bobseine, K.; Kelce, W. R.; Wilson, E. M. Distinguishing androgen receptor agonists and antagonists: distinct mechanisms of activation by medroxyprogesterone acetate and dihydrotestosterone. *Mol. Endocrinol.* **1999**, *13*, 440–454.

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Articles

Three-Dimensional Structure–Activity Relationships of Nonsteroidal Ligands in Complex with Androgen Receptor Ligand-Binding Domain

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We studied the three-dimensional quantitative structure–activity relationships (3D QSAR) of 70 structurally and functionally diverse androgen receptor (AR) binding compounds using the comparative molecular similarity indices analysis (CoMSIA) method. The compound set contained 67 nonsteroidal analogues of flutamide, nilutamide, and bicalutamide whose binding mode to AR was unknown. Docking was used to identify the preferred binding modes for the nonsteroidal compounds within the AR ligand-binding pocket (LBP) and to generate the ligand alignment for the 3D QSAR analysis. The alignment produced a statistically significant and predictive model, validated by random group cross-validation and external test sets ($q^2_{LOO} = 0.656$, SDEP = 0.576, $r^2 = 0.911$, SEE = 0.293; $q^2_{10} = 0.612$, $q^2_5 = 0.571$; pred- $r^2 = 0.800$). Additional model validation comes from the CoMSIA maps that were interpreted with respect to the LBP structure. The model takes into account and links the AR LBP structure, docked ligand structures, and the experimental binding activities. The results provide valuable information on intermolecular interactions between nonsteroidal ligands and the AR LBP.

Introduction

Endogenous androgens testosterone (T) and 5 α -dihydrotestosterone (DHT) are essential steroid hormones for the development, maintenance, and regulation of the male phenotype. Their androgenic and anabolic actions are elicited via androgen receptors (ARs), which function as ligand-dependent transcription factors in the regulation of AR target gene expression.¹ AR belongs to the superfamily of nuclear receptors (NRs) and shares a common modular three-domain structure of the NRs. The transcriptional activation of the ARs, like other NRs, is regulated through agonist and antagonist binding resulting in conformational changes in the ligand-binding domain (LBD) and subsequent recruitment of coregulators.^{1–3}

In the agonist-bound form of AR the LBD folds into a compact structure with the ligand-binding pocket (LBP) enclosed by the carboxyl-terminal helix 12 (H12) (Figure 1).^{4–6} The conformational changes occurring upon antagonist binding are unknown for AR. In related NRs, e.g. estrogen receptor (ER), the active antagonists displace the H12 of the ER LBD into a coactivator recognition surface known as activation function 2 (AF2).^{7,8} The displaced H12 thus prevents coactivators from binding to the receptor.^{7,9} Antagonist-induced displacement is possible in ER because the H12 contains the same recognition sequence (LXXLL) as ER coactivators that interact with the AF2.⁷ However, no evi-

dence has been reported for a similar antagonistic mechanism in AR.

Exogenous AR ligands have for long been used in the treatment of conditions resulting from altered androgen levels or responsiveness.¹⁰ AR targeted pharmaceuticals can structurally be classified into steroidal or nonsteroidal compounds and functionally into androgenic or antiandrogenic compounds. Both androgenic (e.g. testosterone and its derivatives) and antiandrogenic (e.g. cyproterone acetate) functional activities of the steroidal structures are used clinically. Only antiandrogenic compounds of the nonsteroidal class of AR ligands (e.g. bicalutamide, flutamide, and nilutamide) are currently in medicinal use. However, several research groups have recently reported discovery of nonsteroidal androgens.^{11–16}

From the pharmacological point of view, nonsteroidal compounds are superior to the steroidal ones in terms of receptor selectivity and pharmacokinetic properties.^{17,18} These properties make them more attractive alternatives as novel therapeutic agents. Recently, interest has especially been directed toward the development of selective androgen receptor modulators (SARMs) with tissue-specific agonist and antagonist activities.^{10,19}

In the past years a number of experimental structure–activity relationship (SAR) studies have been conducted on a few structural classes of nonsteroidal AR binding compounds.^{11–16,20–28} In this study we applied the 3D QSAR analysis based on the comparative molecular similarity indices analysis (CoMSIA)²⁹ method to gain insight into the structural and chemical features influencing the binding affinity of a panel of 70 AR binding

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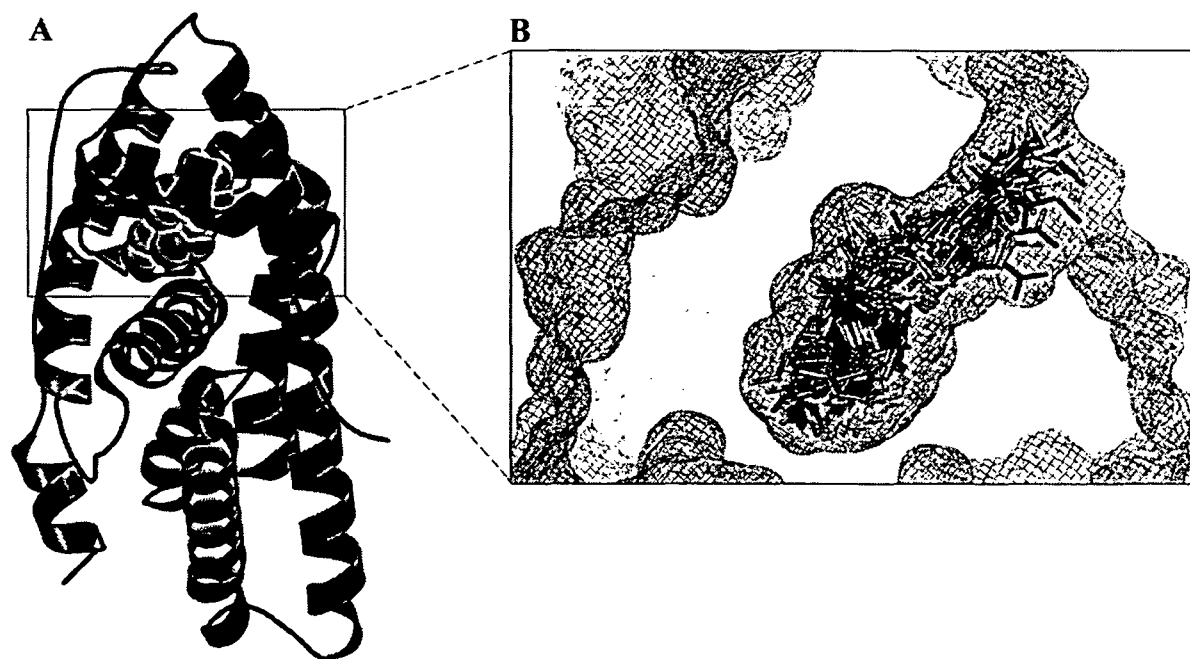


Figure 1. (A) The androgen receptor ligand-binding domain structure presented in the agonist-bound conformation in complex with the natural androgen dihydrotestosterone. Helix 12 is shown in red. (B) A close-up view of the structural alignment of ligands generated by molecular docking. The superposition is shown inside the Connolly solvent accessible surface of the ligand-binding pocket.

compounds containing 67 nonsteroids obtained from the literature.^{11,24–27} Since the structure of AR LBD in complex with nonsteroidal ligands has not yet been experimentally resolved, we used molecular docking to predict the biologically active conformations of the nonsteroids and to create the structural alignment of AR ligands for model building. The generation of a reliable 3D QSAR model requires that the ligands be aligned in a way to maximize the 3D overlap of their structural and functional features. To the best of our knowledge, this is the first report within AR research where molecular docking is combined with 3D QSAR analysis. Docking provides a powerful way to screen the conformational space of the ligands in search of the preferred binding conformation, while taking into account the structure and the chemical environment of the AR LBP. Combining docking and the 3D QSAR analysis have been shown to yield predictive 3D QSAR models.^{30–32} This approach allows also the visualization and interpretation of the CoMSIA maps to be made within the AR LBP, thus enlightening the interactions that are beneficial or detrimental for the binding affinity of the compounds in their proposed binding mode.

Results and Discussion

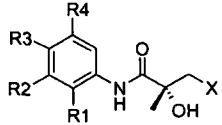
Ligand Alignment. The structures and pharmacological data for 70 AR binding compounds used in the 3D QSAR analysis were obtained from five previous publications reported by two laboratories.^{11,24–27} The results from the two laboratories were merged in our study, as the laboratories follow similar experimental methods in binding affinity measurements. The steroid reference compound DHT is included in the articles of both laboratories and is reported to have affinity values close to each other thus indicating that the laboratories produce comparable affinity data. The compound series

was divided into a training set of 61 compounds and a test set of 9 compounds. The test set was selected to contain compounds spanning a wide activity range and the main structural elements integrated within the training set.

The most crucial and challenging step in 3D QSAR analysis is the generation of the structural alignment of the compounds under study. The aim is to identify an alignment, among numerous possible ones, that represents the biologically active conformations of the compounds. At many instances this task is facilitated if the binding geometry for a compound from the series under investigation has been experimentally observed.^{30,32} This was not the situation in our case, however, as the experimentally determined binding mode of the nonsteroidal AR ligands has not been reported to date. Another challenge in the superpositioning was the structural and functional diversity of the compound set. Structurally the set can be divided into six chemical families (Tables 1–6). Primarily, the compounds are derivatives of the nonsteroidal structures flutamide, nilutamide, and bicalutamide. The steroidal structures of T, DHT, and mibolerone, which are known to bind AR LBP, were included in the published binding affinity data sets and incorporated in the examined set of compounds. Functionally the compounds represent the entire spectrum of pharmacological activities ranging from agonists to partial agonists and antagonists. For all of the compounds the functional activity has not been reported. The competitive binding assays indicate, however, that all the compounds do bind to AR LBP, as they are able to displace the high-affinity radioligand mibolerone from binding to the receptor.

Inclusion of compounds with different functionalities can be a potential source of errors in this study, since

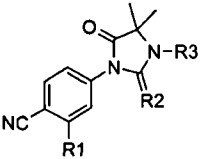
Table 1. Structures of Flutamide Derivatives



compd	R ₁	R ₂	R ₃	R ₄	X	p <i>K</i> _{iExp} ^b	p <i>K</i> _{iPred} ^c
1 ^{a,d}	H	CF ₃	CN	H	Br	8.10	7.86
2 ^d	H	CF ₃	NO ₂	H	Br	9.52	9.19
3 ^d	H	CF ₃	CN	H	I	8.55	8.09
4 ^d	H	CF ₃	NO ₂	H	I	9.07	8.80
5 ^e	H	H	NO ₂	H	Br	7.48	7.82
6 ^e	NO ₂	H	NO ₂	H	Br	5.93	5.63
7 ^e	H	NO ₂	H	NO ₂	Br	7.09	6.67
8 ^e	H	CF ₃	NH ₂	H	Br	7.09	7.17

^a Compound that belongs to the test set. ^b Experimental binding affinity (p*K*_i). ^c Predicted binding affinity (p*K*_i). ^d Reference 11. ^e Reference 27.

Table 2. Structures of Nilutamide Derivatives



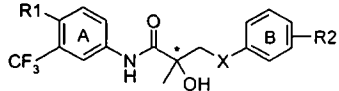
compd	R ₁	R ₂	R ₃	p <i>K</i> _{iExp} ^b	p <i>K</i> _{iPred} ^c
9 ^d	CF ₃	O	CH ₃	7.43	7.17
10 ^d	CF ₃	O	(CH ₂) ₂ OCH ₃	6.92	7.10
11 ^d	CF ₃	O	(CH ₂) ₂ F	6.85	7.04
12 ^d	CF ₃	O	(CH ₂) ₃ F	6.49	7.04
13 ^d	CF ₃	O	(CH ₂) ₄ F	6.67	7.08
14 ^d	CF ₃	O	CH ₂ (<i>m</i> -I-Ph)	6.91	6.49
15 ^d	CF ₃	O	CH ₂ (<i>p</i> -I-Ph)	6.82	6.46
16 ^d	CF ₃	O	(CH ₂) ₂ (<i>m</i> -I-Ph)	6.09	5.87
17 ^{a,d}	CF ₃	O	(CH ₂) ₂ (<i>p</i> -I-Ph)	6.38	7.01
18 ^d	CF ₃	O	(CH ₂) ₃ (<i>m</i> -I-Ph)	6.44	6.47
19 ^d	CF ₃	O	(CH ₂) ₃ (<i>p</i> -I-Ph)	7.19	7.01
20 ^d	CF ₃	O	(CH ₂) ₄ OH	7.59	7.28
21 ^e	CF ₃	S	(CH ₂) ₄ OH	8.65	8.42
22 ^e	I	S	(CH ₂) ₄ OH	9.15	8.96

^{a-c} See Table 1. ^d Reference 26. ^e Reference 25.

the molecular mechanism for AR antagonism and the resulting conformational changes are not known. It is possible that AR displays an antagonistic mechanism similar to the antagonistic mechanism of ERs that involves displacement of H12 into the cofactor-binding cleft on the surface of LBD.^{7,9} However, H12 of AR LBD does not possess the same recognition sequence as the recognition sequences proposed for cofactors and AR amino-terminal sequences that target AR LBD.^{33,34} Also, AR antagonists in this study do not have an extension or "arm", like typical ER antagonists.⁸ Thus, we expect the conformational changes induced by AR antagonists in our ligand set to cause less violent changes in AR LBD than those caused by ER antagonists in ER LBD. For these reasons we did not model the antagonist-bound structure of AR LBD on the basis of known antagonist forms of other steroid receptors. Instead, we aligned all the ligands using a protein model based on the agonist structure of AR LBD (PDB ID: 1gs4).⁶

Docking Simulations. We used the docking program GOLD³⁵ to identify the most favorable conformations for the nonsteroidal compounds within the AR LBP and to study the ligand-receptor interactions. The structural water molecule, which is found in most of the

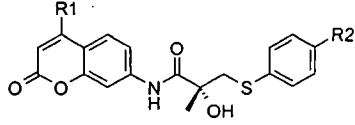
Table 3. Structures of Bicalutamide Derivatives



compd	R ₁	R ₂	X	isomer	p <i>K</i> _{iExp} ^b	p <i>K</i> _{iPred} ^c
23 ^d	CN	<i>p</i> -F	SO ₂	<i>R</i>	7.96	7.32
24 ^d	CN	<i>p</i> -F	SO ₂	<i>S</i>	6.44	6.45
25 ^{a,d}	CN	<i>p</i> -NH ₂	S	<i>R</i>	7.05	6.88
26 ^{a,d}	CN	<i>p</i> -NH ₂	S	<i>S</i>	6.10	6.70
27 ^d	CN	<i>m</i> -NH ₂	S	<i>R</i>	7.19	7.25
28 ^d	CN	<i>p</i> -NCS	S	<i>S</i>	6.37	6.63
29 ^d	CN	<i>m</i> -NCS	S	<i>R</i>	6.64	7.03
30 ^d	CN	<i>m</i> -NCS	S	<i>S</i>	6.89	6.94
31 ^d	CN	<i>p</i> -NCS	SO ₂	<i>R</i>	7.39	7.34
32 ^d	CN	<i>m</i> -NCS	SO ₂	<i>R</i>	6.85	7.28
33 ^d	CN	<i>μ</i> -NHCOCH ₂ Cl	S	<i>R</i>	8.78	8.42
34 ^d	CN	<i>p</i> -NHCOCH ₂ Cl	S	<i>S</i>	6.70	6.84
35 ^d	CN	<i>p</i> -NHCOCH ₂ Br	S	<i>R</i>	7.00	7.38
36 ^d	CN	<i>p</i> -NHCOCH ₂ Cl	SO ₂	<i>R</i>	7.97	7.75
37 ^{a,d}	CN	<i>m</i> -NHCOCH ₂ Cl	SO ₂	<i>R</i>	6.89	7.06
38 ^d	CN	<i>p</i> -NHCOCH ₂ Br	SO ₂	<i>R</i>	6.44	6.92
39 ^d	CN	<i>p</i> -NH ₂	SO ₂	<i>R</i>	7.49	7.51
40 ^d	CN	<i>p</i> -NHCOCH ₃	S	<i>R</i>	8.31	8.26
41 ^d	CN	<i>p</i> -NHCOCH ₃	S	<i>S</i>	5.86	6.25
42 ^d	CN	<i>p</i> -N(COCH ₃) ₂	S	<i>R</i>	6.98	6.90
43 ^d	CN	<i>p</i> -NHCOCH ₃	SO ₂	<i>R</i>	7.80	8.07
44 ^d	CN	<i>p</i> -NHCOCH ₂ CH ₃	S	<i>R</i>	7.57	7.52
45 ^d	CN	<i>p</i> -N(COCH ₂ CH ₃) ₂	S	<i>R</i>	6.18	6.59
46 ^{a,e}	NO ₂	<i>p</i> -NH ₂	S	<i>R</i>	7.24	7.86
47 ^{a,e}	NO ₂	<i>p</i> -NH ₂	S	<i>S</i>	6.28	6.19
48 ^e	NO ₂	<i>p</i> -NHCOCH ₃	S	<i>R</i>	8.54	8.68
49 ^e	NO ₂	<i>p</i> -NHCOCH ₃	S	<i>S</i>	6.91	6.83
50 ^e	NO ₂	<i>p</i> -NHCOCH ₃	SO ₂	<i>R</i>	8.03	8.39
51 ^e	NO ₂	<i>p</i> -NHCOCH ₃	SO ₂	<i>S</i>	6.23	6.09
52 ^e	NO ₂	<i>p</i> -NHCOCF ₃	S	<i>R</i>	8.59	8.82
53 ^{a,e}	NO ₂	<i>p</i> -NHCOCF ₃	SO ₂	<i>R</i>	8.21	8.65
54 ^e	NO ₂	<i>p</i> -NHCOCH ₂ Cl	S	<i>R</i>	8.48	8.63
55 ^e	NO ₂	<i>p</i> -NHCOCH ₂ Cl	S	<i>S</i>	7.01	7.23
56 ^e	NO ₂	<i>p</i> -NHCOCH ₂ Cl	SO ₂	<i>R</i>	8.16	7.73
57 ^e	NO ₂	<i>p</i> -NHCOCH ₂ Cl	SO ₂	<i>S</i>	6.63	6.12
58 ^e	NO ₂	<i>p</i> -NHSO ₂ CH ₃	S	<i>R</i>	7.30	7.34
59 ^e	NO ₂	<i>p</i> -NHSO ₂ CH ₃	SO ₂	<i>R</i>	7.82	8.19

^{a-c} See Table 1. ^d Reference 24. ^e Reference 27.

Table 4. Structures of Bicalutamide Derivatives Bearing a Coumarine Ring

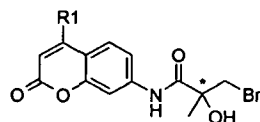


compd	R ₁	R ₂	p <i>K</i> _{iExp} ^b	p <i>K</i> _{iPred} ^c
60 ^d	CH ₃	F	7.04	7.16
61 ^d	CF ₃	H	6.88	6.80
62 ^{a,d}	CF ₃	F	6.47	6.81
63 ^d	CF ₃	NCS	6.87	6.77
64 ^d	CF ₃	NH ₂	6.54	6.65

^{a-c} See Table 1. ^d Reference 27.

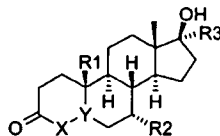
ligand-complexed NR LBD structures (like ER, GR, PR, and AR), was kept as part of the protein structure during the docking.

Crystal structures of AR LBD complexed to steroid agonists, both natural and synthetic, have revealed the common binding mode and the key interactions contributing to the binding of steroids.⁴⁻⁶ Besides the van der Waals interactions between the steroid core and hydrophobic residues of the LBP, the steroids form three hydrogen bonds to the receptor in the wild-type AR. Hydrogen bonds are formed to Arg752 at the carbonyl

Table 5. Structures of Hydroxyflutamide Derivatives Bearing a Coumarine Ring

compd ^a	R ₁	isomer	p <i>K</i> _{iExp} ^b	p <i>K</i> _{iPred} ^c
65 ^d	CH ₃	R	6.56	6.53
66 ^d	CF ₃	R	6.88	6.97
67 ^d	CF ₃	S	6.61	6.61

^a No compounds in this table belong to the test set. ^{b,c} See Table 1. ^d Reference 27.

Table 6. Structures of Steroids

compd ^a	X-Y	R ₁	R ₂	R ₃	p <i>K</i> _{iExp} ^b	p <i>K</i> _{iPred} ^c
68 ^d	CH ₂ -CH	CH ₃	H	H	9.55	9.36
69 ^e	CH-C	CH ₃	H	H	8.85	8.97
70 ^e	CH-C	H	CH ₃	CH ₃	9.12	9.14

^a No compounds in this table belong to the test set. ^{b,c} See Table 1. ^d Reference 11. ^e Reference 25.

oxygen end of the steroid and to Asn705 and Thr877 at the hydroxyl group end of the steroid (Table 6).^{4,5} Hydrogen bonding is considered essential also for nonsteroidal ligand binding.^{20,21} The significance of Asn705 for the activity and binding of nonsteroidal antagonists hydroxyflutamide and bicalutamide (casodex) has been demonstrated in a mutation study.³⁶

Docking simulations were started using a modified AR LBP structure in which two mutations (His701Leu and Ala877Thr) restoring the wild-type LBD sequence were made. Upon docking into this LBP many of the large nonsteroidal compounds with several aromatic rings adopt "sandwich-like" conformations where the rings lie on top of each other. These conformations are not likely the bioactive conformations but merely the result of forcing the ligands to fit into the volume and shape of the agonist-bound conformation of AR LBP. These docking poses do not satisfy important ligand-receptor interactions, and the alignment of such poses does not provide an explanation for the variance of the biological data. To emphasize the hydrogen-bonding interactions of the steroidal and nonsteroidal ligands to Asn705, DHT was used to guide docking, although it is not structurally an optimal template for the large and flexible nonsteroids. Additionally, another mutation (Phe876Ala) was made to allow rational alignment of the bulky nonsteroidal ligands in the LBP. The enlarged volume generated by this mutation corresponds to a region surrounding the five-ring of the steroidal scaffold in their bioactive conformation, i.e. the region on the opposite side of the LBP from the structural water molecule. This mutation resulted in extended binding conformations of nonsteroidal ligands and enabled key interactions between ligand and receptor to be formed. The Phe876Ala mutation could partly account for the structural changes that we assume to take place in the LBD upon binding of large nonsteroidal compounds,

which would otherwise have problems to fit in the AR binding cavity.

The aim of docking was to generate the ligand alignment as automatically and objectively as possible. Unfortunately, the selection of top-scored conformations based on the scoring functions of GOLD, XSCORE,³⁷ or CSCORE³⁸ (including the individual scoring functions implemented in CSCORE) did not produce alignments resulting in statistically significant models. To obtain a well-superposed set of ligands, the alignment based on GOLD scoring was improved by manually selecting docking poses for ligands that deviated from the alignment. In the set of best-ranked conformations according to GOLD there were 10 ligands (12, 14, 22, 32, 36, 44, 48, 50, 52, 54) whose structural features were not superimposed with the rest of the docked ligand set. For these ligands the aromatic A-ring and its substitutions (Table 3) were not aligned with the rest of the docked ligands and did not produce statistical correlation with experimental binding affinities in 3D QSAR analysis using the CoMSIA method. Also the amide group next to the A-ring was often not oriented similarly as in the other docked ligands. For these 10 problematic ligands, GOLD simulation did generate conformations whose structural and functional elements were aligned with the majority of the ligands but which were not top-ranked in GOLD scoring. An average decrease of 2.0 in the GOLD fitness score was detected between the best-ranked conformation and the conformations where the structural and functional elements for these 10 ligands were aligned with the rest of the ligand set. These differences in the GOLD score can be considered to be of minor importance. The molecular alignment resulting from docking of the training set compounds is shown in Figure 1 within the AR LBP visualized with the Connolly solvent accessible surface calculated in BODIL.³⁹

3D QSAR Model. The molecular alignment derived from docking simulations was used for the generation of the 3D QSAR model based on the CoMSIA method.²⁹ This approach allowed us to study the physicochemical characteristics that contribute to the binding affinity of the investigated set of AR ligands. Hydrophobic and hydrogen bond acceptor fields and five PLS components in model building provided the best explanation for the variation in the binding affinity data. The hydrophobic fields contribute 51% to the information content of the final model, while the hydrogen bond acceptor fields represent the remaining 49%.

The leave-one-out (LOO) cross-validation used to determine the optimum number of PLS components for model building produced a correlation coefficient q^2_{LOO} of 0.656 and standard error of prediction (SDEP) of 0.576. Internal correlation coefficient values for 10 random group cross-validation yielded an average q^2_{10} of 0.612 and SDEP of 0.612, and those for five random group cross-validation yielded an average q^2_5 of 0.571 and SDEP of 0.643. The conventional correlation coefficient r^2 obtained by the nonvalidated analysis gave 0.911 with a standard error of estimate (SEE) of 0.293. The q^2 and r^2 values of our 3D QSAR model indicate a statistically significant and stable model.

The r^2 value signifies that approximately 91% of the variance in ligand binding of the present series of

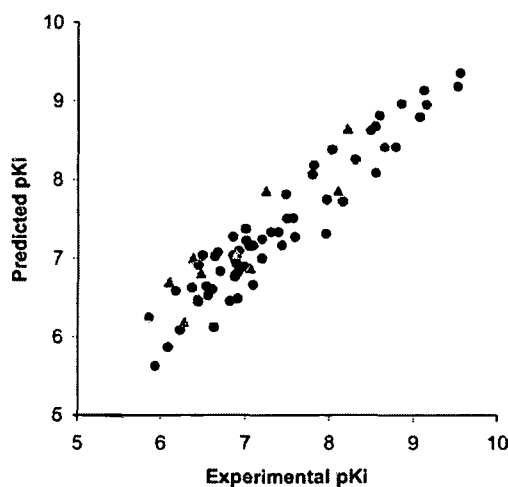


Figure 2. Correlation between the experimental and predicted activities (pK_i) for the training set compounds (●) and test set compounds (▲).

compounds can be explained with the model. The standard error value of the model means that if a compound were predicted to have a 10 nM (K_i) affinity, its actual affinity would fall into a range between 5 and 20 nM ($10^{-8 \pm 0.3}$ M). The error of model can never be less than the experimental error of the data that was used to train the model. The experimental and predicted binding affinity values of the training set ligands are listed in Tables 1–6 and graphed in Figure 2.

Six compounds from the publications where the binding affinity data was collected were excluded from this study (compounds 16, 39, 47, 58 of Yin et al.,²⁷ compound 13 of van Dort et al.,²⁶ and compound (*R*)-7 of Kirkovsky et al.²⁴). They turned out as outliers in 3D QSAR model building and were thus removed from the final model. Compounds 16, 39, 47, and 58 from Yin et al.²⁷ were excluded from the ligand set because their structures caused problems in molecular modeling. Compound 16 has a metal-containing B-ring substituent (*p*-Sn(CH₃)₃) and compound 39 has an A-ring substitution (*p*-NCS) that are not present in any other investigated compound. Compound 47, on the other hand, is the only structure of the bicalutamide derivatives bearing a coumarine ring (Table 4) with a sulfone (–SO₂–) linkage, and compound 58 is the only nilutamide derivative bearing a coumarine ring in the entire compound set. Compounds with singular substituents are likely to cause structural changes in the AR LBP that we were unable to predict with the current modeling programs. Compound 13 from van Dort et al.²⁶ has an *N*-iodopropenyl side chain with a highly reactive double bond. Due to the apparent reactivity we decided to remove it from the training set. For the removal of compound (*R*)-7 from Kirkovsky et al.²⁴ we were unable to find any structural explanation. This compound repeatedly turned out as an outlier in all CoMSIA models we attempted to generate. This result led us to suspect that perhaps the reported experimental value might be erroneous.

Besides internal validation with the training set, we used an external test set of 9 compounds to validate the predictivity of the model. The external validation process can be considered the most valuable validation

method, as these compounds are completely excluded during the training of the model. Prior to prediction, the test set compounds were processed identically to the training set compounds, as described in the Methods section. Visual observation of the top-ranked docking poses by GOLD showed the conformations to be well structurally aligned with the training set compounds and thus were used for the prediction of the binding affinity. All of the test set compounds, which represent the different structural properties incorporated within the training set, are well predicted without any apparent outliers. Low- and high-affinity compounds are clearly separated in the prediction. We chose not to include compounds from the highest affinity end of the scale into the test set as, after all, there are only a few sub-nanomolar binders and they are valuable for the 3D QSAR model building. The chosen external test set yielded a predictive r^2 of 0.800 with a SEE value of 0.367. These values indicate a good predictive power and are in agreement with the statistical values from the internal validation procedures. The predicted versus experimental affinity values for the test set compounds are listed and plotted with the training set compounds in Tables 1–6 (marked with a) and Figure 2, respectively.

While our study was underway, Marhefka et al.⁴⁰ reported a new series of bicalutamide-like compounds in which the metabolically susceptible sulfur linkage was replaced with either oxygen or nitrogen, and new substitutions were introduced into the B-ring. We did not include this high-affinity compound series in the 3D QSAR model building because the training set already had 31 structurally similar compounds (Table 3), but rather used them to further validate our model. We produced 10 possible poses of each of these compounds using the docking procedure described in the Methods section. We then used our 3D QSAR model to predict the binding affinity of each of these poses and picked the one that was predicted to be the best. The docked conformations that give the highest predictions in the 3D QSAR model are well aligned with the ligand set used in this study. The predicted vs the experimental binding affinity value yields a SEE of 0.6. This value corresponds to a situation where a compound predicted to have a 10 nM (K_i) affinity in reality shows an affinity in the range between 2 and 40 nM ($10^{-8 \pm 0.6}$ M). The binding mode inside AR LBP of these novel high-affinity AR ligands can thus be identified with our 3D QSAR model. A similar approach can be used in the identification of active compounds from a molecular library in virtual screening, too. The compound is docked several times to the receptor model, and the representative docking pose for a novel compound is then picked using the best prediction according to the 3D QSAR model. If the selected docking pose is structurally aligned with the training set compounds in this study, the predicted affinity gives a good estimate of the actual binding affinity and the ligand conformation takes into account the chemical and structural features of the AR LBP. The limitation is that the model can only predict K_i of compounds that are structurally described by the training set.

Visualization of the 3D QSAR Model. The statistically relevant results from the 3D QSAR analysis are

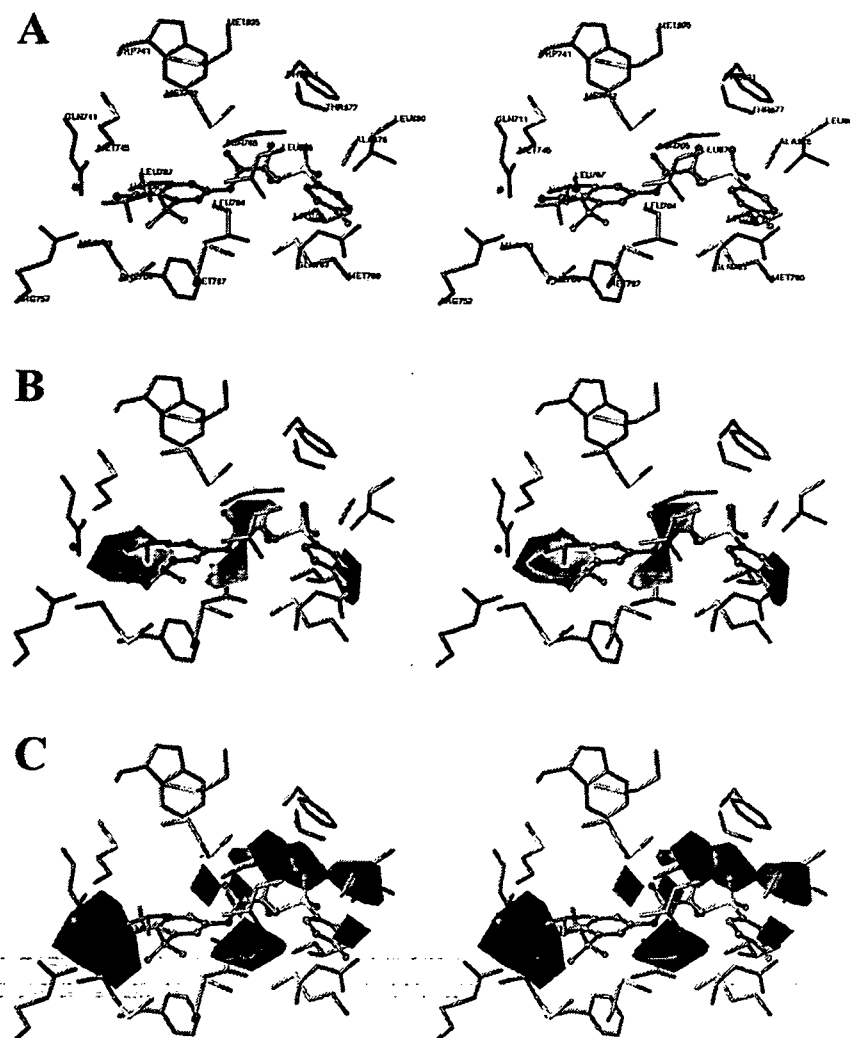


Figure 3. Stereoviews of the LBP and the 3D contour maps of the CoMSIA model represented as $\text{stdev} \times \text{coeff}$ plots. The favorable and unfavorable maps are shown with contribution levels of 80% and 20%, respectively. (A) The LBP residues (sticks) with the docked conformation of compound 23 ((*R*)-bicalutamide) (ball-and-stick). All hydrogens are omitted for clarity, and C^α -carbons are colored in purple. The structural water is shown as a red sphere. (B) The yellow contours represent regions in ligands where an increase in hydrophobicity is connected with enhanced affinity; purple contours represent regions where hydrophobicity is detrimental to affinity, thus favoring hydrophilicity. (C) The blue contours indicate volumes where hydrogen bond acceptors in ligands increase binding affinity; red contours indicate areas where hydrogen bond acceptors decrease binding affinity.

visualized as 3D contour maps. The contour maps of our CoMSIA model based on hydrogen bond acceptor and hydrophobic fields are displayed as PLS $\text{stdev} \times \text{coeff}$ maps in Figure 3. Since the superposition of ligands for the analysis was done using the receptor structure and ligand docking, the CoMSIA maps can be drawn inside the AR binding cavity. The maps of the 3D QSAR model based on the chemical properties and molecular interaction fields of ligands should correlate with the features of the LBP. Also, the ligand interactions with the residues of the binding cavity should provide an explanation of the variation of the experimental binding affinity. In Figure 3, the contour maps of our CoMSIA model are displayed with the binding site residues and with the docked conformation of ligand 23 ((*R*)-bicalutamide) as a reference structure.

Interpretation of the 3D QSAR Maps with Respect to AR LBP. For the analysis of the CoMSIA maps we divided the LBP roughly into three sections: the outer (solvent accessible) part, the center part, and

the inner part. The contours of the favorable hydrophobic and hydrogen bond acceptor fields used in the analysis are drawn at a contribution level of 80%, while the corresponding unfavorable fields are contoured at a contribution level of 20% (Figure 3).

The Outer Part. There is a small favored volume for acceptor interactions close to the solvent accessible surface, located near the amide group of residue Gln783 and the backbone carbonyl oxygen of Arg779 (not shown in Figure 3). The volume next to the side chain of Leu880 and the carbonyl oxygen of Phe876 (Ala876 in the receptor model used for docking) in the outer part of the binding site is unfavorable for acceptor interactions. There is a small hydrophilic volume located above the carbonyl oxygen of Ser778 and close to the volume that is unfavorable for acceptor interactions. Consistent with the aqueous surface, this part of the binding site has no favored volumes for hydrophobic interactions. Receptor structure and the 3D QSAR model consistently indicate that hydrophobic parts of

the ligands that come close to the solvent surface are not beneficial for binding affinity. It is sensible that hydrophilicity is favored in this volume, since polar water molecules can form interactions with the ligands that reach the outer part of the binding site.

The Center Part. According to the 3D QSAR model a volume in the center of the LBP, lined by the main chain carbonyl of Leu873, the aliphatic carbons of side chain Thr877, and the terminal methyl group of Met742, is defined as favorable for acceptor interactions. This result is rather surprising, because at first glance the interactions provided by these side chains seem hydrophobic. Our first impression was that the contour could be an artifact. However, Superstar^{41,42} calculations (data not shown) with water oxygen probe indicate that the volume could accommodate acceptor interactions even though these interactions do not seem very strong. The favored acceptor contour in this volume can partially explain the high binding affinity of e.g. compounds **36**, **50**, and **56**. In their docked conformations a sulfone linkage ($-\text{SO}_2-$) is positioned close to this volume. Although a general observation from the experimental results is that in most cases bicalutamide derivatives with a sulfide linkage show higher affinity than ones with a sulfone linkage, the effect of the linkage also largely depends on the substituent and its position in the aromatic B-ring.^{24,27}

There are two volumes in the center of the LBP where visualized CoMSIA maps indicate unfavorable acceptor interactions. These volumes are placed on the opposite sides of the superimposed ligands. The first unfavorable volume is located in the vicinity of the side chain carbonyl of Asn705 and the side chain aliphatic carbon of Leu704, and on top of the plane of the peptide bond between Leu704 and Asn705. This volume is clearly not a favorable site for acceptor interactions. The unfavorable acceptor interaction volume extends toward the side chain hydroxyl group of Thr877 and the hydrophobic residues Leu701, Leu880, and Phe891. While the hydroxyl group of Thr877 could provide an interaction that is constructive with acceptors, the other residues make the volume mostly hydrophobic. The second unfavorable volume for acceptor interactions is also indicated as favorable for hydrophobic interactions. Residues Met742, Met780, Met787, Leu873, and one edge of the aromatic ring of Phe764 form the borders of this volume in the center of the binding cavity. Thus, both the unfavorable acceptor and the favorable hydrophobic CoMSIA contours are in accordance with the receptor structure. On the whole, the CoMSIA maps described above show that the center of the LBP favors binding of hydrophobic moieties. Moreover, the center part lacks volumes defined as unfavorable for hydrophobic interactions. This result is in agreement with the observed interactions between the endogenous steroid ligand core and the AR LBP.⁵

The Inner Part. The inner part of the LBP, next to the side chains of Gln711 and Arg752 and the structural water molecule, displays a large favored acceptor volume that partly overlaps with a favored hydrophilic volume. Both maps agree with each other and with the receptor structure, since the hydrogen-bonding network between side chains of Gln711 and Arg752 and the water molecule are most likely able to donate a hydro-

gen bond. The inner part of the binding site displays a CoMSIA volume favoring hydrophobicity in a region above the plane of the peptide bond between Met745 and Val746 and close to the side chains of Met745, Val746, and Met749. In this part of the LBP the CoMSIA model is devoid of areas that are unfavorable for acceptor interactions.

Conclusions

In this paper we report a 3D QSAR model (CoMSIA) for a series of 70 AR binding compounds containing 67 nonsteroids. The set contains six structurally distinct scaffolds and a variety of pharmacological activities. Because the binding mode for nonsteroidal AR ligands was unknown, we used molecular docking to identify the bioactive conformations, and to superpose the structurally and functionally diverse ligands. Docking screens the conformational space of the ligands in search of the preferred binding conformation, while taking the chemical and structural features of the AR LBP into account.

The superposition of the ligands made with docking produced a statistically significant 3D QSAR model that has been carefully validated. Further validation for the 3D QSAR model comes from the interpretation of the contour maps explaining variation of the binding affinity of the ligands. The 3D QSAR model is compatible with the protein environment in the binding site, as the interpretation of the contour maps can be reflected to the amino acids of the AR LBP. These results indicate that the superposition is likely to represent the biologically active conformations of the nonsteroidal ligands.

The steps in the model building process are interdependent. The docking simulations depend on the structure of the AR LBP, and the 3D QSAR analysis in turn depends on the superposition of the ligands produced with the docking simulations. The experimentally observed changes in the binding affinity were mapped back to the structural features of the ligands in the 3D QSAR analysis, and the interpretation of the 3D QSAR model fits the chemical environment of AR LBP. Analysis of the maps is also consistent with the possible interactions formed between the nonsteroidal ligands and the AR LBP residues in the docking simulations. As a whole, there is a chain of dependencies between AR LBP, docking simulations, ligand alignment, statistical analysis, and experimental binding activities. The statistical validation thus confirms that there is a link between the different steps in the model building process and the reported experimental binding affinities. Together, the results provide valuable information on how nonsteroidal ligands bind and interact with the AR LBP. Nevertheless, it would be interesting to see whether similar binding geometries as presented here would be detected by experimental structure determination methods.

The procedure described in the paper can be used for, e.g., automated virtual identification of high-affinity AR ligands from chemical databases and structure-based optimization of AR ligands.

Methods

The Protein Data. The crystal structure of androgen receptor LBD in complex with an agonist 9 α -fluorocortisol (PDB ID: 1gs4)⁶ used in docking simulations was retrieved

from the Protein Data Bank.⁴³ 1gs4 is a mutant structure with two mutations in the active site (Leu701His, Thr877Ala). Prior to docking, His701Leu and Ala877Thr mutations restoring the wild-type AR sequence were made. Side chain conformations corresponding to ones observed in the crystal structure of AR LBD from PDB ID 1e3g⁴ were fixed using the rotamer library in the BODIL software.³⁹ An additional mutation (Phe876Ala) was made to allow more space to the solvent surface side of the binding cavity. 9 α -Fluorocortisol and all water molecules except the structural water located between residues Gln711 and Arg752 (Wat957 in 1gs4) were removed from the crystal structure. All hydrogens of the protein structure and of the water molecule were added using Sybyl 6.9.1.⁴⁴

The Ligand Data. The structural and pharmacological data for 70 AR binding compounds were obtained from five publications reported by two laboratories.^{11,24–27} Compounds lacking defined stereochemistry or exact affinity values were excluded from the data set. The results reported by the two laboratories were combined into our study, as the experimental procedures for binding affinity measurements are highly similar. The affinity measurements have been performed using competitive binding assay with cytosolic AR from rat ventral prostate. ³H-Mibolerone was used as the high-affinity ligand in the measurements and triamcinolone acetonide to block interaction of ³H-mibolerone with glucocorticoid and progesterone receptors. Moreover, the incubation times and temperatures in binding assays were the same and the hydroxyl-apatite precipitation was used as a method to determine the protein bound radioactivity from free radioactivity. The binding affinities reported as K_i (nM) were converted to pK_i values ($-\log K_i$) for the derivation of the 3D QSAR model.

The ligands were converted into 3D structures for docking using CORINA, version 2.6.^{45,46} The maximum number of ring conformations for one compound was restricted to three while using an energy window of 20 kJ/mol between the best and the worst conformations. This setting yielded a single conformation for each ligand, except testosterone (2 conformations) and mibolerone (3 conformations). Gasteiger–Hückel atom charges^{47,48} used for 3D QSAR analysis were calculated in Sybyl 6.9.1.

Docking Simulations. The docking program GOLD, version 2.0,³⁵ was used to predict the bioactive conformations and binding modes of the ligands within AR LBP. The binding pocket was defined as a cavity within a radius of 15 Å from atom C9 in 9 α -fluorocortisol in the crystal structure of 1gs4. The docking procedure was repeated 10 times for each of the CORINA-generated ligand conformations using the standard docking parameters in the GOLD manual. Docking of a single ligand conformation was allowed to terminate if the three top-scoring solutions were within a 1.5 Å root-mean-squared deviation (rmsd) of each other.

Docking of the nonsteroidal ligands was biased to emphasize similar hydrogen-bonding patterns as seen in the experimentally determined binding mode of the natural ligand DHT. The crystal structure of AR LBD with DHT (PDB ID: 1i37)⁵ was superimposed with our receptor model used in docking. The bound DHT was then extracted and merged into the receptor model with BODIL to be used as a template for docking. The template similarity constraint option of GOLD was applied by evaluating the overlap of all donor atoms and all acceptor atoms as well as shape overlap between DHT and the ligand being docked, with constraint weights 5.0, 5.0, and 10.0, respectively.

Selection of Docking Poses for 3D QSAR Analysis. Attempts to pick the top-scoring ligand poses using merely GOLD, XSCORE,³⁷ or CSCORE³⁸ scoring functions did not produce statistically significant alignments in the 3D QSAR analysis. As a consequence, manual selection of docking poses that we based on GOLD scoring was included into the alignment generation. For 10 of the ligands the top-ranked docking pose was not properly superimposed with the rest of the docked ligand set. For these problematic cases representatives that are consistent with the alignment produced by the

majority of the ligands were manually selected among the docking poses generated by GOLD simulation.

3D QSAR Analysis. The molecular alignment of the 61 docked ligands in the training set was used to build the 3D QSAR model with the CoMSIA method²⁹ to explore physicochemical properties contributing to binding affinity. The CoMSIA molecular descriptor fields, expressed as steric, electrostatic, hydrophobic, and hydrogen-bonding properties, were calculated using the default settings in Sybyl and correlated with the variations in the binding affinity data using the statistical method of partial least squares (PLS).⁴⁹ Molecular field descriptors with an energy variance less than 2.0 kcal/mol were filtered out from the PLS analysis. The 3D QSAR model was built using hydrophobic and hydrogen bond acceptor fields. The LOO cross-validation method was applied to determine the optimum number of PLS components, corresponding to the highest q^2 value and to the lowest SDEP value. The optimum of five components derived from the LOO cross-validation was used in the development and further validation of the 3D QSAR model.

The predictive value of the 3D QSAR model was validated first with internal cross-validation using 10 and five random groups. Due to the structural variability and the relatively small size of the training set, we did not apply random group cross-validation with a smaller number of groups. Each random group cross-validation procedure was repeated 25 times to calculate the mean q^2 values. Further validation of the predictivity of the model was done with an external test set of 9 compounds not included in 3D QSAR model building. The correlation between the experimental and the predicted activities for the test set compounds for the 3D QSAR model is represented as the predictive r^2 value.

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References

- Gobinet, J.; Poujol, N.; Sultan, C. Molecular action of androgens. *Mol. Cell. Endocrinol.* **2002**, *198*, 15–24.
- Bourguet, W.; Germain, P.; Gronemeyer, H. Nuclear receptor ligand-binding domains: three-dimensional structures, molecular interactions and pharmacological implications. *Trends Pharmacol. Sci.* **2000**, *20*, 381–388.
- Berrevoets, C. A.; Umar, A.; Brinkmann, A. O. Antiandrogens: selective androgen receptor modulators. *Mol. Cell. Endocrinol.* **2002**, *198*, 97–103.
- Matias, P. M.; Donner, P.; Coelho, R.; Thomaz, M.; Peixoto, C.; et al. Structural evidence for ligand specificity in the binding domain of the human androgen receptor. *J. Biol. Chem.* **2000**, *275*, 26164–26171.
- Sack, J. S.; Kish, K. F.; Wang, C.; Attar, R. M.; Kiefer, S. E.; et al. Crystallographic structures of the ligand-binding domains of the androgen receptor and its T877A mutant complexed with the natural agonist dihydrotestosterone. *Proc. Natl. Acad. Sci. U.S.A.* **2001**, *98*, 4904–4909.
- Matias, P. M.; Carrondo, M. A.; Coelho, R.; Thomaz, M.; Zhao, X.-Y.; et al. Structural basis for the glucocorticoid response in a mutant human androgen receptor (Ar^{CCR}) derived from an androgen-independent prostate cancer. *J. Med. Chem.* **2002**, *45*, 1439–1446.
- Shiau, A. K.; Barstad, D.; Loria, P. M.; Cheng, L.; Kushner, P. J.; et al. The structural basis of estrogen receptor/coactivator recognition and the antagonism of this interaction by tamoxifen. *Cell* **1998**, *95*, 927–937.
- Shiau, A. K.; Barstad, D.; Radek, J. T.; Meyers, M. J.; Nettles, K. W.; et al. Structural characterization of a subtype-selective ligand reveals a novel mode of estrogen receptor antagonism. *Nat. Struct. Biol.* **2002**, *9*, 359–364.
- Brzozowski, A. M.; Pike, A. C.; Dauter, Z.; Hubbard, R. E.; Bonn, T.; et al. Molecular basis of agonism and antagonism in the estrogen receptor. *Nature* **1997**, *389*, 753–758.
- Zhi, L.; Martinborough, E. Selective androgen receptor modulators (SARMS). *Annu. Rep. Med. Chem.* **2001**, *36*, 169–180.

- (11) Dalton, J. T.; Mukherjee, A.; Zhu, Z.; Kirkovsky, L.; Miller, D. D. Discovery of nonsteroidal androgens. *Biochem. Biophys. Res. Commun.* **1998**, *244*, 1-4.
- (12) Edwards, J. P.; West, S. J.; Pooley, C. L. F.; Marschke, K. B.; Farmer, L. J.; et al. New nonsteroidal androgen receptor modulators based on 4-(trifluoromethyl)-2(1H)-pyrrolidino[3,2-g]quinolinone. *Bioorg. Med. Chem. Lett.* **1998**, *8*, 745-750.
- (13) Hamann, L. G.; Mani, N. S.; Davis, R. L.; Wang, X. N.; Marschke, K. B.; et al. Discovery of a potent, orally active, nonsteroidal androgen receptor agonist: 4-ethyl-1,2,3,4-tetrahydro-6-(trifluoromethyl)-8-pyridono[5,6-g]-quinoline (LG121071). *J. Med. Chem.* **1999**, *42*, 210-212.
- (14) Edwards, J. P.; Higuchi, R. I.; Winn, D. T.; Pooley, C. L. F.; Caferro, T. R.; et al. Nonsteroidal androgen receptor agonists based on 4-(trifluoromethyl)-2H-pyran[3,2-g]quinolin-2-one. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 1003-1008.
- (15) Zhi, L.; Tegley, M. T.; Marschke, K. B.; Jones, T. K. Switching androgen receptor antagonists to agonists by modifying C-ring substituents on piperidino[3,2-g]quinolinone. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 1009-1012.
- (16) Higuchi, R. I.; Edwards, J. P.; Caferro, T. R.; Ringgenberg, J. D.; Kong, J. W.; et al. 4-Alkyl- and 3,4-dialkyl-1,2,3,4-tetrahydro-8-pyridono[5,6-g]quinolines: potent, nonsteroidal androgen receptor agonists. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 1335-1340.
- (17) Cockshott, I. D.; Cooper, K. J.; Sweetmore, D. S.; Blacklock, N. J.; Denis, L. The pharmacokinetics of casodex in prostate cancer patients after single and during multiple dosing. *Eur. Urol.* **1990**, *18*, 10-17.
- (18) Teutsch, G.; Goubet, F.; Battmann, T.; Bonfils, A.; Bouchoux, F.; et al. Non-steroidal antiandrogens: synthesis and biological profile of high-affinity ligands for the androgen receptor. *J. Steroid Biochem. Mol. Biol.* **1994**, *48*, 111-119.
- (19) Negro-Vilar, A. Selective androgen receptor modulators (SARMs): a novel approach to androgen therapy for the new millennium. *J. Clin. Endocrinol. Metab.* **1999**, *84*, 3459-3462.
- (20) Tucker, H.; Crook, J. W.; Chesterson, G. J. Nonsteroidal antiandrogens. Synthesis and structure-activity relationships of 3-substituted derivatives of 2-hydroxypropionamides. *J. Med. Chem.* **1988**, *31*, 954-959.
- (21) Morris, J. J.; Hughes, L. R.; Glen, A. T.; Taylor, P. J. Nonsteroidal antiandrogens. Design of novel compounds based on an infrared study of the dominant conformation and hydrogen-bonding properties of a series of anilide antiandrogens. *J. Med. Chem.* **1991**, *34*, 447-455.
- (22) Hamann, L. G.; Higuchi, R. I.; Zhi, L.; Edwards, J. P.; Wang, X. N.; et al. Synthesis and biological activity of a novel series of nonsteroidal, peripherally selective androgen receptor antagonists derived from 1,2-dihydropyridono[5,6-g]quinolines. *J. Med. Chem.* **1998**, *41*, 623-639.
- (23) Burden, P. M.; Ai, T. H.; Lin, H. Q.; Akinci, M.; Costandi, M.; et al. Chiral derivatives of 2-cyclohexylideneperhydro-4,7-methanoindenes, a novel class of nonsteroidal androgen receptor ligand: synthesis, X-ray analysis, and biological activity. *J. Med. Chem.* **2000**, *43*, 4629-4635.
- (24) Kirkovsky, L.; Mukherjee, A.; Yin, D.; Dalton, J. T.; Miller, D. D. Chiral nonsteroidal affinity ligands for the androgen receptor. 1. Bicalutamide analogues bearing electrophilic groups in the B aromatic ring. *J. Med. Chem.* **2000**, *43*, 581-590.
- (25) Van Dort, M. E.; Robins, D. M.; Wayburn, B. Design, synthesis, and pharmacological characterization of 4-[4,4-dimethyl-3-(4-hydroxybutyl)-5-oxo-2-thioxo-1-imidazolidinyl]-2-iodobenzonitrile as a high-affinity nonsteroidal androgen receptor ligand. *J. Med. Chem.* **2000**, *43*, 3344-3347.
- (26) Van Dort, M. E.; Jung, Y. W. Synthesis and structure-activity studies of side-chain derivatized arylhydantoin for investigation as androgen receptor radioligands. *Bioorg. Med. Chem. Lett.* **2001**, *11*, 1045-1047.
- (27) Yin, D.; He, Y.; Perera, M. A.; Hong, S. S.; Marhefka, C.; et al. Key structural features of nonsteroidal ligands for binding and activation of the androgen receptor. *Mol. Pharmacol.* **2003**, *63*, 211-223.
- (28) He, Y.; Yin, D.; Perera, M.; Kirkovsky, L.; Stourman, N.; et al. Novel nonsteroidal ligands with high binding affinity and potent functional activity for the androgen receptor. *Eur. J. Med. Chem.* **2002**, *37*, 619-634.
- (29) Klebe, G.; Abraham, U.; Mietzner, T. Molecular similarity indices in a comparative analysis (CoMSIA) of drug molecules to correlate and predict their biological activity. *J. Med. Chem.* **1994**, *37*, 4130-4146.
- (30) Sippl, W. Binding affinity prediction of novel estrogen receptor ligands using receptor-based 3-D QSAR methods. *Bioorg. Med. Chem.* **2002**, *10*, 3741-3755.
- (31) Wolohan, P.; Reichert, D. E. CoMFA and docking study of novel estrogen receptor subtype selective ligands. *J. Comput.-Aided Mol. Des.* **2003**, *17*, 313-328.
- (32) Tervo, A. J.; Nyrönen, T. H.; Rönkkö, T.; Poso, A. Comparing the quality and predictiveness between 3D QSAR models obtained from manual and automated alignment. *J. Chem. Inf. Comput. Sci.* **2004**, *44*, 807-816.
- (33) He, B.; Kempainen, J. A.; Wilson, E. M. FXXLF and WXXLF sequences mediate the NH₂-terminal interaction with the ligand binding domain of the androgen receptor. *J. Biol. Chem.* **2000**, *275*, 22986-22994.
- (34) He, B.; Minges, J. T.; Lee, L. W.; Wilson, E. M. The FXXLF motif mediates androgen receptor-specific interactions with coregulators. *J. Biol. Chem.* **2002**, *277*, 10226-10235.
- (35) Jones, G.; Willett, P.; Glen, R. C.; Leach, A. R.; Taylor, R. Development and validation of a genetic algorithm for flexible docking. *J. Mol. Biol.* **1997**, *267*, 727-748.
- (36) Poujol, N.; Wurtz, J.-M.; Tahiri, B.; Lumbroso, S.; Nicolas, J.-C.; et al. Specific recognition of androgens by their nuclear receptor. *J. Biol. Chem.* **2000**, *275*, 24022-24031.
- (37) Wang, R.; Lai, L.; Wang, S. Further development and validation of empirical scoring functions for structure-based binding affinity prediction. *J. Comput.-Aided Mol. Des.* **2002**, *16*, 11-26.
- (38) Clark, R. D.; Strizhev, A.; Leonard, J. F.; Matthew, J. B. Consensus scoring for ligand/protein interactions. *J. Mol. Graphics Modell.* **2002**, *20*, 281-295.
- (39) Lehtonen, J. V.; Still, D.-J.; Rantanen, V.-V.; Ekholm, J.; Björklund, D.; et al. BODIL: a molecular modeling environment for structure-function analysis and drug design. *J. Comput.-Aided Mol. Des.* **2004**, *18*, 401-419.
- (40) Marhefka, C. A.; Gao, W.; Chung, K.; Kim, J.; He, Y.; et al. Design, synthesis, and biological characterization of metabolically stable selective androgen receptor modulators. *J. Med. Chem.* **2004**, *47*, 993-998.
- (41) Verdonk, M. L.; Cole, J. C.; Taylor, R. SuperStar: a knowledge-based approach for identifying interaction sites in proteins. *J. Mol. Biol.* **1999**, *289*, 1093-1108.
- (42) Verdonk, M. L.; Cole, J. C.; Watson, P.; Gillet, V.; Willett, P. SuperStar: improved knowledge-based interaction fields for protein binding sites. *J. Mol. Biol.* **2001**, *307*, 841-859.
- (43) Berman, H. M.; Westbrook, J.; Feng, Z.; Gilliland, G.; Bhat, T. N.; et al. The Protein Data Bank. *Nucleic Acids Res.* **2000**, *28*, 235-242.
- (44) Sybyl 6.9.1; Tripos Inc.: St. Louis, MO, 2003.
- (45) Sadowski, J.; Gasteiger, J. From atoms and bonds to three-dimensional atomic coordinates: model builders. *Chem. Rev.* **1993**, *93*, 2567-2581.
- (46) Sadowski, J.; Gasteiger, J.; Klebe, G. Comparison of automatic three-dimensional model builders using 639 X-ray structures. *J. Chem. Inf. Comput. Sci.* **1994**, *34*, 1000-1008.
- (47) Purcell, W. P.; Singer, J. A. A brief review and table of semiempirical parameters used in the Hückel molecular orbital method. *J. Chem. Eng. Data* **1967**, *12*, 235-246.
- (48) Gasteiger, J.; Marsili, M. Iterative partial equalization of orbital electronegativity: a rapid access to atomic charges. *Tetrahedron* **1980**, *36*, 3219-3222.
- (49) Cramer, R. D., III; Bunce, J. D.; Patterson, D. E. Crossvalidation, bootstrapping, and partial least squares compared with multiple regression in conventional QSAR studies. *Quant. Struct.-Act. Relat.* **1988**, *7*, 18-25.

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and accessory sex organ weights in rats (chronic test) [19]. This paper presents the results obtained with 3 compounds, which were selected on the basis of their high RBAs for the AR. Their biological profiles were compared to those of known non-steroidal antiandrogens. Two compounds were investigated more extensively: RU 59063 which, in its tritiated form, is a good candidate as a specific, high-affinity AR marker, and RU 56187 which displayed potent antiandrogenic activities *in vivo*.

EXPERIMENTAL

Synthesis of *N*-substituted arylthiohydantoin

Melting points were determined on a Kofler hot stage apparatus and are uncorrected. Spectral data were recorded on the following spectrometers: IR, Nicolet 5 SX, in chloroform solution unless stated otherwise; NMR, Bruker AC or AM in CDCl₃ with TMS as an internal standard; UV, Perkin Elmer Lambda 9 in ethanol. Microanalyses were in agreement with calculated values ($\pm 0.3\%$) for the elements cited. Chromatographic purifications were performed using 50–100 parts (w/w) of Merck 60 silica gel (0.04–0.063 mm). All reactions were conducted under a nitrogen atmosphere.

2-Alkylamino-2-cyano-propanes 1a–1c

These compounds were prepared according to the general procedure of Exner *et al.* [20] by slow addition of the relevant amine to neat acetone cyanohydrin. They were not isolated, but best used directly for the transformation 3→4.

2-Trifluoromethyl-4-isothiocyanato-benzonitrile 3

The aniline 2 [21] (2.23 g, 12 mmol) was added over 10 min to a well-stirred heterogeneous mixture of thiophosgene (1 ml, 13 mmol) in water (22 ml) at room temperature (RT). The stirring was continued for 1 h, leading to a progressive decolourization of the medium. The reaction mixture was extracted with chloroform (3 × 10 ml), dried over magnesium sulphate and evaporated to dryness under reduced pressure. The crude desired compound was obtained as a brownish syrup (3 g, >100%) and was used as such for the next step. IR: 2017 cm⁻¹ (N = C = S), 2224 and 2210 cm⁻¹ (CN), 1609, 1562 and 1497 cm⁻¹ (aromatics). An analytical sample can be obtained by chromatography (toluene–cyclohexane, 1:1): m.p. 40°C; UV: λ max = 235 nm ($\epsilon = 15100$) and 309 nm ($\epsilon = 29300$); analysis: C, H, F, N, S.

4-(5-Imino-2-thioxo-3,4,4-trimethyl-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile 4a

To a solution of the crude isothiocyanate 3 (3 g, 12 mmol) in THF (23 ml) and triethylamine (0.23 ml) was added 2-methylamino-2-cyanopropane (1.15 g, 11.7 mmol). The reaction mixture was refluxed for 40 min and then evaporated to dryness under reduced pressure. The residue was purified by chroma-

tography (CH₂Cl₂–acetone, 95:5) and recrystallized from isopropanol, yielding 2.629 g (66%) of the desired imine 4a. m.p.: 173–174°C; UV: λ max = 233 nm ($\epsilon = 20500$), 256 nm ($\epsilon = 24100$); ¹H-NMR: $\delta = 1.59$ (s, 6H, gem-diMe), 3.29 (s, 3H, NMe), 7.76 (d, 1H, H-5), 7.88 (d, 1H, H-3) and 7.98 (d, 1H, H-6); analysis: C, H, F, N, S.

The following compounds were prepared by the same procedure using the relevant cyanoamines: 4-[4,4-dimethyl-3-(2-hydroxyethyl)-5-imino-2-thioxo-1-imidazolidinyl]-2-trifluoromethyl-benzonitrile 4b. Yield = 75%; m.p.: 181°C; UV: λ max = 233 nm ($\epsilon = 20300$), 259 nm ($\epsilon = 23700$); TLC (SiO₂, Merck F₂₅₄, CH₂Cl₂–acetone, 6:4) R_f = 0.27; analysis: C, H, F, N, S. 4-[4,4-Dimethyl-3-(4-hydroxybutyl)-5-imino-2-thioxo-1-imidazolidinyl]-2-trifluoromethyl-benzonitrile 4c. Yield = 61.5%; amorphous solid; TLC (SiO₂, Merck F₂₅₄, CH₂Cl₂–acetone, 6:4) R_f = 0.25; ¹H-NMR: $\delta = 1.59$ (s, 6H, gem-diMe), 1.65 and 1.94 (m, 2 × 2H, the protons on C-2 and C-3 of the butyl chain), 3.72 (m, 4H, protons on C-1 and C-4 of the butyl chain)

4-(5-Oxo-2-thioxo-3,4,4-trimethyl-1-imidazolidinyl)-2-trifluoromethyl-benzonitrile 5a

The imine 4a (2.207 g, 6.76 mmol) was suspended in 6N aqueous HCl (44 ml) and heated to reflux temperature for 1 h. The reaction mixture was poured onto ice-water (1:1, 20 g) and extracted with methylene chloride (4 × 10 ml). The combined organic extracts were dried over magnesium sulphate, evaporated to dryness and recrystallized from isopropanol, affording 2.10 g (95%) of 5a. m.p.: 171°C; UV: λ max = 231 nm ($\epsilon = 18600$), 252 nm ($\epsilon = 23800$); ¹H-NMR: $\delta = 1.58$ (s, 6H, gem-diMe), 3.32 (s, 3H, NMe), 7.78 (d, d, 1H, H-5), 7.91 (d, 1H, 3-H) and 7.96 (d, 1H, H-6); analysis: C, H, F, N, S.

4-[4,4-Dimethyl-3-(2-hydroxyethyl)-5-oxo-2-thioxo-1-imidazolidinyl]-2-trifluoromethyl-benzonitrile 5b

2N aqueous hydrochloric acid (10 ml, 20 mmol) was added to a solution of the imine 4b (4.6 g, 12.7 mmol) in methanol (65 ml) and refluxed for 1 h. The reaction mixture was cooled to RT, poured into cold water (300 ml) and extracted with ethyl acetate (3 × 30 ml). The organic fraction was dried over magnesium sulphate and evaporated under reduced pressure. The residue was purified by chromatography (cyclohexane–ethyl acetate, 1:1) and recrystallized from CH₂Cl₂–cyclohexane, affording 4.37 g (95%) of 5b. m.p.: 130°C; UV: λ max = 232 nm ($\epsilon = 18700$), 254 nm ($\epsilon = 22700$); ¹H-NMR: $\delta = 1.61$ (s, 6H, gem-diMe), 2.16 (s, 1H, OH), 3.94 (m, 2H, N-CH₂), 4.05 (m, 2H, CH₂-O); analysis: C, H, F, N, S.

4-[4,4-Dimethyl-3-(4-hydroxybutyl)-5-oxo-2-thioxo-1-imidazolidinyl]-2-trifluoromethyl-benzonitrile 5c

Proceeding as for the preparation of 5b, compound 5c was obtained in 78.7% yield. m.p.: 78–79°C; UV: λ max = 232 nm ($\epsilon = 19500$), 254 nm ($\epsilon = 24000$);

$^1\text{H-NMR}$: $\delta = 1.60$ (s, 6H), 1.67 (m, 2H), 1.96 (m, 2H), 3.72 (m, 4H), 7.78 (d, 1H), 7.90 (m, 1H) and 7.96 (m, 1H); analysis: C, H, F, N, S.

Animals

Male Sprague-Dawley rats and Swiss mice were purchased from Iffa Credo (France). Male golden Syrian hamsters were obtained from Charles River (U.S.A.) and female New Zealand rabbits from ESD (France).

Compounds

[6,7- ^3H]R 1881 (sp. act.: 57 Ci/mmol), [1,2- ^3H]testosterone (sp. act.: 54 Ci/mmol) and [5- ^3H]RU 59063 (sp. act.: 25 Ci/mmol) were prepared by RU radiomolecules Lab. L[2,3- ^3H]ornithine (sp. act.: 46.5 Ci/mmol) was obtained from NEN.

Testosterone, dihydrotestosterone (DHT), R 1881, Anandron (nilutamide = RU 23908), Casodex (ICI 176,334), hydroxyflutamide, cyproterone acetate, triamcinolone acetonide and RU 28362 were synthesized according to described procedures.

Homogenization and assay buffers

Buffer A: 10 mM Tris-HCl, pH 7.4, 0.25 M sucrose; buffer B: buffer A + 2 mM dithiothreitol; buffer C: buffer B + 0.1 mM phenylmethylsulphonyl fluoride and 20 mM sodium molybdate; buffer D: 50 mM Tris-HCl, pH 7.4, 5 mM dithiothreitol, 0.25 mM pyridoxal phosphate, 0.1 mM EDTA.

Dextran coated charcoal (DCC) was prepared by suspending 1.25% Norit A charcoal and 0.625% dextran T80 in the buffer used for tissue homogenization.

RBA for steroid receptors and human plasma sex hormone binding globulin (SHBG)

The RBAs of test compounds were evaluated in a routine receptor screening described in detail previously [17]. It included the androgen (AR), progesterin (PR), glucocorticoid (GR), mineralocorticoid (MR) and estrogen (ER) receptors. AR, PR, GR, MR and

ER were prepared respectively from prostate of 1 day castrated rats (180–200 g), uterus of immature estradiol-primed rabbits (≈ 1 kg), thymus of adrenalectomized (ADX) male rats (≈ 160 g), kidney of ADX rats (160 g), uterus of 18-day-old mice. All subsequent procedures were carried out at 0–4°C.

Prostates were homogenized in buffer C (1:8, w/v), rabbit uterus (1:50, w/v), mouse uterus (1:25, w/v), kidney (1:3, w/v) in buffer A and thymus (1:10, w/v) in buffer B with a glass-glass Porter.

The homogenates were centrifuged at 209,000 g for 30 min. 125 μl aliquots of the supernatant (cytosol) were incubated for 24 h (5 h at 25°C for ER) with 2.5 or 5 nM of the relevant ^3H ligand [[^3H]testosterone ([^3H]T) for AR, [^3H]R 5020 for PR, [^3H]dexamethasone for GR, [^3H]aldosterone with 10^{-6} M RU 28362 [22] for MR and [^3H]estradiol for ER] in the presence of increasing concentrations (1–25,000 nM) of cold reference or test compounds.

Plasma was prepared from human female blood, diluted (1:100) with buffer A and 125 μl aliquots were incubated for 24 h with 20 nM of [^3H]T in the presence of increasing concentrations of cold T or RU 59063.

Bound ^3H ligand measurement by DCC adsorption technique: a 0.1 ml aliquot of incubated cytosol or plasma was stirred for 10 min with 0.1 ml DCC solution in a 96 well microtitre plate and centrifuged for 10 min at 800 g. The radioactivity of a 0.1 ml supernatant sample was counted.

RBA calculation: the RBA was defined as the ratio of the concentration of the reference compound over the concentration of the competitor required to inhibit ^3H ligand binding by 50% and multiplied by 100. The RBAs of T, progesterone, dexamethasone, aldosterone and estradiol were taken equal to 100.

Binding to AR of various species and to human plasma

AR was prepared from the following target tissues of 1 day castrated animals: cytosols of rat prostate (1:5, w/v), hamster (140–160 g) prostate and flank organs

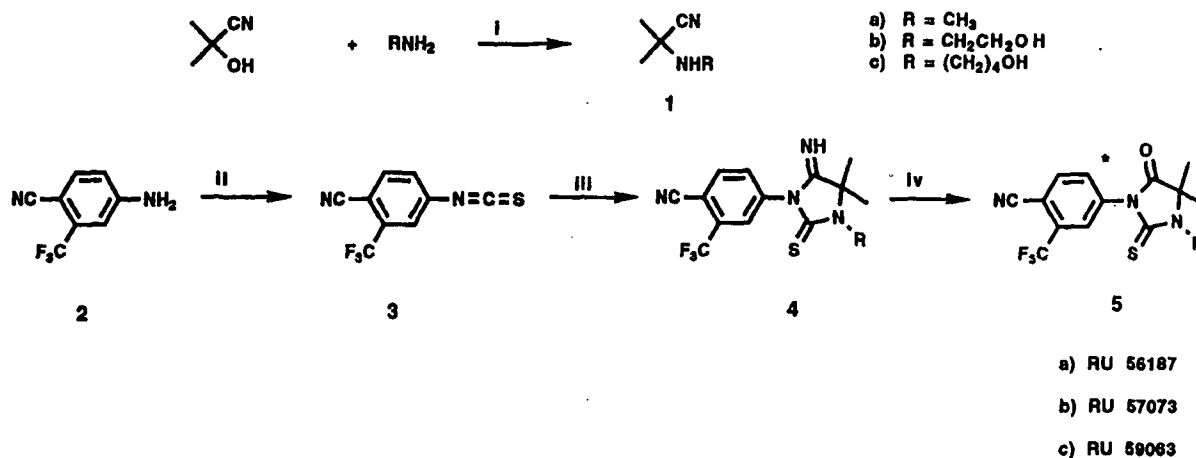


Fig. 1. Synthesis of *N*-substituted arylthiohydantoin and structures of the most representative compounds of this series. (i) Neat, room temperature (RT); (ii) CCl₄, H₂O, RT; (iii) 1, NEt₃, THF, RT; (iv) 2N HCl, MeOH, reflux. *Site of labelling in [^3H]RU 59063.

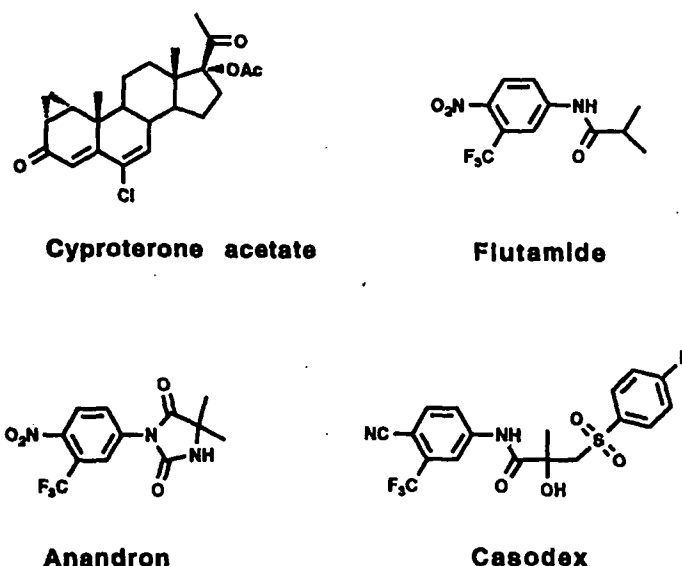


Fig. 2. Chemical structures of antiandrogens commonly used in therapeutics.

(1:5, w/v) and male mouse (30 g) kidney (1:3, w/v) were prepared in buffer C, as described above, except that flank organs were frozen in liquid nitrogen and pulverized in a mortar before homogenization.

The human AR (hAR) and a truncated hAR (htAR) were obtained as follows: hAR cDNA was cloned from T47D cells and inserted in the eucaryotic expression vector pSG5 (Prof. Chambon, LGME, Strasbourg, unpublished data). The chimera GAL4-htAR was obtained by inserting in the XhoI site of the expression vector pGAL4M polyII [23] a PCR-amplified fragment containing the D-E regions of hAR (amino acids 625 to 920). These receptors were expressed in COS-1 cells as follows: cells were plated at 2×10^6 cells/9 cm dish in Dulbecco's modified Eagle's medium (Gibco) containing 5% DCC-treated foetal calf serum and transfected with 5 μ g plasmid DNA/dish using the calcium phosphate precipitation technique. Cells were harvested about 40 h after transfection and the cytosols were prepared as described above.

0.1 ml aliquots of cytosols or human plasma (1:100) were incubated with concentrations (0.1 to 20 nM) of [3 H]T or [3 H]RU 59063 for 24 h at 0°C. Non-specific binding was determined in parallel incubations with the 3 H ligand in the presence of a 500-fold excess of the corresponding cold compound. Bound radioactivity

was determined as described above. Scatchard analysis [24] was used to determine the concentration of binding sites N (fmol/mg protein) and the association constants K_d .

In vivo studies

TP and the test compounds were administered by subcutaneous route (s.c.) in sesame oil containing 5% benzyl alcohol or in corn oil containing 10% ethanol. Orally they were given in an aqueous solution containing 0.5% methylcellulose.

Mouse renal ODC activity. Male mice weighing about 30 g (3 to 6 animals) were castrated. 24 h later, they received simultaneously a single dose of the test compound by oral or s.c. route and 3 mg/kg of TP by s.c. route. 16 h later, the animals were sacrificed, the kidneys removed, pooled and homogenized in buffer D at 4°C. The cytosols were prepared as described above. ODC activity was evaluated according to Djurhuus [25] with slight modifications. Briefly aliquots (triplicate) of 50 μ l of cytosol were incubated with 0.025 μ Ci of [3 H]L-ornithine and 10 mM of cold L-ornithine for 1 h at 37°C. Samples were then applied to dry Whatman paper strips and washed 3 times with 0.1 M ammonia in order to eliminate L-ornithine. [3 H]Putrescine retained by the paper strip was counted. The results were expressed as fmol of

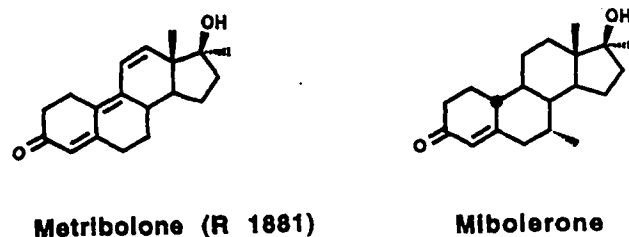


Fig. 3. Structures of the synthetic androgens widely used as AR markers.

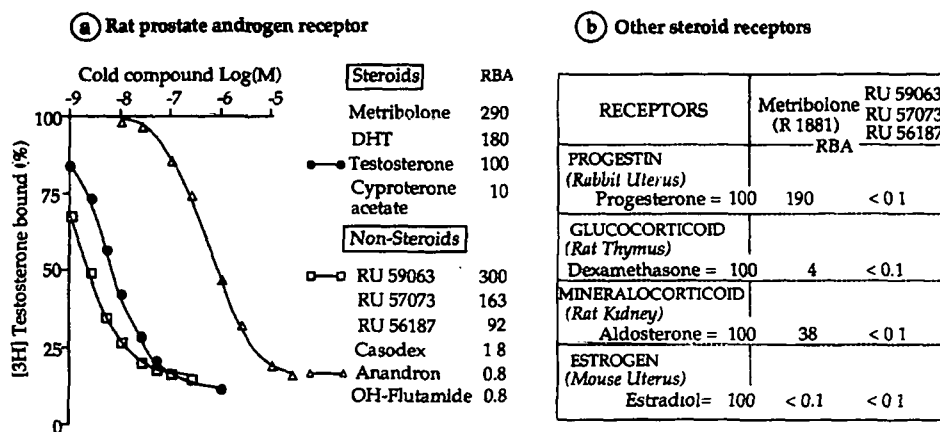


Fig. 4. RBAs for the 5 classical steroid receptors.

[³H]putrescine formed per mg of protein per hour. The proteins were determined according to the method of Bradford [26].

Accessory sex organ weight in rats. Immature male rats weighing about 100 g were castrated. 24 h later, groups of 5 animals received daily (8 administrations over 10 days, as indicated in Fig. 8) simultaneously 0.5 mg/kg of TP by s.c. route and the test compounds either by oral or s.c. route. 24 h after the last administration the animals were sacrificed, the seminal vesicles and the prostate removed and fixed for 72 h in demineralized water containing 10% formaldehyde. Then the organs were carefully dissected and weighed.

Groups of 5 adult male rats weighing about 200–220 g received daily increasing doses of RU 56187 by oral route according to the scheme of administration described above.

RESULTS AND DISCUSSION

RBAs for the steroid receptors

The RBAs of several *N*-substituted arylthiohydantoin, (see synthetic scheme in Fig. 1), were determined on a routine receptor screening [17] and

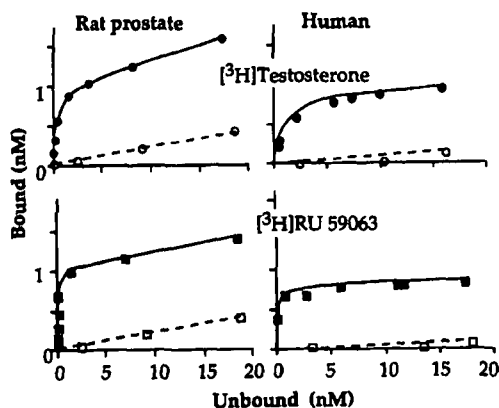


Fig. 5. Saturation curves of [³H]T and [³H]RU 59063 binding to rat prostate and hAR androgen receptors. (●, ■) Total binding; (○, □) non-specific binding.

compared to those of cyproterone acetate (CPA), Casodex, Anandron and hydroxyflutamide, the active metabolite of flutamide (see structures in Fig. 2) and to that of R 1881, one of the two compounds (see structures in Fig. 3) with mibolone (MB), widely used as AR markers [27–29]. As shown in Fig. 4, RU 59063, RU 57073 and RU 56187 exhibit exceptionally high RBAs for the rat prostate AR. The compound with the highest affinity, RU 59063, inhibits in a concentration-dependent manner the binding of [³H]T to rat AR. Its RBA is similar to that of R 1881, and respectively 1.7, 3, 30, 166 and 375 times higher than those of DHT, T, CPA, Casodex and Anandron or hydroxyflutamide. The RBAs obtained for these latter compounds are in good agreement with those published elsewhere [1, 13, 16].

As shown in the table inserted in Fig. 4, these three new non-steroidal compounds are totally devoid of any binding to other steroid receptors up to 25 μM whereas R 1881 exhibits, respectively strong, moderate and weak RBAs for the rabbit uterus PR, rat kidney MR and rat thymus GR. The latter data are consistent with those published previously [17, 30]. Furthermore MB, has been shown to possess a slightly higher RBA than that of R 1881 for AR and, respectively a 2, 4, and 7 times lower one for PR, MR and GR [31]. The very promising biochemical profile of RU 59063, led us to label it in order to evaluate its binding parameters for the AR of various species.

Binding parameters of [³H]RU 59063 and [³H]T for AR of various species

The binding parameters (number of binding sites, *N*, and association constant, *K_a*) of RU 59063 and T were determined in the cytosol of target tissues of species commonly used to test androgens or antiandrogens: rat prostate, hamster flank organ and prostate, mouse kidney. They were also evaluated in the cytosol of COS cells in which the full-length (hAR) or a truncated (htAR) human AR were expressed. As illustrated in Fig. 5, incubation with [³H]RU 59063, or [³H]T, show saturable binding of rat prostate and COS

Table 1. Binding parameters (N , K_a) of [^3H]T and [^3H]RU 59063 for the AR of different species

Binding parameters	Compounds	Rat prostate	Mouse kidney	Hamster		Human	
				Prostate	F.O.*	Full length	Truncated
N (fmol/mg protein)	Testosterone	219	20	17	132	367	820
	RU 59063	226	24	15	145	327	920
K_a (10^9 M^{-1})	Testosterone	1.9	1.2	1.3	0.9	0.8	0.5
	RU 59063	6.1	1.6	4.3	7.2	6.2	6.2

*F.O., flank organ.

cells (hAR) cytosols. Furthermore its non-specific binding, like that of [^3H]T, is low. This characteristic is a necessary requirement for a receptor marker. N and K_a were deduced from these binding data using the Scatchard analysis and are shown in Table 1. RU 59063 and T gave rise to the same number of binding sites whatever the target tissues. RU 59063 exhibited a very high K_a , from 3 to 13 times higher than that of T, according to the tissue. The reported K_a values (10^9 M^{-1}), are the mean of 3 determinations for rat prostate AR [$K_a(\text{RU}) = 4-8.4$ vs $K_a(\text{T}) = 1.7-2.1$], for hAR [$K_a(\text{RU}) = 5-7$ vs $K_a(\text{T}) = 0.7-1.2$] and for htAR [$K_a(\text{RU}) = 5-9$ vs $K_a(\text{T}) = 0.4-0.6$]. A single determination was performed for the other tissues. Whereas the K_a s of RU 59063 for rat AR, hAR and htAR are identical, those of T are different in spite of the fact that the amino acid sequences of the hormone binding domains of rat and human AR are strictly identical [32]. The origin of this discrepancy is under investigation. Preliminary results seem to indicate that it is not related to 5α -reductase activity.

In addition, the binding parameters of [^3H]RU 59063 for rat prostate cytosol were compared to those of [^3H]R 1881 and [^3H]T (data not shown). [^3H]R 1881 (0.1–20 nM) was incubated in the presence of a 100-fold excess of triamcinolone acetonide (TA) in order to block its binding to PR and GR [30]. Under these experimental conditions, the 3 ligands gave rise to

the same N : 196, 196 and 176 fmol/mg protein for RU 59063, R 1881 and T, respectively. However, the K_a of RU 59063 ($8.4 \times 10^9 \text{ M}^{-1}$) was higher than those of R 1881 ($6 \times 10^9 \text{ M}^{-1}$) and T ($2 \times 10^9 \text{ M}^{-1}$). Finally, the comparison of the K_a values of RU 59063 reported here with those published for R 1881 and MB, in the rat [30–34], mouse [35] and human [30, 32, 33, 36], confirms that RU 59063 is one of the most potent AR markers known so far and undoubtedly the most selective.

Binding to human plasma SHBG

One major problem in the assay of AR in human tissue with [^3H]DHT or [^3H]T is the interference by plasma proteins such as SHBG which binds these two hormones with a very high affinity [31]. It was therefore important to assess the binding of RU 59063 to human plasma. As depicted in Fig. 6 (left panel), it does not affect up to $2.5 \times 10^{-5} \text{ M}$, the binding of [^3H]T to human SHBG whereas cold T reduces it by 50% at a concentration close to 10^{-8} M . The RBA (<0.1%) of RU 59063 for SHBG is similar to the values reported for other non-steroidal antiandrogens [16], but weaker than those published for R 1881 and MB [30, 31]. Moreover, as illustrated in Fig. 6 (right panel), the incubation of [^3H]RU 59063 with diluted human plasma (1:100) gave only rise to a non-specific binding whereas [^3H]T, as expected, displayed a saturable binding.

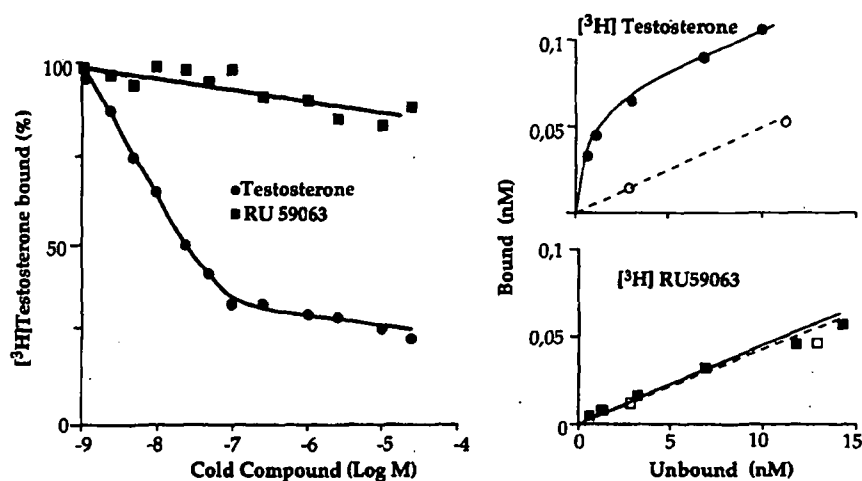


Fig. 6. Inhibition of [^3H]T binding to human plasma SHBG by cold T and RU 59063 (left panel) and saturation curves of [^3H]T and [^3H]RU 59063 binding to human plasma (right panel). (●, ■) Total binding; (○, □) Non-specific binding.

Table 2. Antiandrogenic activity on renal ODC in Cx mice and prostate weight in Cx immature rats

Compounds	Cx mouse Renal ODC		Cx rat Prostate weight	
	Inhibition of the effect of TP %			
	Dose: 1.7 mg/kg s.c. ^a	3 mg/kg p.o. ^b	1 mg/kg s.c.	1 mg/kg p.o.
RU 56187	65	94	69	60
RU 57073	30	67	37	45
RU 59063	64	—	21	23
Casodex	3 mg/kg 46	80	58	0
		64	28	0
Anandron	3 mg/kg 46	—	44	29
		—	10 mg/kg 67	10 mg/kg 59

^as.c., subcutaneously; ^bp.o., per orally.

All together, these biochemical studies suggest that RU 59063 is the most suitable ligand known so far for assaying the AR. It presents the following advantages over the existing steroidal ligands: unlike DHT and T, it is devoid of any specific binding to human plasma and unlike R 1881 and MB, it does not interact with other steroid receptors. The latter ligands exhibited a notably strong binding to the PR which is present in androgen-dependent tissues such as human prostate [30, 33]. In order to overcome this drawback, they are commonly used, for assaying the AR, in the presence of a large excess of cold TA or cold DHT [30, 33, 34]. But it has been reported that, in some cases, the inclusion of TA caused underestimation of AR whereas the use of DHT led to an overestimation [30].

In vivo antiandrogenic activity

The antiandrogenic activities of RU 56187, RU 57073, RU 59063, Anandron and Casodex were compared on a preliminary two-step bioassay, performed in male castrated animals supplemented with TP given s.c.: the renal ODC activity in mice (acute test), and the prostate weight assay in rats (chronic test).

In mice, a single injection of 3 mg/kg TP induced, 16 h later, a 50-fold increase in renal ODC activity relatively to vehicle-treated animals (528 ± 41 vs 10 ± 2 fmol putrescine/mg protein/h; $n = 15$). The androgen-specificity of this response was verified by injecting by s.c. route 30 mg/kg of the following compounds: estradiol, the pure glucocorticoid RU 28362 [37], the progestin R 5020 and aldosterone. None of these compounds were able to induce ODC activity (data not shown). In immature rats 8 day's treatment with TP induced an 8-fold increase in prostate weight relative to vehicle-treated animals (167 ± 8 vs 21.2 ± 3 mg; $n = 25$).

In these two models, the compounds were administered concomitantly with TP by s.c. or oral route, and the results obtained were expressed as the percentage of inhibition of the TP effect (TP-treated animals were arbitrarily assigned a value of 0 and vehicle-treated animals a value of 100%). At doses indicated in Table 2, RU 56187 exhibited the most potent antiandrogenic activity whatever the test and the route of administration. On the prostate weight, it proved to be about 3 and 10 times more active than Casodex and Anandron, respectively. This activity is not as high as expected from the differences in RBAs. Preliminary results suggest that, *in vivo*, RU 56187 is rapidly transformed to a low-affinity metabolite.

RU 56187 was selected from this preliminary screening and was studied in more detail by oral route on the 2 models mentioned above as well as in intact rats. As shown in Fig. 7, it antagonized dose-dependently the TP-induced ODC activity in Cx mice. It was fully effective at a dose close to 3 mg/kg while being devoid of agonistic activity when given alone at a dose of 10 mg/kg. On this test, its ED_{50} was about 0.6 mg/kg.

In Cx rats (Fig. 8), it inhibited in a dose-dependent manner the TP-induced accessory sex organ weight increase. It was fully effective on seminal vesicle (SV) and prostate weights at doses of 3 and 10 mg/kg, respectively with ED_{50} s of 0.6 mg/kg (SV) and 1 mg/kg (prostate).

RU 56187 also prevented the effect of endogenous androgens. As shown in Fig. 9, it caused a dose-related reduction of the accessory sex organ weights in intact adult rats with ED_{50} s of 1 mg/kg (SV) and 2.7 mg/kg (prostate). In a similar model, (14 days oral administration) the ED_{50} s reported for Casodex [14] were 2.5 mg/kg (SV) and 7.4 mg/kg (prostate).

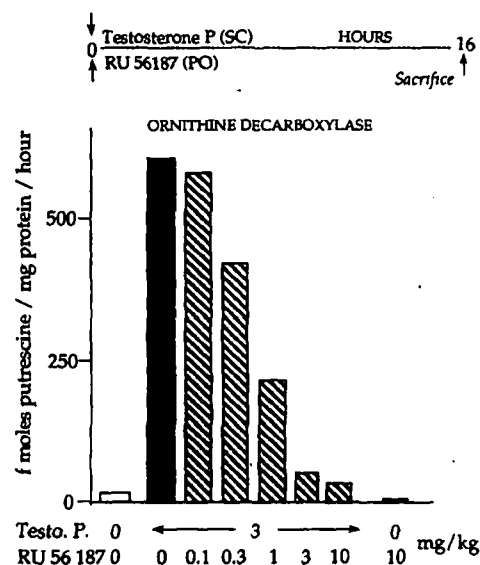


Fig. 7. Oral antiandrogenic activity of RU 56187 on TP-induced renal ODC activity in Cx mice.

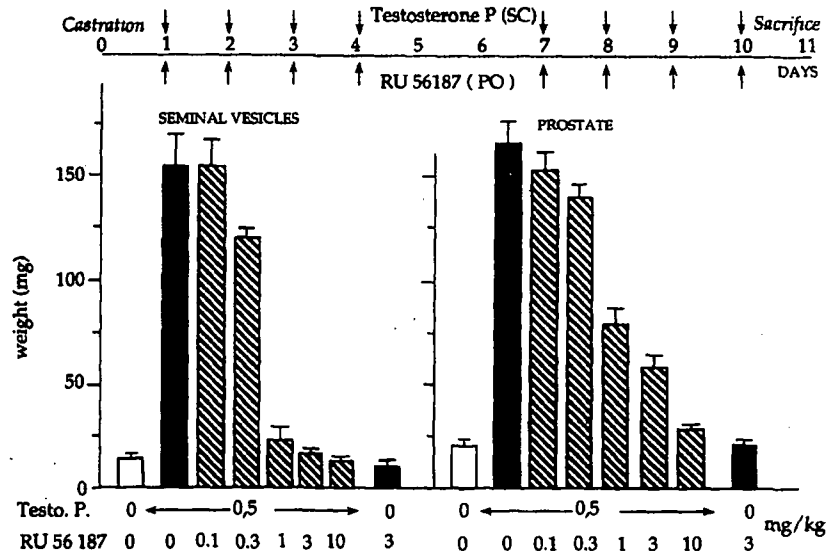


Fig. 8. Oral antiandrogenic activity of RU 56187 on TP-induced sex accessory organ weight in Cx immature rats.

CONCLUSION

The new arylthiohydantoin antiandrogens are characterized by exceptionally high RBAs for AR: higher than or close to those of the natural androgens T and DHT and 100 times higher than those of previously known non-steroidal antiandrogens. One of them, RU 59063, in its tritiated form, fulfills most of the criteria required for an ideal AR marker. Its K_d for the AR is at least equal to those of R 1881 (metribolone) and MB (mibolerone) and according to species, 3 to 13 times higher than that of T. Unlike existing steroid markers (R 1881, MB), it binds exclusively to AR and unlike T and DHT, it does

not interact with SHBG. *In vivo*, another analogue RU 56187 has proved to be the most potent pure antiandrogen known so far. In rats, when given orally, it is, respectively 3 and 10 times more active than Casodex and Anandron. Thus, this compound may be of potential use in the treatment of androgen-dependent diseases such as prostate cancer.

Further work is in progress to extend the usefulness of this new series of compounds, for instance in the area of γ and positron-emitting agents [38] and in the investigation of the mode of action of non-steroidal antiandrogens.

REFERENCES

- 1 Moguilewsky M and Bouton M. M. How the study of the biological activities of antiandrogens can be oriented towards the clinic. *J. Steroid Biochem.* 31 (1988) 699-710.
- 2 Neumann F.: Pharmacology and potential use of cyproterone acetate *Horm. Metab. Res.* 9 (1977) 1-13
- 3 Neumann F. and Topert M.: Pharmacology of antiandrogens *J. Steroid Biochem.* 25 (1986) 885-895
- 4 Neumann F. Pharmacology and clinical use of cyproterone acetate In *Pharmacology and Clinical Uses of Inhibitors of Hormone Secretion and Action* (Edited by B. J. A. Furr and A. E. Wakeling). Bailliere Tindall, London (1987) pp. 132-159
- 5 Mowszowicz I., Bieber D. E., Chung K. W., Bullock L. P. and Bardin C. W. Synandrogenic and antiandrogenic effect of progestins: comparison with non-progestational antiandrogens. *Endocrinology* 95 (1974) 1589-1599.
- 6 Neri R., Peets E. and Watnick A.: Anti-androgenicity of flutamide and its metabolite Sch 16423. *Biochem Soc Trans.* 7 (1979) 565-569.
- 7 Raynaud J. P., Bonne C., Moguilewsky M., Lefebvre F. A., Bélanger A. and Labrie F.: The pure antiandrogen RU 23 908 (Anandron)®, a candidate of choice for the combined antihormonal treatment of prostatic cancer: a review *The Prostate* 5 (1984) 299-311.
- 8 Neri R. and Kassem N.: Pharmacology and clinical uses of flutamide In *Pharmacology and Clinical Uses of Inhibitors of Hormone Secretion and Action* (Edited by B. J. A. Furr and A. E. Wakeling) Bailliere Tindall, London (1987) pp 160-169
- 9 Moguilewsky M, Bertagna C. and Hucher M. Pharmacological and clinical studies of the antiandrogen Anandron *J Steroid Biochem* 27 (1987) 871-875.

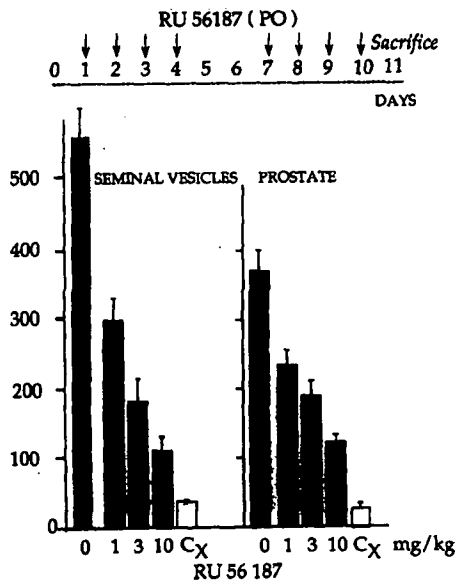


Fig. 9. Oral antiandrogenic activity of RU 56187 on sex accessory organ weight in adult intact rats.

10. Moguilewsky M., Cotard M., Proulx L., Tournemine C. and Raynaud J. P.: What is an antiandrogen and what is the physiological and pharmacological rationale for combined "castration" + antiandrogen" therapy. In *Prostate Cancer, Part A: Research, Endocrine Treatment, and Histopathology Progress in Clinical and Biological Research* (Edited by G. Murphy, S. Khoury, R. Küss, C. Chatelain and L. Denis). Alan Liss, New York, Vol. 243A (1987) pp. 315-340
11. Harris M. G., Coleman S. G., Faulds D. and Chrisp P.: Nilutamide. A review of its pharmacodynamic and pharmacokinetic properties, and therapeutic efficacy in prostate cancer *Drugs Aging* 3 (1993) 9-25
12. Furr B. J. A., Curry V. B., Woodburn J. R., Chesterson R. and Tucker H.: ICI 176, 334: a novel nonsteroidal peripherally selective antiandrogen. *J. Endocr.* 113 (1987) R7-R9.
13. Furr B. J. A.: "Casodex" (ICI 176,334)—A new, pure, peripherally-selective antiandrogen. preclinical studies. *Horm. Res.* 32 (1989) 69-76.
14. Tucker H.: Non-steroidal anti-androgens in the treatment of prostate cancer. *Drugs Future* 15 (1990) (3) 255-265
15. Mahler C. and Denis L. Clinical profile of a new non-steroidal antiandrogen. *J. Steroid Biochem Molec. Biol* 37 (1990) 921-924.
16. Ayub M. and Levell M. J.: The effect of ketoconazole related imidazole drugs and antiandrogens on [³H] R1881 binding to the prostatic androgen receptor and [³H] 5 α -dihydrotestosterone and [³H]cortisol binding to plasma proteins *J. Steroid Biochem.* 33 (1989) 251-255.
17. Ojasoo T. and Raynaud J. P.: Unique steroid congeners for receptor studies. *Cancer Res.* 38 (1976) 4186-4198.
18. Bullock L. P.: Androgen and progestin stimulation of ornithine decarboxylase activity in the mouse kidney. *Endocrinology* 112 (1983) 1903-1909.
19. Dorfman R. I.: Biological activity of antiandrogens. *Br. J. Dermatol* 82 (1970) 3-8.
20. Exner L. J., Luskin L. S. and De Benneville P. L.: α -(N-Alkylamino)-nitriles. *J. Am Chem. Soc.* 75 (1953) 4841-4842.
21. Alastair T. G. Amide derivatives, process for their manufacture, pharmaceutical or veterinary compositions containing them and a method of producing an antiandrogenic effect in a warm-blooded animal. European Patent, EP 0 002 892 (1979).
22. Ojasoo T., Moguilewsky M., Philibert D. and Raynaud J. P.: Study and assay of steroid hormone receptors. *Proc. Symp. Analysis of Steroids*, Sopron, Hungary (1987) 17-44.
23. Tora V., White J., Brou C., Tasset D., Webster V., Scheer E. and Chambon P.: The human estrogen receptor has two independent nonacidic transcriptional activation functions. *Cell* 59 (1989) 477-487
24. Scatchard G.: The attraction of protein for small molecules and ions. *Ann N.Y Acad. Sci.* 51 (1949) 660-672
25. Djurhuus R.: Ornithine decarboxylase (EC 4.1.1.17) assay based upon the retention of putrescine by a strong cation-exchange paper. *Analyt. Biochem.* 113 (1981) 352-355.
26. Bradford M. M.: A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Analyt. Biochem.* 72 (1976) 248-254.
27. Bonne C. and Raynaud J. P.: Methyltrienolone, a specific ligand for cellular androgen receptors. *Steroids* 26 (1975) 227-232.
28. Asselin J., Melancon R., Gourdeau Y., Labrie F., Bonne C. and Raynaud J. P.: Specific binding of [³H]methyltrienolone to both progestin and androgen binding components in human benign prostatic hypertrophy. *J. Steroid Biochem.* 10 (1979) 483-486
29. Schilling K. and Liao S.: The use of radioactive 7 α ,17 α -dimethyl-19-nortestosterone (mibolerone) in the assay of androgen receptor. *The Prostate* 5 (1984) 581-588.
30. Stebbings W. S. L., Anderson E., Puddefoot J. R., Farthing M. J. G. and Vinson G. P.: A hybrid ligand method for androgen receptor measurement. *J. Steroid Biochem.* 31 (1988) 181-186.
31. Carlson K. E. and Katzenellenbogen J. A.: A comparative study of the selectivity and efficiency of target tissue uptake of five tritium-labeled androgens in rat. *J. Steroid Biochem.* 36 (1990) 549-561
32. Chang C., Kokontis J. and Liao S.: Structural analysis of complementary DNA and amino acid sequences of human and rat androgen receptors. *Proc. Natn. Acad. Sci. U.S.A.* 85 (1988) 7211-7215.
33. Trash A. M., Muller R. E. and Woutz H. H.: Binding of 7 α , 17 α -dimethyl-19-nortestosterone (Mibolerone) to androgen and progesterone receptors in human and animal tissues. *Endocrinology* 118 (1986) 1327-1333
34. Bannister P., Sheridan P. and Losowsky M. S.: Use of new radioactive ligand, 7 α , 17 α -dimethyl [17 α -methyl ³H] 19-nortestosterone for the estimation of androgen receptors in rat liver cytosol *J. Steroid Biochem* 23 (1985) 121-123.
35. Kemp C. J. and Drinkwater N. R.: Genetic variation in liver tumor susceptibility, plasma testosterone levels and androgen receptor binding in six inbred strains of mice. *Cancer Res* 49 (1989) 5044-5047.
36. Young C. Y. F., Qiu S., Prescott J. L. and Tindall D. J.: Overexpression of a partial human androgen receptor in *E. coli*: characterization of steroid binding, DNA binding, and immunological properties. *Molec. Endocr.* 4 (1990) 1841-1849
37. Teutsch G., Costerousse G., Deraedt R., Benzoni J., Fortin M. and Philibert D.: 17 α -alkynyl-11 β , 17-dihydroxyandrostane derivatives: a new class of potent glucocorticoids. *Steroids* 38 (1981) 651-665.
38. Liu A., Dence C. S., Welch M. J. and Katzenellenbogen J. A.: Fluorine-18-labeled androgens: radiochemical synthesis and tissue distribution studies on six fluorine-substituted androgens, potential imaging agents for prostatic cancer *J. Nucl. Med* 33 (1992) 724-734.



Preliminary Pharmacokinetics and Metabolism of Novel Non-steroidal Antiandrogens in the Rat: Relation of their Systemic Activity to the Formation of a Common Metabolite

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The non-steroidal antiandrogens, RU 58841 and RU 56187 are amongst the most active of a new series of *N*-substituted aryl hydantoins or thiohydantoins. Their pharmacokinetics and principal metabolic profiles have been evaluated in rat plasma after intravenous administration of a 10 mg/kg dose. Both compounds disappear relatively rapidly from the plasma (elimination half-life of the order of 1 h), but they form a common metabolite, the *N*-desalkyl derivative, RU 56279, which is eliminated much more slowly. The percentage transformations of each into RU 56279, estimated from the AUCs of the metabolite compared with the AUC obtained after administration of RU 56279 itself, were respectively 1% and 77%. In parallel, their *in vivo* activity, as well as that of their metabolites, was determined with respect to parameters related to systemic antiandrogenic effects (prostate and seminal vesicle weights). The results showed that: (1) the common metabolite, RU 56279, is clearly antiandrogenic; (2) there appears to be a relationship between the percentage formation of this metabolite and the systemic antiandrogenic activity of the compounds. Thus, the pharmacological profile of RU 58841 which displays a potent local antiandrogenic activity without systemic effects can be related to its very low propensity to form the *N*-desalkyl metabolite.

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INTRODUCTION

The two non-steroidal antiandrogens, RU 56187 and RU 58841, were selected from a series of Anandron-derived compounds designed for the potential treatment of androgen-dependent disorders. These compounds are hydantoin (RU 58841) or thiohydantoin (RU 56187) derivatives characterized by the presence of an alkyl group on the nitrogen in position 3 (Fig. 1); hydroxybutyl in the case of the former and methyl in the latter. Although closely related structurally they differ pharmacologically. RU 58841 is very active by topical route on the hamster flank organ, but shows little effect on either weights of accessory sex organs such as prostate, seminal vesicle or levels of testosterone in both hamster and rat by oral,

subcutaneous or topical route [1]. By contrast, RU 56187 displays very strong systemic antiandrogen activity whatever the model studied and the route of administration [2]. In order to explain this apparent dissociation and possibly to relate the extent of the systemic effects of these compounds to their respective metabolic profiles, a preliminary study of their pharmacokinetics and metabolism was undertaken. The study was carried out in the rat by intravenous route, which by definition eliminates all absorption phases and allows a direct comparison of metabolism. The relatively high dose used (10 mg/kg) was chosen so that after a single administration the compounds and their metabolites could readily be measured using HPLC with UV detection. The antiandrogenic activity of RU 56187 and its metabolites was subsequently evaluated after oral administration, this being the route of choice in the case of the use of this compound for the treatment of prostate cancer in man. Concerning RU

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58841 which is destined to be used topically for the treatment of acne, alopecia and hirsutism, its activity as well as that of its metabolites was evaluated using the subcutaneous route in order to mimic its complete passage through the skin.

MATERIALS AND METHODS

Chemicals

The hydantoin derivative, RU 58841, the thiohydantoin derivative, RU 56187 (structures: Fig. 1) and the putative metabolites, RU 56191, 56279, 58336 and 59416, were synthesised in the Chemistry Department at Roussel Uclaf (structures of metabolites: Fig. 2).

All solvents and reagents used were of HPLC or analytical grade. Acetonitrile (low UV) was obtained from J. T. Baker (Phillipsburg, NJ, U.S.A.). Methanol and tetrahydrofuran were obtained from Merck (Darmstadt, Germany). Water used for the preparation of buffers was purified with a Milli-Q system (Millipore, Bedford, MA, U.S.A.).

Stock solutions were prepared in methanol (40 µg/ml) and stored at 4°C.

Animals

Pharmacokinetic studies. Male Sprague-Dawley rats (4 in each group), weighing 200 g on average were used; they received a single i.v. (tail vein) dose in PEG 300 (2 ml/kg). They were killed by decapitation and the blood was collected in heparinized tubes (Li) and rapidly centrifuged (3000 g, 4°C, 15 min). Plasma analysis was carried out immediately (without congelation). Concerning the cutaneous treatment, the animals were shaved on the back, using an electric shaver, then they received a single cutaneous administration of the product dissolved in ethanol 99.9% (50 µl/rat).

Pharmacological studies. Male Sprague-Dawley rats weighing 100 g were purchased from Iffa Credo (L'Abresle, France), housed in our central animal facilities under a 12 h dark/light cycle and given food and water *ad libitum*. The animals were castrated under anesthesia with Imalgene 500 (Rhone Mérieux), 24 h later (day 1) they received 0.5 mg/kg of testosterone propionate (TP, Roussel Uclaf) alone or together with the compounds at a dose of 1 mg/kg, or vehicle. Animals were treated daily, from day 1-4 and from day 7-10. TP was administered by s.c. route in germ corn oil (Mayoland, Benedicta, France) containing 10% ethanol and the compounds either by oral route, in aqueous solution containing 0.5% methylcellulose (Colorcon) or by s.c. route, in germ corn oil containing 10% ethanol. The animals were sacrificed by exsanguination under anesthesia 24 h after the last treatment. The prostates and seminal vesicles were removed and fixed for 72 h in demineralized water containing 10% formol (Merck); they were then dissected and weighed.

Each group consists of 5 animals/dose. The antiandrogenic activity was expressed as a percentage of inhibition of the TP effect (TP-treated animals were arbitrarily assigned a value of 0% and vehicle-treated animals a value of 100%).

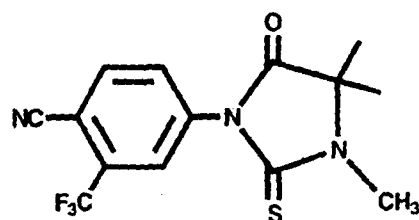
Statistical analysis

The means of the values and the standard error of the mean (SEM) were calculated for each dose. Statistical analysis were performed using Dunnett's [3] test. $P(*) < 0.01$ were considered to indicate a significant difference between the groups.

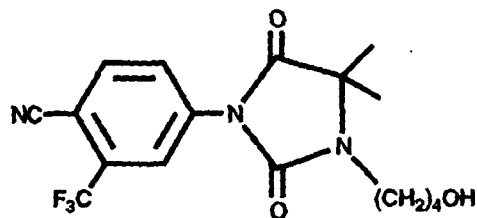
Analytical methods

Chromatography. The high-performance liquid chromatographic (HPLC) system consisted of a Hewlett Packard ternary pump model HP 1090 operated at a flow rate of 1 ml/min, a Spectra Physics SP 8780 autosampler equipped with a 20 µl loop, a Spectra Physics SP 8490 variable-wavelength Spectromonitor operated at 260 nm or a Perkin Elmer LC-235 diode array detector and a Spectra Physics SP 4290 integrator.

For RU 58841 studies, an Ultrabase Kromasil octyl analytical column (150 × 4.6 mm, 5 µm particle size) from Shandon Scientific-SFCC (Eragny, France) was used. In order to separate RU 58841 and its two metabolites, a mobile phase consisting of a 60/40 (v/v) mixture of 50 mM potassium dihydrogenophosphate



RU 56187



RU 58841

Fig. 1. Chemical structures of RU 56187 and RU 58841.

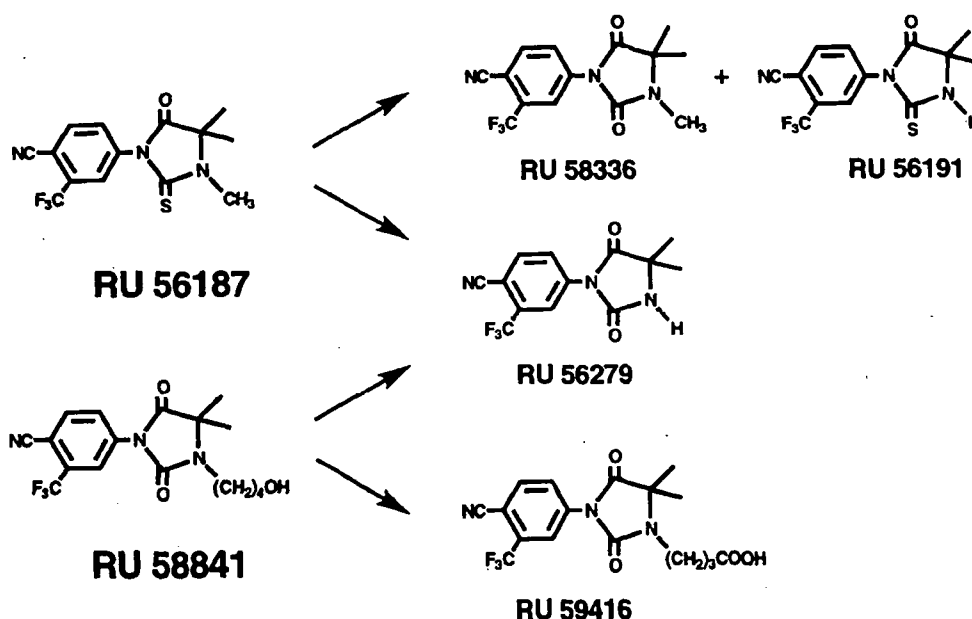


Fig. 2. Proposed principal metabolism scheme of RU 56187 and RU 58841 in the rat.

adjusted to pH 5.2 and tetrahydrofuran (= System I) was used.

RU 56187 and its metabolites were separated on a Nucleosil Cyano analytical column (150 × 4.6 mm, 5 μm particle size) from Colochrom (Gagny, France), with a mobile phase composed of acetonitrile and 0.1 M potassium phosphate buffer, pH 7.0; 35/65(v/v) (= System II).

Plasma sample extraction. A Vac Elut processing station SPS 24 (Analytichem) was used for extracting plasma samples. The complete method is described for RU 58841; for RU 56187, only the internal standard differs (compound of the same chemical series).

To 1 ml of plasma was added 0.8 μg (20 μg of a methanolic solution) of internal standard (RU 57073, compound of the thiohydantoin series). The mixture was vortex-mixed for 10 s, transferred onto octadecylsilane-bonded silica micro-columns (Bond Elut C18, 1 ml capacity, Analytichem International, Harbor City, CA, U.S.A.) and pretreated successively with 1 ml methanol and 1 ml purified water. Endogenous components in plasma were removed by washing the columns once with 1 ml of 1 M NaCl and 3 times with 1 ml purified water. The bound compounds were eluted with 1 ml methanol and 20 μl aliquots of this direct methanolic extract were injected into the chromatographic system. All the metabolites described here were extracted using this method.

Calibration and calculation. For both compounds and their metabolites, the limit of detection was evaluated as 0.005 μg/ml. Calibration curves were constructed by addition of known amounts of RU 58841 and its metabolites (0.010–8 μg/ml), together with the internal standard, to aliquots of control plasma (1 ml). The

calibration graphs were found to be linear over the range used, with correlation coefficients >0.998 and *y*-intercepts close to zero. Unweighted least-square regression lines were generated using peak-height ratios (RU compound/internal standard).

When plasma samples were spiked to final concentrations of 0.5 μg/ml, RU 58841 and its metabolite RU 56279 were totally recovered, the absolute recovery of the acid metabolite RU 59416 was 80%. The recovery of RU 56187, in the same conditions of concentration was 91%.

Identification of metabolites: analysis by mass-spectrometry/thermospray. The principal metabolites of RU 58841 and RU 56187 were tentatively identified by HPLC-SM (thermospray) as well as by the comparison of the peaks on the chromatograms with those of reference compounds.

All experiments were carried out on a Finnigan Model 4600 single quadrupole mass spectrometer equipped with a Data general Nova 4S computer using Superincos Software and fitted with a modified TSP II thermospray ionisation source (Finnigan Mat, San Jose, CA, U.S.A.). Typical operating conditions were as follows:

—Jet (source block) temperature, 230°C (Aerosol 245°C); vaporizer temperature, 105°C; repeller voltage, 10 V.

—For compounds tested, the sensitivity was found to be better when focusing on negative ions instead of positive ions. The negative thermospray profiles of standards and metabolites are relatively simple, with just one major ion predominating. The molecular ion (M-H⁻) is the base peak of the spectra and no

fragments of lower mass are observed. It is the mode used for the two metabolites of RU 58841 and for the two N-H metabolites of RU 56187. For RU 58336 (containing no mobile proton), the positive ionization mode was used (formation of $M + NH_4^+$). The structures of the 3 metabolites were confirmed by using a discharge electrode, to form M^- (1 kV).
 —Data were collected over the mass range of 150–500 amu using 1 s scans in the centroid mode.
 —The HPLC system connected to the inlet of the thermospray interface consisted of a Model SP 8700 pump (Spectra Physics), the solvent system in both cases was ammonium acetate 0.02 M in water/acetonitrile; the UV 260_{nm} elution profile was obtained on a Spectraflow 757 Kratos detector equipped with a special high pressure resistant cell to permit on line coupling with TSP mass spectrometer.

RESULTS

Pharmacokinetics and metabolism of the thiohydantoin, RU 56187, in the rat after i.v. administration of 10 mg/kg

Unchanged RU 56187. 5 min after injection, the plasma concentrations of RU 56187 are of the order of 2.4 $\mu\text{g/ml}$, they then decrease rapidly; at 6 h the mean value is 0.030 $\mu\text{g/ml}$. The experimental AUC (0–6 h) is equal to 1.58 $\mu\text{g/ml} \times \text{h}$ and the terminal elimination half-life is of the order of 1.4 h [Fig. 3(a)].

Principal metabolites (see Fig. 2). The first metabolite identified results from the oxidation of the thio function to give the corresponding hydantoin, RU 58336. This metabolism is very rapid; the maximum concentration (1.6 $\mu\text{g/ml}$) being observed at approx. 5 min. After 6 h its concentration is 0.4 $\mu\text{g/ml}$ and it is no longer detected beyond this time.

The principal metabolite is the *N*-demethylated hydantoin, RU 56279 (resulting from both oxidation and *N*-dealkylation): from 5 min, it is present at a mean concentration of 0.5 $\mu\text{g/ml}$ and the maximum appears at 6 h (mean concentration = 6 $\mu\text{g/ml}$). It decreases very slowly afterwards: 24 h after administration the mean concentration is 2.6 $\mu\text{g/ml}$ and after 72 h it is still present at a concentration of 0.7 $\mu\text{g/ml}$.

The *N*-demethylated thiohydantoin (RU 56191) was detected but not quantified; its concentrations at 4, 6 and 24 h were below or equal to 0.05 $\mu\text{g/ml}$.

The mass spectra of the three metabolites from a plasma extract are shown in Fig. 4 (a, RU 56279; b, RU 56191; c and d, RU 58336). They are in good agreement with those of the reference compounds.

Pharmacokinetics and metabolism of the hydantoin, RU 58841, in the rat after i.v. administration of 10 mg/kg

Unchanged RU 58841. 5 min after injection the plasma concentrations of RU 58841 are of the order of 6 $\mu\text{g/ml}$, they then decrease rapidly; at 6 h the mean value is 0.015 $\mu\text{g/ml}$. The experimental AUC (0–6 h) is equal to 3.61 $\mu\text{g/ml} \times \text{h}$ and the terminal elimination half-life is of the order of 1 h [Fig. 3(b)].

Principal metabolites (see Fig. 2). The principal metabolite is the acid (RU 59416) resulting from oxidation of the primary alcohol function on the butyl side-chain. It is present from the first sampling time (5 min) at concentrations higher than those of RU 58841 (around 9 $\mu\text{g/ml}$). They continue to rise up to 2 h (mean value 32 $\mu\text{g/ml}$) then decrease. At 24 h it is no longer detectable.

The *N*-desalkyl metabolite (RU 56279) appears later. It is only detected after 4 h and shows a maximum at 6 h (mean value 0.125 $\mu\text{g/ml}$). At 24 h its

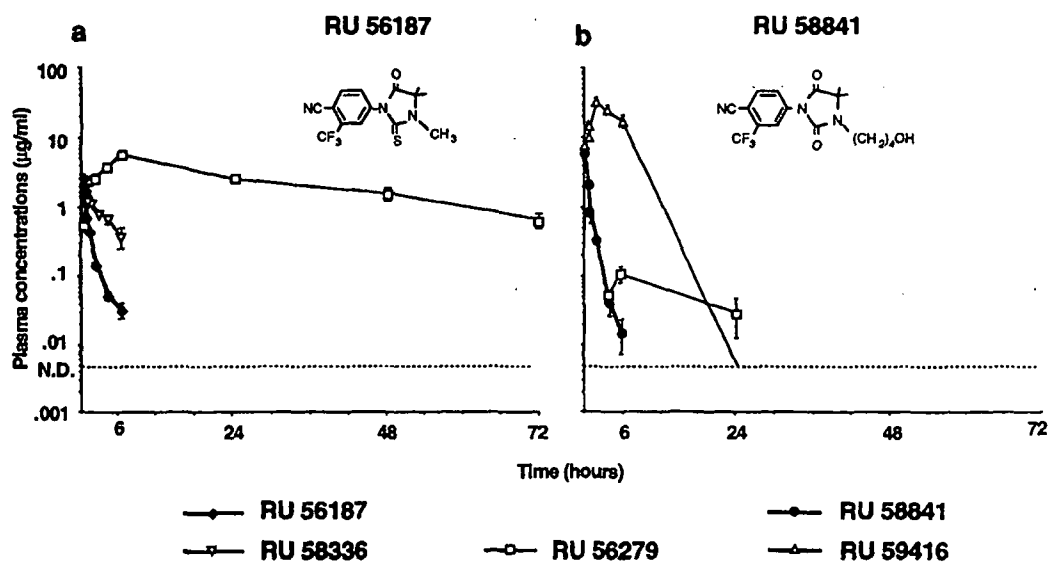


Fig. 3. Plasma concentration–time curves of parent compound and metabolites after i.v. administration of 10 mg/kg of RU 56187 (left) or RU 58841 (right). Each point represents the mean \pm SEM of 4 animals.

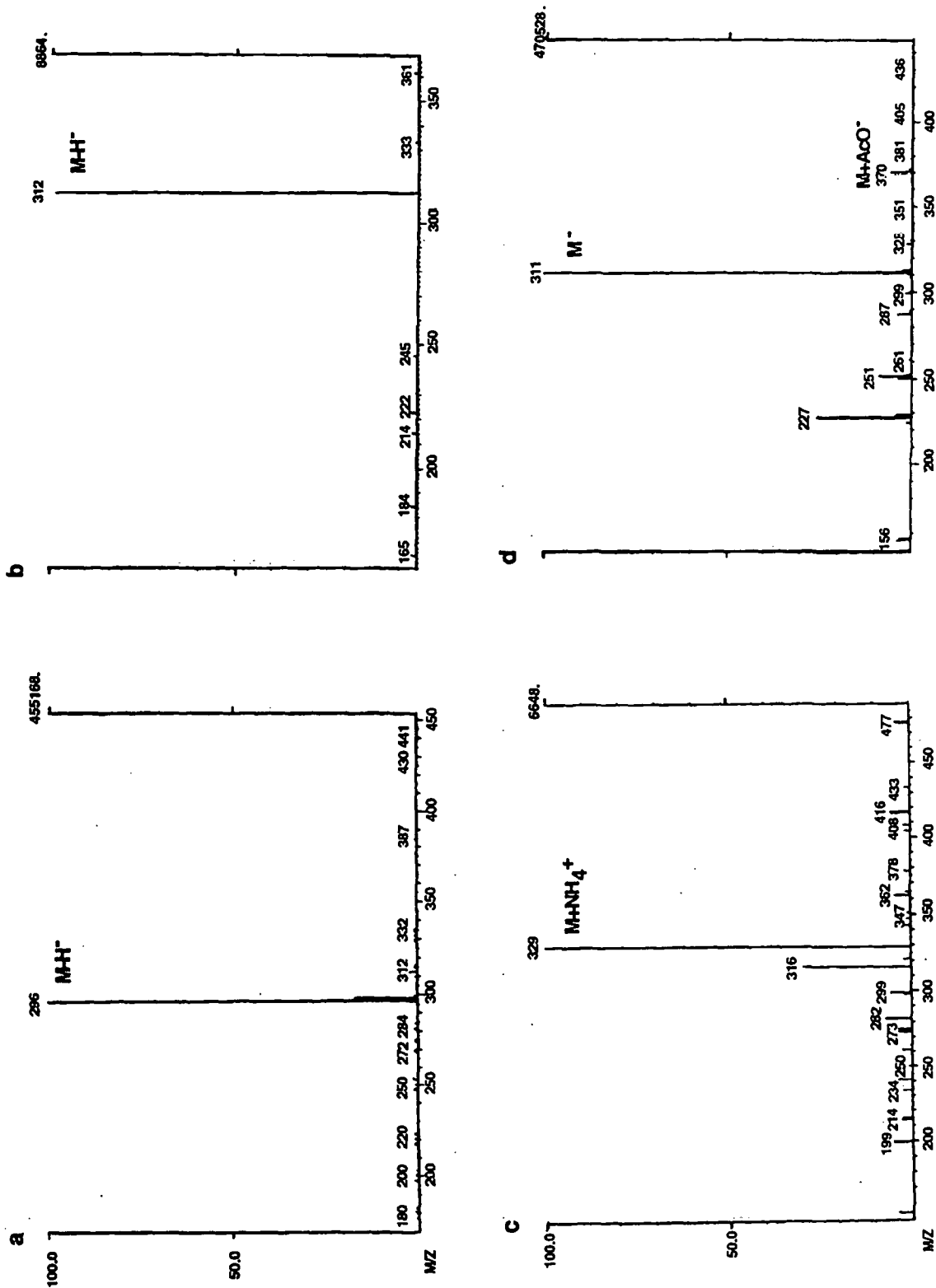


Fig. 4. Mass spectra of RU 56187 metabolites in a plasma extract: (a) RU 56279; (b) RU 56191 (both by thermospray ionisation, mode (-)); (c) RU 58836 (thermospray ionisation, mode (+)); (d) RU 58836 (thermospray discharge, mode (-)).

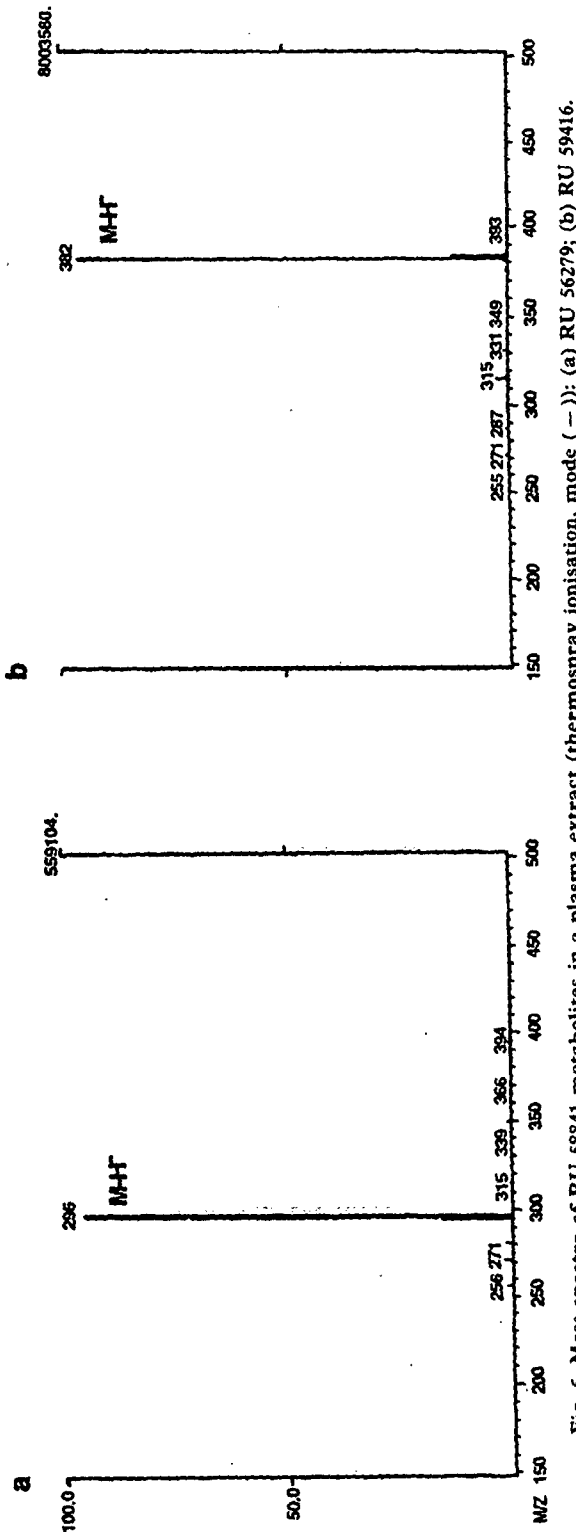


Fig. 5. Mass spectra of RU 58841 metabolites in a plasma extract (thermospray ionisation, mode (—)): (a) RU 56279; (b) RU 59416.

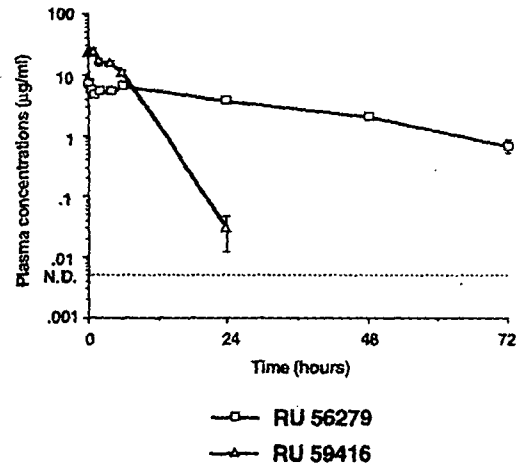


Fig. 6. Plasma concentration-time curves of RU 56279 or RU 59416 after i.v. administration of 10 mg/kg of RU 56279 or RU 59416. Each point represents the mean \pm SEM of 4 animals.

concentration is about 4 times lower ($0.03 \mu\text{g/ml}$) and it is no longer detected at 48 h.

Fig. 5 represents the mass spectra of the two metabolites from a plasma extract, which are in good agreement with those of the reference compounds (a, RU 56279; b, RU 59416).

Quantification of the fraction of the common *N*-desalkyl-hydantoin metabolite (RU 56279) formed from RU 58841 or RU 56187

The proportion of metabolite (RU 56279) formed from RU 58841 or RU 56187 was determined by comparison of the AUCs obtained after administration of the two parent compounds or of RU 56279 itself [4].

I.v. administration of RU 56279 (10 mg/kg). The curve corresponding to the mean concentrations is shown in Fig. 6. The experimental AUC, from 0 to 72 h, is equal to $230.0 \mu\text{g/ml} \times \text{h}$ and the terminal elimination half-life is of the order of 20 h.

After i.v. administration of RU 56187, the experimental AUC corresponding to RU 56279 is equal to $176.4 \mu\text{g/ml} \times \text{h}$; therefore, the proportion of metabolite formed, given by the ratio of the AUC of RU 56279 after administration of RU 56187 and the AUC of RU 56279 after administration of the metabolite itself, is equal to 77% (taking into account the difference in M.W.).

After i.v. administration of RU 58841 (10 mg/kg), the experimental AUC corresponding to RU 56279 is of the order of $2.26 \mu\text{g/ml} \times \text{h}$, thus, the proportion of metabolite formed, calculated in the same way, is about 1%.

Quantification of the fraction of acid metabolite (RU 59416) formed after administration of RU 58841

I.v. administration of RU 59416 (10 mg/kg). The curve corresponding to the mean concentrations is shown in Fig. 6. The experimental AUC from 0 to 24 h, is equal to $190 \mu\text{g/ml} \times \text{h}$ and the terminal elimination

half-life is of the order of 2 h (the latter was determined approximately using only the 3 points at 4, 6 and 24 h, since the concentrations decrease very rapidly between 6 and 24 h).

The proportion of metabolite RU 59416 formed from RU 58841, given by the AUC ratios as above, is 93%.

Pharmacokinetics and metabolism of RU 56187 and RU 58841, in the rat after cutaneous administration of 10 mg/kg

Limited curves of the parent compounds and of their principal metabolites, at the same dose of 10 mg/kg, are shown in Fig. 7. Following cutaneous application of RU 56187, plasma concentrations were determined at 4, 6 and 24 h. Unchanged compound was recovered at all 3 times at very low concentrations (maximum equal to 0.014 µg/ml after 6 h); RU 56279 reached concentrations equal to 1 µg/ml after 24 h; RU 58336 was not detected. For RU 58841 and its metabolites, assays were only performed 6 and 24 h after cutaneous application: the parent compound was quantified only at 6 h (mean concentrations equal to 0.012 µg/ml); RU 56279 concentrations at 6 h are about half those obtained after i.v. administration; RU 59416 was only detected at 6 h, at concentrations equal to 1 µg/ml.

Antiandrogenic activity of RU 56187 and its metabolites by oral route

The oral activity of RU 56187 was compared with that of its metabolites RU 56279 and 59416 characterized after i.v. administration. Castration leads to a strong decrease in sex organ weight which is prevented by TP. After a chronic treatment at a dose of 1 mg/kg, RU 56187 induced 61 and 78% decreases in prostate

Table 1. Oral antiandrogenic activity of RU 56187 and its metabolites administered at 1 mg/kg, with respect to prostate and seminal vesical weights

	Prostate weight (mg)	Seminal vesicle weight (mg)
Castrated animals	19.9 ± 1.6	13.8 ± 1.8
Castrated + TP	160.1 ± 7.3	178.7 ± 9.6
Castrated + TP + RU 56187	74.0 ± 9.0*	49.3 ± 9.5*
Castrated + TP + RU 56191	52.3 ± 3.4*	19.6 ± 3.1*
Castrated + TP + RU 56279	80.7 ± 6.1*	33.0 ± 2.2*
Castrated + TP + RU 58336	83.6 ± 6.9*	31.1 ± 4.2*

Each value represents the mean of 5 animals ± SEM. **P* < 0.01, using Dunnett's test, indicates a significant difference between the groups.

and seminal vesicle weights respectively compared with castrated animals supplemented with TP. Concerning RU 56191, 56279 and 58336, their administration reduces prostate weights by 67, 57 and 53% and seminal vesicle weights by 89, 88 and 86% respectively (Table 1).

Antiandrogenic activity of RU 58841 and its metabolites by subcutaneous route

The antiandrogenic activity of RU 58841 was compared with that of its metabolites RU 56279 and 59416 identified after i.v. administration. After chronic treatment at a dose of 1 mg/kg, RU 58841 and 59416 did not induce any significant decreases in prostate or seminal vesicle weights. On the contrary, RU 56279 caused a strong decrease in these parameters; with reductions in prostate and seminal vesicle weights of 76 and 94%, respectively (Table 2).

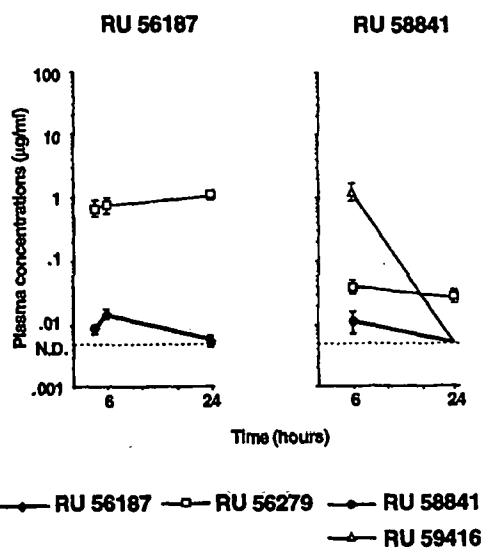


Fig. 7. Plasma concentration-time curves of parent compound and metabolites after cutaneous administration of 10 mg/kg of RU 56187 (left) or RU 58841 (right). Each point represents the mean ± SEM of 4 animals.

DISCUSSION

Antiandrogens are compounds of potential therapeutic interest for the treatment of either systemic androgen-dependent disorders such as prostate cancer or topical ones such as acne, alopecia or hirsutism [5]. Several non-steroid products (flutamide, Anandron) are available for the treatment of prostate cancer [6, 7]

Table 2. Subcutaneous antiandrogenic activity of RU 58841 and its metabolites administered at 1 mg/kg, with respect to prostate and seminal vesicle weights

	Prostate weight (mg)	Seminal vesicle weight (mg)
Castrated animals	17.7 ± 1.2	11.0 ± 0.6
Castrated + TP	169.5 ± 10.0	156.8 ± 7.1
Castrated + TP + RU 58841	146.0 ± 8.9	118.9 ± 8.4
Castrated + TP + RU 56279	47.8 ± 6.0*	19.6 ± 2.1*
Castrated + TP + RU 59416	143.3 ± 10.3	134.2 ± 11.4

Each value represents the mean of 5 animals ± SEM. **P* < 0.01, using Dunnett's test, indicates a significant difference between the groups.

or in development (Casodex) [8] but at the moment there is no topically acting compound which is devoid of systemic effects. As a result of structure-activity studies, new molecules with dissociated effects have been developed [2].

RU 56187 which shows strong systemic inhibitory activity on accessory sex organ weights (prostate and seminal vesicles) could be used for the treatment of prostate cancer [2]. However, despite an affinity for the androgen receptor *in vitro* about 100 times higher than that of the compounds cited above, it appears to be only 3–10 times more active *in vivo* [2]. RU 58841 on the other hand, which is a powerful topical antiandrogen with respect to the hamster flank organ but with very weak systemic effects, is of interest for the treatment of acne [1]. Its affinity is in good agreement with its pharmacological profile [1].

The metabolism of this series of compounds has not yet been described. That of Anandron, a hydantoin which differs from RU 56187 by the presence of a nitro group on the benzene ring, has been described in man and in various animal species [9, 10], but its metabolites contribute very little to its activity [11]. It therefore seemed of interest to make an initial study of the principal metabolites of RU 56187 and RU 58841, in an attempt to explain their dissociated effects and the difference between the *in vivo* and *in vitro* results observed for RU 56187. Since these compounds have a very short elimination half-life (about 1 h), the difference in their activity on accessory sex organs would not seem to be related to their rate of elimination and could not explain the systemic activity of RU 56187. It could, however, be explained by a metabolism product.

After *i.v.* administration of RU 56187, three metabolites were identified in the plasma: RU 58336 which is formed by oxidation of the thio function, RU 56191 arising from an *N*-demethylation and RU 56279 resulting from both these biotransformations. RU 56279 is the principal metabolite: about 80% of RU 56187 administered is transformed into RU 56279 which is found at relatively high concentrations. Its elimination is slow, the half-life being about 20 h. RU 58336 is present at concentrations of the order of those of RU 56187 but it is rapidly eliminated. RU 56191 was detected but not quantified. Preliminary results (not yet published) have shown that these three compounds display a low affinity for the androgen receptor. Thus, in order to investigate their activities, they were administered orally to castrated rats supplemented with testosterone propionate, an experimental model very sensitive to the systemic activities of antiandrogens [12]. After 2 weeks of treatment, they cause an important reduction in prostate and seminal vesicle weights, equivalent to that induced by RU 56187 itself. Thus the systemic activity of this compound which is not well correlated with its affinity, could be mediated by its metabolites and specially by the most important of them: the *N*-desmethyl hydantoin derivative (RU

56279). This metabolite is found at high concentration in the plasma and it remains there for a long time so, despite a low affinity, it displays a long acting antiandrogenic effect both on prostate and seminal vesicle weights.

As far as RU 58841 is concerned, two principal metabolites have been identified: RU 56279, resulting from *N*-dealkylation and RU 59416, the acid formed by oxidation of the butyl alcohol function. RU 56279 is formed in a very low proportion, representing about 1% of RU 58841 administered, compared with RU 59416, which represents 93%. RU 58841, a topically active compound has little systemic activity. Its metabolites were therefore administered *s.c.* so as to mimic the complete passage of the compounds through the skin and to measure their effects at the level of the prostate and seminal vesicles. In these conditions, RU 56279 causes an important reduction in prostate and seminal vesicle weights (as after oral administration). RU 59416 on the other hand does not significantly modify these parameters. Furthermore, it displays a very low affinity for the androgen receptor (preliminary results, not yet published). Thus the lack of systemic effects of RU 58841 could be explained by the fact that it is metabolized principally to a compound which is inactive on accessory sex organ weight in the rat.

Thus, the dissociation of the effects RU 56187 and 58841 would therefore seem to be related to the extent of their metabolism and to the proportion of the common *N*-desalkyl metabolite formed.

RU 58841 being at present in clinical development for the local treatment of acne, alopecia and hirsutism, its pharmacokinetics have also been studied after cutaneous administration. The same metabolites were found in the plasma but at concentrations much lower than after *i.v.* administration. These results showing that RU 58841 is transformed primarily into an inactive metabolite suggest that, provided that its metabolism in man is the same as in the rat, it could be used topically without the risk of affecting accessory sex organ weight.

Acknowledgement—The authors are extremely grateful to R. Legrand, from the Physics Department, Roussel Uclaf, for his help in analysing and determining the structures of the metabolites.

REFERENCES

1. Battmann T., Bonfils A., Branche C., Humbert J., Goubet F., Teutsch G. and Philibert D.: RU 58841, a new specific antiandrogen: a candidate of choice for the treatment of acne, androgenic alopecia and hirsutism. *J. Steroid Biochem. Molec. Biol.* 48 (1994) 55–60.
2. Teutsch G., Goubet F., Battmann T., Bonfils A., Bouchoux F., Cerede E., Gofflo D., Gaillard-Kelly M. and Philibert D.: New non-steroidal antiandrogens: synthesis and biological profile of high affinity ligands for the androgen receptor. *J. Steroid Biochem. Molec. Biol.* 48 (1994) 111–119.
3. Dunnett C. W.: A multiple comparison procedure for comparing several treatments with a control. *J. Am. Stat. Assoc.* 50 (1955) 1096–1121.

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<p>(21) International Application Number: PCT/US96/10286 (22) International Filing Date: 13 June 1996 (13.06.96) (30) Priority Data: 08/491,130 16 June 1995 (16.06.95) US (71) Applicant (for all designated States except US): BIOPHYSICA FOUNDATION [US/US]; 3333 North Torrey Pines Court #100, La Jolla, CA 92037 (US). (72) Inventors; and (75) Inventors/Applicants (for US only): SOVAK, Milos [DE/US]; 3333 North Torrey Pines Court #100, La Jolla, CA 92037 (US). BRESSI, Jerome, C. [US/US]; P.O. Box 126874, San Diego, CA 92112 (US). DOUGLASS, James, Gordon, III [US/US]; 4066 Moratalla Terrace, San Diego, CA 92130 (US). CAMPION, Brian [US/US]; 122 S. Rios Avenue #5, Solana Beach, CA 92075 (US). WRASIDLO, Wolfgang [US/US]; 307 Prospect Street, La Jolla, CA 92037 (US). (74) Agents: ROWLAND, Bertram, I. et al.; Flehr, Hohbach, Test, Albritton & Herbert, Suite 3400, 4 Embarcadero Center, San Francisco, CA 94111-4187 (US).</p>	<p>(81) Designated States: AL, AM, AT, AU, AZ, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TR, TT, UA, UG, US, UZ, VN, ARIPO patent (KE, LS, MW, SD, SZ, UG), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), ÖAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG).</p> <p>Published <i>With international search report.</i></p>	
<p>(54) Title: ANDROGENIC DIRECTED COMPOSITIONS</p>		
<p>(57) Abstract</p> <p>Substituted phenylthiohydantoin are provided for use in detecting the presence of tumor cells having androgenic receptors and providing for cytostatic and cytotoxic activity toward such cells. The subject compounds provide for vehicles for specific targeting to the endrogenic receptor containing cells of cytostatic and/or cytotoxic agents, heavy or light radioactive or radioopaque atoms, and the like for detection and treatment of cancer cells involving androgenic receptors or blocking androgenic receptors.</p>		

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ANDROGENIC DIRECTED COMPOSITIONS

INTRODUCTION

Technical Field

The field of this invention is diagnosis and treatment of androgenic related
5 neoplasia and blockage of androgenic receptors.

Background

The growth of prostate cancer (CaP) depends upon the presence of androgen
(male) hormones, acting via androgen receptors contained in the cell's nucleus. The
only effective, albeit temporary, therapy of prostate cancer is based upon
10 interference of male hormone production or activity, using estrogenic steroids or
non-steroidal substances to block the cancer cells' androgen receptors. There are a
number of problems with these therapies. Steroidal estrogens had to be abandoned
due to their high cardiovascular toxicity. The only steroidal compound clinically
used today is cyproterone acetate. However, it also binds to the glucocorticoid and
15 progesterin receptors. Current, clinically-used non-steroidal anti-androgens such as
Flutamide, Casodex or Anandron do not bind sufficiently to androgen receptors to
achieve their complete blockage. None of the current anti-androgens provide
permanent relief. It is suspected that the incomplete blockage of the receptors may
be the reason why, with time, the therapy invariably becomes ineffective as the CaP
20 cells mutate having proliferated metastatically. At that phase, the cells cannot be
substantially influenced by any known chemotherapy or radiation.

There is the further consideration that the current armamentarium for the
diagnostic staging of prostate cancer is extremely poor and yet essential in choosing
the therapeutic mode. Proof of metastatic dissemination beyond the prostate
25 excludes surgery and relegates these patients to systemic therapy. With improved
diagnostic staging, unnecessary prostatectomies, a major and potentially mutilating

surgery, could be avoided.

Only recently, an assay has become available for the detection of CaP cells circulating in the blood. However, that finding alone does not imply the existence of metastases. Typically, early metastases occur in the lymph nodes and the later
5 ones develop in the bones. While ^{99}Tc scans can visualize bone defects, the lymph node metastases are extremely difficult to locate since typically, the infiltrated nodes are neither enlarged nor show changes on either magnetic resonance or x-ray computed tomography. Further, because of their low metabolic rate, the pathological nodes cannot be identified by positron emission spectrography using
10 ^{18}F -deoxyglucose. Lymph node biopsy is possible only in the pelvic area. Early metastases in inaccessible paraaortic lymph nodes cannot be detected and consequently these patients are operated upon needlessly. Recently developed radiolabeled monoclonal antibodies against prostate cancer have only a limited use due to their low target specificity and long persistence in the blood pool, liver and
15 spleen, which interferes with the imaging.

There have been a number of attempts to develop a CaP radionuclide scanning agent. Several radioiodinated androgen steroids were made, but they suffer from synthetic complexity. Steroidal androgens labeled with ^{18}F were synthesized as a potential PET imaging agent for prostate cancer, but their practicability is limited
20 due to the complicated synthesis and need for specialized rare equipment (PET scanners) to detect positron emitting radionuclides. There is a further consideration that androgens promote CaP growth.

There is, therefore, substantial interest in developing novel compounds which can provide for the diagnosis and therapy of prostate cancer.

25

Relevant Literature

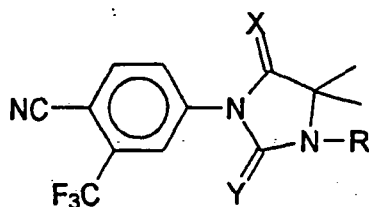
N-aryl substituted imidazolidiones have been reported in DE32 22 523; Offenlegungsschrift 26 49 925; WO88/03404; EP0 436426; EP0 494819; EP0 580459, and Teutsch, J. Steroid Biochem. Molec. Biol. (1994) 48:111-119. The
30 activity of the trifluoromethyl, nitro- and trifluoromethyl, cyanophenyl derivatives as high-affinity ligands for the androgen receptor are reported in Teutsch, *supra.*, as well as in many of the foregoing patents.

where the linking group may include one or more functionalities, such as amino, oxy, and non-oxo-carbonyl, where amides and esters may be involved, e.g. urethanes; and the third group will involve carbocyclic aryl groups, particularly iodoaryl, which may be bonded to the nitrogen of the hydantoin through a linking
 5 group of from one to eight, usually two to six carbon atoms, preferably two to three carbon atoms, where the linking group may include an amino, oxy or non-oxo-carbonyl functionality, particularly ester or amide, and the aryl group may be substituted with oxy, amino, non-oxo-carbonyl, and derivatives thereof. As the aryl group, phenyl is of particular interest.

10 Tissue comprising cells with androgen receptors include prostate tissue, ovary tissue, testes, etc. Hosts of interest include primates, e.g. humans, domestic animals and pets.

The first group of the compounds of the subject invention will have the following formula:

15



20

wherein:

25 X is oxygen or nitrogen, with the proviso that when R is iodoaryl, X may be sulfur;

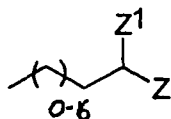
Y is sulphur, with the proviso that when R is iodoaryl group, Y may be sulphur, oxygen or nitrogen, preferably X and Y are different;

R is an organic group, which may be aliphatic and may comprise one or
 30 more heteroatoms, alicyclic, aromatic, heterocyclic, or combinations thereof, where heteroatoms include oxygen, nitrogen, sulphur etc., to be further defined below.

The first group of compounds will comprise monothiohydantoin, where the other oxo group of the hydantoin will be oxygen or nitrogen. These groups will, for

the most part, have R having the following formula:

5



wherein:

Z is hydroxyl, amino, a substituted amino or a 4-diazolyl, particularly a 4-(1',3'-imidazolyl);

Z' is hydrogen, hydroxyl, or may be taken together with Z to provide for olefinic or acetylenic unsaturation, or a 2,2-dimethyldioxolane.

The substituents on amino nitrogen may be varied widely, depending upon the use of the compound. For cytotoxicity or antiproliferative activity, the amino group may be unsubstituted or substituted, particularly with the single acyl group, where the acyl group may serve to enhance the activity of the compound by changing its pharmacokinetic activities, by providing for a second cytotoxic or antiproliferative compound, by providing for a chelating agent for chelating a metal ion, particularly a radioactive metal or non-metallic ion, for carrying a radioopaque atom, or the like. Radioactive elements include fluorine, iodine, technetium, etc. Other metals of interest include gadolinium and the like.

Similarly, the hydroxyl, particularly the terminal hydroxyl, may be employed as a site for linking, forming ethers or esters, where the groups bound to oxygen will come within the above description; activations and displacement with other groups of interest, e.g. fluorine, sulphur alkyls and the like.

In addition, iodoaryl groups may be employed which are linked to the nitrogen through an alkyl chain, where the alkyl chain may be of from 1 to 6, usually from 1 to 4, preferably from 2 to 4 carbon atoms. The iodoaryl group may be linked directly to the carbon of the alkyl group or linked through a heteroatom, particularly nitrogen or oxygen, e.g. amide, secondary amine, ether, ester, etc. where the iodoaryl group may have a non-oxo-carbonyl or amino group linked to an annular carbon atom as part of the linking chain. The iodoaryl will generally have from 1 to 4, usually 2 to 4, more usually 2 to 3 iodine atoms, and may be further

substituted with oxy, particularly hydroxy or alkoxy of from 1 to 3 carbon atoms, or amino, or a substituted amino (mono- or disubstituted), having alkyl substituents having a total of 1 to 6 carbon atoms, more usually 1 to 4 carbon atoms, and 0 to n-1 oxy groups, where n is the number of carbon atoms in the substituent. A variety of
5 aminosubstituted symmetrically substituted triiodoisophthaldiamides and diaminosubstituted symmetrically substituted triiodobenzamides have been reported in the literature, where the nitrogen atoms are substituted with acyl groups, alkyl groups or oxyalkyl groups of 1 to 6, usually 1 to 4 carbon atoms and 0 to n-1 oxy groups. See, for example, U.S. Patent Nos. 4,547,357; 4,021,481; 4,364,921 and
10 4,341,756 and references cited therein. The carboxyl group may be used to link the iodoaryl group to the thiohydantoin through the alkyl chain. Alternatively, iodine may be bonded to an sp_2 carbon atom of an alkenyl group.

Illustrative R groups include: allyl, propynyl, aminoethyl, aminopropyl, 2-hydroxypropyl, 3-hydroxypropyl, 2-hydroxyethyl, 2,3-dihydroxypropyl,
15 2-hydroxy-3-acetoxypropyl, 4-benzamidobutyl, 4-fluorobutyl, 4-iodobut-3-enyl, 2-iodoprop-2-enyl, cis & trans-3-iodo-prop-2-enyl, 3-(4'-oxazolyl-1,3)propyl, 2-(4'-diazolyl)ethyl, 3-(propionamido)propyl, N-phenoxy carbonyl 2-aminoethyl, N-methoxycarbonyl 2-aminoethyl, 3-(3',5'-diiodo-4'-dimethylaminophenyl)propyl, 2-(3',4',5'-triiodophenyl)propyl, N-(cysteinyl, glycyl, glycyl) 2-aminoethyl,
20 (3',6',9'-triazanonoxy)ethyl, p-hydroxyphenylpropyl, and the carboxamide of N-nitritotriacetic acid and 2-aminoethyl.

Alternatively, various cytotoxic agents may be employed, which are joined to the subject hydantoins by any convenient linking group, which does not significantly diminish the cytotoxic or antiproliferative activity of the compound. Compounds of
25 interest include methotrexate, taxol, 5-fluorouracil, adriamycin, bleomycin, and the like.

The subject compounds can be prepared in accordance with conventional ways, varying the particular procedure based on the particular side groups. The preparation of hydantoins conveniently involves the use of an isocyanate and a
30 substituted α -aminoacetonitrile. By appropriate choice of the isocyanate and the α -aminoacetonitrile, one may arrive at the final product in a single step.

Alternatively, by employing various protective groups, which may be subsequently removed, or providing for substituents which become involved in the formation of

the hydantoin or may provide for sites for further derivatization. Various procedures are described in EPO Publication Numbers 0 494 819 and 0 580 459. Also, a significant number of examples may be found in the subject experimental section.

5 The subject compositions find a variety of uses associated with prophylactic and therapeutic opportunities. By providing for substituents which allow for detection by x-rays, molecular resonance imaging, radioactivity, or the like, regions of a mammalian host, particularly humans, can be investigated, where the regions are associated with an androgenic receptor. Thus, cells or tissues associated with the
10 androgenic receptors may be visualized, so as to identify neoplasms, benign tumors, mobile cells, etc. Thus, by having substituents which have radioactive atoms, heavy metals, heavy atoms such as iodine, or the like, one can visualize physiological structures associated with androgenic receptors.

 In addition, the subject compounds have proliferative inhibitory capability in
15 inhibiting the proliferation of cells having androgenic receptors and dependent upon signal transduction associated with the androgenic receptors. The subject compounds are found to have a high affinity for the androgenic receptors, demonstrating enhanced activity as compared to prior substituted hydantoins.

 In addition, the subject hydantoins can be used as vehicles for transporting
20 other cytotoxic agents to the androgen receptor comprising cells. Thus, while at the same time inhibiting androgenic activation, other pathways which inhibit proliferation may also be addressed. Thus, one can greatly enhance the therapeutic index of a known chemotherapeutic agent by directing the chemotherapeutic agent to specific sites in the host.

25 The subject compositions may be formulated in accordance with conventional ways for use *in vivo*. The subject compounds are found to be stable in human plasma at physiological temperatures. The subject compounds are found to have substantially greater cytostatic and cytotoxic effects in inhibiting cell growth for neoplastic cells, as compared to normal cells, i.e. having a high therapeutic index.
30 The subject compositions are readily formulated in conventional carriers, such as saline, phosphate buffered saline, vegetable oils, ethanol, or other physiologically acceptable carrier.

 The concentrations used for the subject compounds in diagnosis and therapy

will be varied widely, depending upon the purpose of the compound, the patient being treated, the stage of the disease, whether the subject compounds are being used by themselves or in a combination therapy, the manner of administration, the responsiveness of the cancer cells to the drug, and the like. The particular dosage
5 can be determined empirically. Other components of the formulation may include buffers, stabilizers, excipients, or the like. Depending upon the particular compound and its formulation, administration may be oral or parenteral, including intravascular, subcutaneous, intratumoral, intraperitoneally, etc.

The subject compounds may also be used in competitive assays for evaluating
10 other compounds as to their cytotoxic or cytostatic effect. Thus, specific cell lines may be employed where the effect of an agent on the cytotoxic level of a subject compound may be determined in relation to the survival rate of the target cells. Also, in mixtures of cells containing neoplastic androgenic receptor containing cells the subject compounds can be used to eliminate the neoplastic cells in the presence
15 of normal cells. Thus, in a variety of cultures, where androgenic receptor containing cells may be susceptible to becoming or are tumorous, by maintaining a cytotoxic level of the subject compounds in the medium, the cells may be selectively killed.

The following examples are offered by way of illustration and not by way of
20 limitation.

EXPERIMENTAL

The following compounds were prepared according to the general method described by Teutsch et. al., J. Steroid Biochem. Molec. Biol. 1994; 1:11-119.

25

Example 1

4-[3'-(2''-(N-t-butoxycarbonyl)-aminoethyl)-4',4'-dimethyl-5'-imino-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-136)

Crude 2-trifluoromethyl-4-isothiocyanoato-benzonitrile (700 mg, 3.07 mmol)
30 was dissolved in THF (6.0 mL). At room temperature, triethylamine (59 μ L, 0.42 mmol) was added to the stirring solution followed by 2-(1',2'-ethyldiamino-N-t-butoxycarbonyl)-2-cyanopropane (682 mg, 3.00 mmol). The reaction was refluxed for 40 minutes under a N₂ atmosphere and then the solvent was removed under

reduced pressure. The resulting brown residue was purified by silica gel chromatography (CH₂Cl₂/acetone, gradient) and treated with activated carbon to yield 951 mg (68.1%) of light yellow powder.

mp: 81 °C (dec); UV (MeOH): λ_{\max} = 234 nm (ϵ = 18841) and
5 260 nm (ϵ = 21454)

Example 2

4-[3'-(2'',2''-dimethyl-1'',3''-dioxolane-4''-methyl)-4',4'-dimethyl-5'-imino-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-163)

10 BP-163 was prepared and purified as described in Example 1 using the amino cyanopropane prepared from 2,2-dimethyl-1,3-dioxolane-4-methanamine and acetone cyanohydrin. Yield = 63.3%.

UV (MeOH): λ_{\max} = 230 nm (ϵ = 23528), 244 nm (ϵ = 22733), and
15 258 nm (ϵ = 24590);

Example 3

4-[3'-(2''-propenyl)-4',4'-dimethyl-5''-imino-2'-thioxo-1''-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-208)

20 BP-208 was prepared and purified by the same method described in Example 1 using the amino cyanopropane prepared from allylamine and acetone cyanohydrin. Yield = 67.3%.

Example 4

25 4-[3'-(2''-propynyl)-4',4'-dimethyl-5'-imino-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-211)

BP-211 was prepared as described in Example 1 using the amino cyanopropane prepared from propargyl amine and acetone cyanohydrin. The compound was purified by chromatography (CH₂Cl₂/acetone, 100% (50:50 gradient by 10% segments) and isolated as an orange oil. The product was not further
30 characterized and was carried as is into the hydrolysis step. (Example 12)

Example 5

4-[3'-(2''-{4''-imidazolyl}ethyl)-4',4'-dimethyl-5'-imino-2'-thioxo-1'-

imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-210)

BP-210 is prepared as described in Example 1 using the amino cyanopropane prepared from 4-(aminoethyl)imidazole and acetone cyanohydrin. The compound is purified by column chromatography and isolated as a pale yellow oil. It is used in the subsequent hydrolysis without further purification. (Example 13)

Example 6

4-[3'-(2"-p-hydroxyphenylethyl)-4',4'-dimethyl-5'-imino-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-212)

BP-212 was prepared as described in Example 1 using the amino cyanopropane prepared from p-hydroxyphenethylamine and acetone cyanohydrin. Following silica gel chromatography (CH₂Cl₂/acetone; gradient), a pale yellow solid was obtained, which was taken directly into the hydrolysis step without further characterization. (Example 14)

15

Example 7

4-[3'-(2"-aminoethyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-138)

BP-136 (300 mg, 0.66 mmol) was dissolved in MeOH (3.5 mL) and 2N HCl (.065 mL) with stirring at room temperature. The reaction mixture was refluxed for two hours, then the solvent was removed under reduced pressure, and the resulting solid was crystallized as the hydrochloride from isopropanol. Yield 204 mg (79.0%).

mp: >200(C; UV (MeOH): δ_{\max} = 234 nm (18441) and 252 nm (ϵ = 20891)

25

Example 8

4-[3'-(2",3"-dihydroxypropyl)-4',4'-dimethyl-5'-oxo-2'-thio xo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-135)

BP-135 was prepared in the manner described in Example 7 using the appropriate imine (BP-163, example 2). The product was isolated by pouring the reaction mixture over a mixture of ice and water. The product was extracted with EtOAc, dried over MgSO₄ and the solvent removed under reduced pressure. BP-

30

254 nm ($\epsilon=21499$)

Example 12

4-[3'-(2''-propynyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-199)

BP-199 was prepared from the appropriate imine (BP-211, example 4) in the same manner as described in Example 7. The product was isolated as colorless crystals from CH_2Cl_2 /hexane.

mp: 120-121°C (dec); UV: $\lambda_{\text{max}}=206$ nm ($\epsilon=17328$), 232 nm ($\epsilon=18068$), and 252 nm ($\epsilon=22003$).

Example 13

4-[3'-(2''-{4''-imidazolyl}ethyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-213)

BP-213 was prepared from the appropriate imine (BP-210, example 5) in the same manner as described in example 7. The crude product was purified by column chromatography and isolated as a colorless solid in high purity ((96%, HPLC).

UV: $\lambda_{\text{max}}=234$ nm ($\epsilon=14113$) and 254 nm ($\epsilon=1604$).

Example 14

4-[3'-(2''-p-hydroxyphenylethyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-214)

BP-214 is prepared from the corresponding imine (BP-212, example 6) in the same manner as described in Example 7. The crude product is crystallized from CH_2Cl_2 /hexane as colorless crystals.

Example 15

4-[3'-(2''-N-acetylaminoethyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-139)

The free amine of BP-138 (100 mg, 0.28 mmol) was dissolved in $(\text{Ac})_2\text{O}$ (5.0 mL) and allowed to stir at room temperature for 30 minutes. The solvent was then removed under reduced pressure and the resulting off-white solid was purified by silica gel chromatography (CH_2Cl_2 /acetone; 95:5) to yield 102 mg (91.6%) of

pure compound.

mp: 77-79°C (dec); UV (MeOH): λ_{\max} = 234 nm (ϵ = 18694) and
254 nm (ϵ = 21499)

5

Example 16

4-[3'-(2"-aminoethyl-N-(glycyl-N'"-(2'"-(triphenylmethylthioacetyl)-glycine)))-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-197)

- 10 Dicyclohexylcarbodiimide (DCC, 1.1 mg, 5.35×10^{-3} mmol) and the free base of BP-138 (1.9 mg; 5.35×10^{-3} mmol) were added to a stirring solution of N-[2-triphenylmethylthioacetyl]-glycyl-glycine (2.0 mg, 4.46×10^{-3} mmol) in THF (0.200 mL) at room temperature. The reaction was heated at 35°C for two hours and then purified by preparative HPLC without further work-up. Yield = 50.2%.

15

Example 17

4-[3'-(4"-oxybutyl-O-glycyl-N'"-(2-(triphenylmethylthioacetyl)-glycine))-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-198)

- 20 To a stirred solution of N-[2-(triphenylmethylthioacetyl)-glycyl-glycine (2.0 mg, 4.46×10^{-3} mmol) in THF (2.00 mL) was added DCC (1.1 mg, 5.35×10^{-3} mmol), 4-[3'-(4"-hydroxybutyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile (RU 59063, 2.1 mg, 5.35×10^{-3} mmol) [Synthesized as described by Teutsch et. al., supra] and a crystal of DMAP. After stirring at
25 room temperature for 45 minutes, the product was isolated by preparative HPLC. Yield = 56.8%.

Example 18

- 30 4-[3'-(2"-aminoethyl-N-(glycyl-N'"-(2-thioacetyl)-glycine)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-207)

Bu₃SiH is added to a stirring solution of BP-197 in 10% TFA/CH₂Cl₂ and is purified by preparative HPLC without further work-up. This product can now be used as a substrate for complexing with ⁹⁹Tc by standard methods.

Example 19

4-[3'-(4"-oxybutyl-O-glycyl-N'"-(2-(thioacetyl)-glycine))-4',4'-dimethyl-5'-oxo-2''-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-209)

Bu₃SiH is added to a stirring solution of BP-198 in 10% TFA/CH₂Cl₂ and is
5 purified by preparative HPLC without further work-up. This product can now be used as a substrate for complexing with ⁹⁹Tc by standard methods.

Example 20

4-[3'-trans-(2"-propenyl-3"-tributylstannyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-
10 1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-237)

BP-199 (1.05 g) was dissolved in dry toluene (100 mL) under N₂. Bu₃SnH (1.12 mL) and AIBN (68.5 mg) were added and the reaction mixture heated to reflux. After stirring for 24 hours, additional aliquots of Bu₃SnH (0.40 mL) and AIBN (10 mg) were added. After further stirring for 3 hours at reflux, the reaction
15 was allowed to cool to room temperature and the volatiles removed under vacuum. The crude product was purified by column chromatography and isolated as a pale oil (1.67 g).

HPLC analysis indicated the presence of two isomers.

20 Example 21

4-[3'-trans-(2"-propenyl-3"-*iodo)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-305)

BP-237 is dissolved in a small amount of methanol. Radioiodination is accomplished using Na[¹²⁵I]I or Na[¹³¹I]I or Na[¹²³I]I by known methods (see Hunter
25 & Greenwood, Nature, 1962; 194:495-496). TLC with autoradiography indicates 50-75% radiochemical yield.

Example 22

4-[3'-(4"-methanesulfonyloxybutyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-
30 imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-232)

RU-59063 (described by Teutsch *et al.*, supra and Example 17), was dissolved in methylene chloride, pyridine was added and the solution cooled to 0°C. Under N₂, methanesulfonic anhydride was added slowly and the reaction allowed to

warm to room temperature. The solution was cooled and pyridinium hydrochloride is filtered. The product was purified column chromatography (silica gel, CHCl₃/acetone; gradient 100% (85:15) and isolated as a colorless solid. m.p. 114-115°C.

5 Example 23

4-[3'-(4''-fluorobutyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-218)

a.) ¹⁹F BP-218

RU 59063 (2.2 g) was placed in a 100 mL Schlenk flask with a stir bar and
10 placed under N₂. Dry methylene chloride was added (15 mL) and the solution stirred under N₂ for 10 minutes. Pyridine (1.66 mL) was added, the solution cooled to -78°C with dry ice/acetone bath. Dimethyl aminosulphur trifluoride (DAST, 0.905 mL) was added dropwise and the reaction stirred at -78°C for 4 hours. The solution was then allowed to warm to room temperature and then taken to dryness.
15 The product was isolated as a colorless oil by column chromatography (260 mg).

b.) ¹⁸F BP-218

[¹⁸F] Fluoride ion was produced by proton irradiation of oxygen-18 enriched (96% isotopic enrichment) held in an all-silver cyclotron target (330 μL target
20 volume). The aqueous [¹⁸F] fluoride was converted to a no-carrier added Kryptofix 2.2.2/K₂CO₃/¹⁸F; prepared by addition of the [¹⁸O] water/[¹⁸F] solution to a mixture of the aminopolyether 4,7,13,16,21,24-hexaoxa-1,10-diazabicyclo [8.8.8]hexacosane (Kryptofix 2.2.2, 26.0 mg, 0.069 mmole) and potassium carbonate (2.3 mg, 0.0166 mmole) in a Vacutainer®. The vessel was placed in an oil bath at 110°C,
25 and water was removed under a gentle stream of N₂, assisted by azeotropic distillation, each employing 0.5-0.8 mL CH₃CN.

The Kryptofix/K₂CO₃/¹⁸F solution (1-50 mCi) in anhydrous acetonitrile (500 (L) was added to 2.0 mg of (4-[3'-(4''-methanesulfonyloxybutyl)-4'-4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile) (BP-232). The
30 reaction mixture was heated for one hour at 110°C and then cooled before being injected onto a preparative HPLC system. The HPLC purification was performed on a C-18 reverse-phase preparative column and eluted with a 65:35 CH₃CN/H₂O solvent mixture (2 mL/min). Column effluent was monitored by a flow-through

radiation detector at 254 nm. The desired F-18 compound eluted at ~ 19 minutes. The solvents were evaporated in vacuo and the ¹⁸F BP-218 was reformulated in saline.

Both radio-HPLC and radio-TLC were used to determine radiochemical
5 purity. Purity by HPLC was determined using an ODS reverse-phase column, eluting with acetonitrile/water (80/20) with UV detection at 254 nm and a flow-through radiation detector. The retention time for F-18 BP-218 was 6.2 min.

Radio-TLC were performed as follows: silica gel plates; CHCl₃/acetone (95:5); F-18 BP-218 (R_f=0.5).

10

Example 24

7{5'',5''-dimethyl-4''-oxo-3''-[4'''-cyano-3'''-trifluoromethylphenyl-1'-imidazolidinyl]-2''-thioxo-1''-ethylcarbamoxy}paclitaxel. (BP-196)

A round bottom flask charged with paclitaxel (60 mg, 0.07 mmol), imidazole
15 (90 mg, 1.32 mmol) and a magnetic stir bar was placed under a N₂ atmosphere. CH₂Cl₂ (1.5 mL) was added and the solution was stirred at room temperature. To the solution was added portionwise a solution of 1.0 M ClSiEt₃ in THF (5 x 100 μL, 0.5 mmol). The progress of the reaction was monitored by HPLC. Upon completion, the 2'-(triethylsiloxy)paclitaxel was purified by preparative HPLC
20 yielding 51.3 mg (75%). Purity by HPLC 97%. Proton NMR of the product matched values given in the literature [Chandhary et. al., J. Org. Chem. 1993; 58(15):3798-3799]

A round bottom flask charged with 2'-(triethylsiloxy)paclitaxel (30 mg, 0.03 mmol) and p-nitrophenylchloroformate (310 mg, 1.50 mmol) and a magnetic stir bar
25 was placed under a N₂ atmosphere. A solution of pyridine (200 μL, 0.247 mmol) in CH₃CN (1.0 mL) was added and the mixture stirred at room temperature for 30 minutes. The product 2'-(triethylsiloxy), 7-(p-nitrophenylcarboxy)paclitaxel was purified by preparative HPLC yielding 24.2 mg (69%). Purity by HPLC was 96%.

To a round bottom flask charged with 2'-(triethylsiloxy), 7-(p-nitrophenyl-
30 carbonoxy)paclitaxel (28.0 mg, 0.014 mmol), 4-[3'-(2''-aminoethyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile (2 X 8.0 mg, 0.44 mmol) and a magnetic stir bar was added CH₂Cl₂ (300 μL). The solution was stirred at room temperature for 4 hours and the product, 2'-(triethylsiloxy)-7{5'',5''-

dimethyl-4"-oxo-3"-[4"-cyano-3"-trifluoromethylphenyl-1'-imidazolidinyl]-2"-thioxo-1"-ethylcarbamoxy}paclitaxel, was purified by preparative HPLC yielding 8.2 mg (85%). Purity by HPLC 97%.

To a round bottom flask charged with 2'-(triethylsiloxy)-7{5",5"-dimethyl-
5 4"-oxo-3"-[4"-cyano-3"- trifluoromethylphenyl-1'-imidazolidinyl]-2"-thioxo-1"-ethylcarb amoxy}paclitaxel (5.0 mg, 0.004 mmol) and a stir bar was added formic acid (250 μ L). The solution was stirred at room temperature for 15 minutes and the volatiles removed under vacuum. BP-196 was purified by preparative HPLC yielding 4.6 mg (>99%). The purity by HPLC was 99%.

10

Example 25

4-[3'-(2"-{4"--(2"'*iodo)imidazolyl}ethyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidnyl]-2-trifluoromethyl-benzonitrile . (BP-216)

BP-213 is dissolved in methanol. Radioiodination is accomplished with
15 chloramine-T and Na[¹²⁵I]I or Na [¹³¹I]I or Na [¹²³I]I by standard methods [Hunter and Greenwood, Nature, 1962; 194: 495-496] The product is purified by HPLC.

Example 26

20 4-[3'-gem-(2"-propenyl-2"-tributylstannyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-300)

BP-199 (2.30 g) was placed in a three-neck 500 mL round bottom flask fitted with two rubber septa, an N₂ adapter and stir bar. Dry toluene was added (30 mL) followed by HSnBu₃ (2.48 g). Pd(PPh₃)₄ (151 mg) was dissolved in toluene (30
25 mL) and added quickly to the previously prepared solution. After 24 hours of stirring at room temperature, an additional aliquot of Pd(PPh₃)₄ (50 mg) was added and the reaction heated at 65°C for 3 hours followed by stirring at room temperature for 48 hours. The reaction mixture was taken to dryness and the product(s) purified by column chromatography. HPLC analysis (C18 reverse phase, 75:25 ACN/H₂O)
30 suggested that the major product was BP-300 and the minor product was BP-237 (79:21), based on NMR comparison of the corresponding iodo compounds.

(Example 29)

Example 27

4-[3'-trans--(2"-propenyl-3"-iodo)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-305)

and

5 4-[3'-cis--(2"-propenyl-3"-iodo)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-307)

BP-237 (82% pure, with the remainder the corresponding cis isomer BP-354, 370 mg) was dissolved in CHCl₃ (5 mL) and cooled to 0°C. In a separate flask I₂ was dissolved (146 mg) in CHCl₃ (15 mL) and added to the solution of BP-237.

10 After 2 hours at room temperature, the volatiles were removed and the crude products separated and purified using column chromatography.

Example 28

15 4-[3'-(6"-methanesulfonyloxyhexyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-328)

BP-327 (10.4g) was dissolved in methylene chloride (130 mL), pyridine (2.5 mL) was added and the solution was cooled to 0°C under N₂. Methanesulfonic anhydride (5.5 g) was dissolved in methylene chloride (100 mL) and the resulting clear solution added slowly to the former solution. After 30 minutes at 0°C, the
20 solution was allowed to warm to room temperature at which time the volatiles were removed under vacuum. The crude product was dissolved in a minimum of chloroform, filtered, and purified using silica gel column chromatography. Combining the appropriate fractions followed by removal of volatiles gave the product as a light brown oil (8.8 g, 98% pure by HPLC).

25

Example 29

4-[3'-(6"-thiohexyl)hexyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-332)

BP-328 (1.10 g) was dissolved in methylene chloride (35 mL). In a separate
30 flask were placed hexanethiol (315 μL) and toluene (10 mL). Sodium methoxide (403 μL, 5.5 M) was added and the solution stirred for ten minutes. The resulting emulsion was added dropwise to the BP-328 solution with rapid stirring. After stirring for 12 h, the solution was stripped down and the crude product was purified

by column chromatography and isolated as a clear oil (160 mg) in 25% recovered yield. Additionally, unreacted BP-328 was also recovered (50%).

5 Example 30

4-[3'-{2''-N-(p-hydroxy phenethyl) amidoethyl}-4',4'-dimethyl-5'-oxo-2'-thio-1'-imidazolidinyl]-2-trifluoromethyl benzonitrile. (BP-231)

A 100 mL Schlenk flask was charged with BP-138 (430 mg, 1.20 mmol), Bolton-Hunter reagent (318 mg, 1.20 mmol) and a stir bar. Anhydrous THF (5 mL)
10 was added via a gas tight syringe and the reaction mixture stirred under N₂(g) at room temperature. After one hour, the volatiles were removed under vacuum and the crude product purified using column chromatography (230-400 mesh SiO₂, 20 g, packed with CHCl₃) using gradient elution (100% CHCl₃ (80:20 CHCl₃/Acetone). The appropriate fractions (as determined by TLC) were combined and the volatiles
15 removed to give the product as a white solid (385 mg) in 64% yield. The purity by HPLC was 99.0%. UV (MeOH): λ_{max}=206 nm (ε=9553), 228 nm (ε=9872), 254 nm (ε=8339).

Example 31

20 4-[3'-{2''-(N-3'''',5'''-diiodo-4'''-hydroxy phenethyl) amidoethyl}-4',4'-dimethyl-5'-oxo-2'-thio-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-248)

BP-231 (54.2 mg, 0.107 mmol) and chloramine-T (60 mg) was placed in a round bottom flask and CHCl₃ (6 mL) added. Iodine was added (6.05 mg).
25 Methanol (3 mL) was added dropwise at room temperature with stirring. The solution turned orange. After one hour, the reaction was quenched (Na₂S₂O₅ 50 mg in 5 mL H₂O) and the products extracted into CHCl₃ (2 x 10 mL). The combined organics were dried and the volatiles removed. The crude product was purified using column chromatography (SiO₂, 5 g, CHCl₃) with a gradient elution (100
30 CHCl₃ (95:5 CHCl₃/Acetone). Purity was 97% based on HPLC. Mass Spec: MH+ (757).

Example 32

4-[3'-(6''-hydroxyhexyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethyl-benzonitrile. (BP-327)

The amino cyanopropane derived from 6-hydroxyhexyl amine and acetone cyanohydrin (13.9 g, 75.7 mmol) was dissolved in THF (100 mL). In a separate
5 flask was placed the substituted aryl isothiocyanate (17.2 g, 75.7 mmol) to which was added THF (50 mL) and NEt₃ (2.0 mL). The latter orange solution was added to the former with stirring. After 12 hours, the volatiles were removed under vacuum to give the crude imine cyclization product as an viscous orange oil. This product was dissolved in methanol (350 mL) and subjected to HCl (2N, 94 mL,
10 0.187 mmol). Heat evolved. After 30 minutes, the volatiles were removed under vacuum. The product was purified using column chromatography (250 g, SiO₂, CHCl₃) and a gradient elution (100 CHCl₃ (80:20 CHCl₃/Acetone). 26.0 g of product was obtained (light brown oil). Purity by HPLC: 98.8%.

15

Testing:

All compounds were tested for stability by incubation in human plasma at 38°C for three hours and subsequent analysis by high pressure liquid chromatography. All compounds tested were found to be stable under these conditions.

20

All compounds were screened on a panel of normal and cancer human cell lines, including human prostate cancer cell lines, PC-3, DU-145, and LnCAP. The purpose of this experiment was to assess cell growth inhibition by measuring cytotoxicity and cytostatic effects.

Cells (10⁴/well) were plated on 96 well plates with the following controls:
25 no cells and toxic control (1 x 10⁻³M sodium dodecyl sulfate (SDS). The drug was diluted in ethanol and added directly to the wells. Plates were incubated at 37°C under 5% carbon dioxide in sterile air, in a humidified incubator for 72 hours. A solution (50 μl of 2,3-bis-(methoxy-4-nitro-5-sulfophenyl)-5-
[(phenylamino)carbonyl]-2H-tetrazolium hydroxide (XTT), 1 mg/mL) in phosphate
30 buffered saline (PBS, 100 mM) was added to each well, In the presence of viable cells, this colorless clear solution is enzymatically transformed to give a pink coloration, read at 450 nm using a microplate reader (Molecular Devices Thermomax). The inhibition of cell growth was measured by hemocytometer,

TABLE I

Inhibition of Cell Proliferation at 72 hours:
Cytotoxic Effects of the Selected Novel Anti-Androgens.

5

10

15

20

Cell Line	Tumor	IC ₅₀ (M)		
		BP-82	BP-196	BP-199
DU-145	Human Prostate (receptor poor)	1.39 x 10 ⁻⁵	8.67 x 10 ⁻⁷	8.51 x 10 ⁻⁵
Ln CAP	Human Prostate (with androgen receptors)	6.60 x 10 ⁻⁵	1.31 x 10 ⁻⁷	8.20 x 10 ⁻⁷
PC-3	Human Prostate (few androgen receptors)	3.15 x 10 ⁻⁵	3.72 x 10 ⁻⁸	1.32 x 10 ⁻⁷
MCF-7	Human Breast	5.00 x 10 ⁻⁵	9.89 x 10 ⁻⁷	1.00 x 10 ⁻⁴
MCF-7/ADR	Human Breast (adriamycin resistant)	1.51 x 10 ⁻⁵	1.00 x 10 ⁻⁵	1.00 x 10 ⁻⁵
Ovcar 3	Human Ovary	9.65 x 10 ⁻⁵	5.00 x 10 ⁻⁸	> 10 ⁻⁴
Molt-4	Human T-cell Leukemia	4.88 x 10 ⁻⁵	1.47 x 10 ⁻⁷	> 10 ⁴
L-1210	Mouse Leukemia	2.50 x 10 ⁻⁵	9.70 x 10 ⁻⁷	1.10 x 10 ⁵
	<u>Normal</u>			
NH DF	Dermal Fibroblast (human)	9.17 x 10 ⁻⁵	1.07 x 10 ⁻⁷	>10 ⁻⁴
HLF-1	Normal Lung Diploid (human)	3.90 x 10 ⁻⁵	8.06 x 10 ⁻⁶	>10 ⁻⁴
CHO	Chinese Hamster Ovary	3.45 x 10 ⁻⁵	8.76 x 10 ⁻⁶	1.28 x 10 ⁻⁵

TABLE III

Anti-androgenic potency (IC_{50}) of current
and novel anti-androgens.

5

Transactivation assay in CV1-3.9.2 cells;
Stimulation with 0.1 nM testosterone)

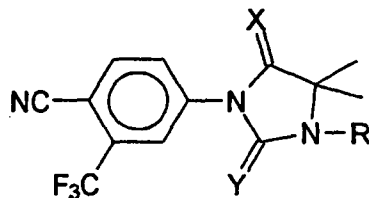
COMPOUND	IC_{50} [nM]
10 Cyproterone Acetate	11
RU59063†	23
Hydroxyflutamide	35 (Binding Affinity [Kf]*=280
Casodex	180
BP134	21
15 BP135	158
BP136	200
BP137	20
BP138	139
BP139	239
20 BP199	15 (Binding Affinity [Kf]*=5
BP82	= 6.5
BP163	217
BP 307	7 (Binding Affinity [Kf]*=24
BP 305	100 (Binding Affinity [Kf]*=15
25 BP 306	10 (Binding Affinity [Kf]*=23
BP 82	~6.5 (Binding Affinity [Kf]*=28
BP 231	260 (Binding Affinity [Kf]*=56
BP 328	NA (Binding Affinity [Kf]*=52
BP 218	NA
30 BP 332	NA

* Kf=competition factor, Kf=1=same as R1881
†Described by Teutsch, (Ref. 1)

WHAT IS CLAIMED IS:

1. A compound of the formula:

5



10

wherein:

X is oxygen or nitrogen, with the proviso that when R is amino, oxy or iodo substituted aryl, X is sulfur, oxygen or nitrogen;

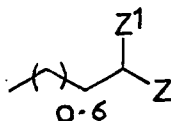
Y is sulphur, with the proviso that when R is said aryl group, Y is sulphur, oxygen or nitrogen;

R is an organic group comprising an aliphatic linking group of from 0 to 2 oxy groups, 0 to 1 amino group, 0 to 1 halo group, or 0 to 1 imidazolyl group, wherein said oxy groups, said amino group and said imidazolyl group have from 0 to 1 substituent.

2. A compound according to Claim 1, wherein R comprises an annular ring amino or oxy substituted aralkyl group for iodination or a polyiodoaralkyl group, wherein said aryl portion is linked to said alkyl portion by a carbon-carbon bond or through a heteroatom.

3. A compound according to Claim 1, wherein R is of the formula:

30



5

wherein:

Z is hydroxyl, amino, substituted amino, halo or 4-diazolyl;

Z¹ is hydrogen, hydroxyl, or may be taken together with Z to provide for olefinic or acetylenic unsaturation, or a 2,2-dimethyldioxalane.

10

4. A compound according to Claim 3, wherein Z and Z¹ are taken together.

5. A compound according to Claim 3, wherein Z is hydroxyl.

15 6. A compound according to Claim 3, wherein Z is amino.

7. A compound according to Claim 3, wherein Z is monosubstituted amino, and said amino substituent being acyl or alkyl of from one to ten carbon atoms.

20 8. A compound according to Claim 3, wherein Z is monosubstituted amino, and said amino substituent is a chelating group.

9. A compound according to Claim 3, wherein Z is monosubstituted amino, and said amino substituent is an antibiotic.

25

10. A compound according to Claim 9, wherein said antibiotic is paclitaxel.

11. A compound according to Claim 3, wherein Z is a substituted amino group, wherein the substituent of said substituted amino group is a polyiodoaryl group.

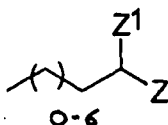
30

12. In a method for specifically directing an agent to cells comprising an androgenic receptor by adding said agent to a mammalian host comprising said cells, the improvement which comprises:

said agent being a compound according to Claim 1.

13. A method according to Claim 12, wherein said substituent is an antibiotic.
- 5 14. A method according to Claim 12, wherein said substituent comprises a radioactive atom or heavy atom.
15. A method according to Claim 1, wherein R is of the formula:

10



15 wherein:

Z is hydroxyl, amino, halo or 4-diazolyl;

Z¹ is hydrogen, hydroxyl, or may be taken together with Z to provide for olefinic or acetylenic unsaturation, or a 2,2-dimethyldioxalane.

- 20 16. A compound selected from the group consisting of: 4-[3'-(2"-propenyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidiny]-2-trifluoromethyl-benzonitrile; 4-[3'-(2"-(N-t-butoxycarbonyl)-aminoethyl)-4',4'-dimethyl-5'-imino-2'-thioxo-1'-imidazolidiny]-2-trifluoromethyl-benzonitrile; 4-[3'-(2"-N-acetylaminoethyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidiny]-2-trifluoromethyl-benzonitrile; 4-[3'-(2"-propynyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidiny]-2-trifluoromethyl-benzonitrile; 4-[3'-trans-(2"-propenyl-3"-*iodo)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidiny]-2-trifluoromethyl-benzonitrile; 4-[3'-cis-(2"-propenyl-3"-iodo)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidiny]-2-trifluoromethyl-benzonitrile; 4-[3'-(6"-thiohexyl)hexyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidiny]-2-trifluoromethyl-benzonitrile; 4-[3'-(2"-{4'''-(2'''*iodo)imidazolyl}ethyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidiny]-2-trifluoromethyl-benzonitrile; 4-[3'-(4"-fluorobutyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidiny]-2-trifluoromethyl-benzonitrile; 4-[3'-trans-(2"-propenyl-3"-tributylstanny)-4',4'-
- 25
- 30

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US96/10286

A. CLASSIFICATION OF SUBJECT MATTER																				
IPC(6) :A61K 31/415; C07D 233/84, 233/86, 233/72, 233/88, 405/04, 405/06. US CL :514/386, 342, 391; 548/311.1, 317.1, 318.5, 320.1, 320.5. According to International Patent Classification (IPC) or to both national classification and IPC																				
B. FIELDS SEARCHED																				
Minimum documentation searched (classification system followed by classification symbols) U.S. : 514/386, 342, 391; 548/311.1, 317.1, 318.5, 320.1, 320.5.																				
Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched Please See Extra Sheet.																				
Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)																				
C. DOCUMENTS CONSIDERED TO BE RELEVANT																				
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.																		
X	US 5,411,981 A (GAILLARD-KELLY ET AL.) 02 May 1995, examples 22, 23, 25, 31, 32, 43-46, 58, 65, 69-81, and 84-91 as well as column 46, line 55 to column 47, line 28.	1-16																		
X	EP 0,494,819 A1 (ROUSSEL-UCLAF) 15 JULY 1992, examples 22, 23, 25, 31, 32, and 43-46 as well as page 28, line 15 to page 29, line 8.	1-16																		
A	EP 0,436,426 B1 (ROUSSEL-UCLAF) 10 July 1991, entire document.	1-16																		
<input type="checkbox"/> Further documents are listed in the continuation of Box C. <input type="checkbox"/> See patent family annex.																				
<table border="0"> <tr> <td>* Special categories of cited documents:</td> <td>"T"</td> <td>later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention</td> </tr> <tr> <td>"A" document defining the general state of the art which is not considered to be of particular relevance</td> <td>"X"</td> <td>document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone</td> </tr> <tr> <td>"E" earlier document published on or after the international filing date</td> <td>"Y"</td> <td>document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art</td> </tr> <tr> <td>"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)</td> <td>"&"</td> <td>document member of the same patent family</td> </tr> <tr> <td>"O" document referring to an oral disclosure, use, exhibition or other means</td> <td></td> <td></td> </tr> <tr> <td>"P" document published prior to the international filing date but later than the priority date claimed</td> <td></td> <td></td> </tr> </table>			* Special categories of cited documents:	"T"	later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention	"A" document defining the general state of the art which is not considered to be of particular relevance	"X"	document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone	"E" earlier document published on or after the international filing date	"Y"	document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art	"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	"&"	document member of the same patent family	"O" document referring to an oral disclosure, use, exhibition or other means			"P" document published prior to the international filing date but later than the priority date claimed		
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Date of the actual completion of the international search 04 SEPTEMBER 1996		Date of mailing of the international search report 04 OCT 1996																		
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(54) **ANTI-ANDROGENIC PYRROLIDINES WITH TUMOR-INHIBITING ACTION**

Publication Classification

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(51) **Int. Cl.⁷** **C07D 43/02; A61K 31/4178**

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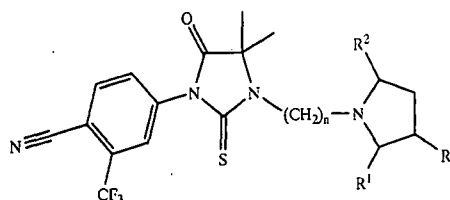
(57) **ABSTRACT**

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This invention relates to anti-androgenic N-[ω-[3-[4-cyano-3-(trifluoromethyl)-phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]alkyl]-substituted pyrrolidines of general formula I,

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(22) **Filed:** May 10, 2004



Related U.S. Application Data

(60) Provisional application No. 60/470,182, filed on May 14, 2003.

(30) **Foreign Application Priority Data**

May 9, 2003 (DE)..... 103 22 108.5

with a strongly pronounced antiproliferative profile of action; process for the production of the compounds of general formula I, pharmaceutical preparations and the use for the production of pharmaceutical agents.

ANTI-ANDROGENIC PYRROLIDINES WITH TUMOR-INHIBITING ACTION

[0001] This application claims the benefit of the filing date of U.S. Provisional Application Ser. No. 60/470,182 filed May 14, 2003.

[0002] This invention relates to anti-androgenic N-[ω -[3-[4-cyano-3-(trifluoromethyl)-phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]alkyl]-substituted pyrrolidines, with a strongly pronounced antiproliferative action profile, process for their production, as well as pharmaceutical preparations that contain the pyrrolidines according to the invention and their use for the production of pharmaceutical agents.

[0003] In industrialized countries, prostate cancer, after lung cancer, is the second main cause of death by cancer in men. In men over 55 years of age, 4% of all deaths can be attributed to a prostate tumor disease, and it is expected that the proportion in men over 80 increases to up to 80% of deaths. The death rate is still relatively low, but it increases yearly by about 14%. The number of men in whom a prostate tumor was diagnosed has increased by 30% in recent years, which can be attributed less to an increasing number of new diseases but rather that the population is generally older, that diagnostic processes have improved and that systematic screening programs were introduced (E. J. Small, D. M. Reese, *Curr. Opin. Oncol.* 2000, 12, 265-272).

[0004] The prostate tumor grows in an androgen-dependent manner in early stages. As long as the tumor is locally limited to the prostate, it can be removed by surgical intervention or by radiation therapy, whereby these methods are associated with corresponding risks. In the cases in which the tumor is no longer locally limited and has already formed metastases, the tumor is treated palliatively by reduction of the testosterone level in the blood. This is carried out either surgically by castration or medicinally by treatment with anti-androgens (bicalutamide, cyproterone acetate, flutamide), LHRH-agonists (buserelin, zoladex), LHRH-antagonists (cetorelix) or 5 α -reductase inhibitors (finasteride). Since the adrenal androgen synthesis remains unaffected in a surgical castration, more recently a combined surgical and medicinal treatment is frequently performed (S. Leewansangtong, E. D. Crawford, *Endocrine-Related Cancer* 1998, 5, 325-339). This treatment, however, has only temporary success, since after at the latest two years, it generally results in renewed growth of the tumor, which in most cases is then hormone-independent (L. J. Denis, K. Griffith, *Semin. in Surg. Onc.* 2000, 18, 52-74). Despite intensive research in the last 50 years, up until now there has been no effective treatment against these advanced stages. The 5-year survival rate in these patients is less than 15%.

[0005] There are different indications that show that the androgen receptor plays an important role in the development and the growth of the prostate tumor not only in the early hormone-dependent stages of the tumor progression, but also in late hormone-independent stages of the tumor progression.

[0006] The androgen receptor belongs to the family of steroid hormone receptors that act as transcription factors. The androgen receptor binds androgens, by which it is stabilized and protected from a quick proteolytic degradation. After hormone bonding, it is transported into the

nucleus, where it activates certain genes by binding to so-called androgen-responsive DNA elements that are in promoter regions (D. J. Lamb et. al. *Vitam. Horm.* 2001, 62, 199-230).

[0007] Studies on prostate tumors show that in 30% of advanced tumors, an amplification of the androgen receptor gene locus was detected. In other cases, a number of mutations were found in the androgen receptor gene, which are localized in various domains of the androgen receptor molecule and result in altered receptor properties. Mutated receptors can either have a higher affinity for androgens, be constitutively active, change their ligand specificity, so that they are activated by other steroid hormones or even anti-androgens, be activated via interactions with molecules from other growth-promoting signal-transmitting methods, change the interaction with cofactors, or activate other target genes (J. P. Elo, T. Visakorpi, *Ann. Med.* 2001, 33, 130-41).

[0008] The identification of anti-androgens, which inhibit not only the natural androgen receptor but also its mutated forms and have an enhanced antiproliferative effect on tumor cells, would presumably be very helpful in treating prostate tumors in various stages. Such compounds can significantly change the period until the tumor growth recurs.

[0009] Studies with nonsteroidal anti-androgens have shown that they have advantages compared to the steroidal compounds and are therefore to be preferred. Thus, with nonsteroidal compounds, a more selective action with fewer adverse side effects can be achieved. In contrast to the steroidal anti-androgens, the known nonsteroids bicalutamide and flutamide lack, e.g., the progestagenic activity, and in addition, their use results in an increase in the testosterone level in the serum, which clinically could result in development of potency (P. Reid, P. Kantoff, W. Oh, *Investigational New Drugs* 1999, 17, 271-284).

[0010] Nonsteroidal anti-androgens are described in U.S. Pat. No. 5,411,981 or U.S. Pat. No. Re. 35,956 (phenylimidazolidine derivatives), in WO 97/00071 (specifically substituted phenyldimethyl hydantoins as well as their imino- or thione derivatives), in WO 00/37430 (phenylalanine, phenyl hydantoins as well as phenyl ureas), in WO 01/58855 (aminopropanilides) and in EP 1122242 (substituted cyanophenylpiperazine describes, i.a., [4-cyano-3-(trifluoromethyl)phenyl]-substituted thiohydantoins with a short-chain, terminally substituted radical, whereby in the case of the chain, this is preferably a C₁-C₄-chain.

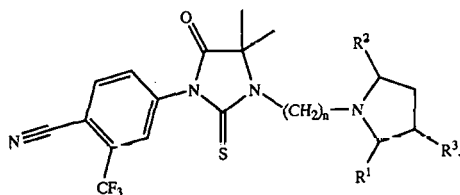
[0011] The compounds that are explicitly disclosed in U.S. Pat. No. Re. 35,956 have an anti-androgenic action, but only a slightly antiproliferative action in cells that originate from human prostate cancers.

[0012] For an effective therapy of androgen-dependent tumors and/or other proliferative diseases, an additional antiproliferative action is necessary.

[0013] The object of this invention therefore consists in making available orally bioavailable anti-androgenic compounds with increased antiproliferative action that can inhibit the growth of androgen-dependent benign or malignant tumors or alleviate or heal androgen-dependent proliferative diseases.

[0014] This object is achieved according to the invention by the N-[ω -[3-[4-cyano-3-(trifluoromethyl)phenyl]-5,5-

dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]alkyl]-substituted pyrrolidines of general formula I:



[0015] in which

[0016] n can mean an integer between 6 and 9,

[0017] R¹ and R², independently of one another, can mean a hydrogen atom, an unbranched C₁-C₄-alkyl group, a branched C₃-C₅-alkyl group, an unbranched hydroxy-C₁-C₄-alkyl group, a branched hydroxy-C₃-C₅-alkyl group, an unbranched C₁-C₄-alkoxy-C₁-C₄-alkyl group, a branched C₃-C₅-alkoxy-C₃-C₅-alkyl group, an unbranched C₁-C₄-alkanoyloxy-C₁-C₄-alkyl group, a branched C₃-C₅-alkanoyloxy-C₃-C₅-alkyl group, a (pyrrolidin-1-yl)methyl group, a carboxy group, a C₁-C₄-alkoxycarbonyl group or an aminocarbonyl group,

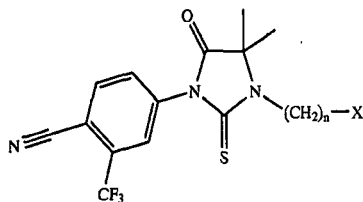
[0018] or

[0019] R¹ and R² together can mean a 2-hydroxypropane-1,3-diyl bridge;

[0020] R³ can mean a hydrogen atom or a hydroxy group.

[0021] It was found that the antiproliferative action of compounds of the N-[3-[4-cyano-3-(trifluoromethyl)phenyl]-5,5-dimethylthiohydantoin type can be increased, surprisingly enough, while retaining anti-androgenic activity, if the N-1-nitrogen carries a pyrrolidin-1-ylalkyl substituent. The compounds according to the invention are distinguished by an alkylene chain of a defined length range, which connects the pyrrolidine nucleus to the thiohydantoin nucleus. Depending on the combination of the heterocyclic end group with the length of the alkylene chain that connects the latter with the thiohydantoin nucleus, a more or less pronounced additional effect that results in the destabilization of the androgen receptor can occur.

[0022] This invention comprises a process for the production of the compounds of general formula I according to the invention, in which compounds of general formula II

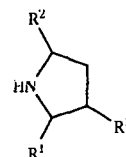


[0023] in which

[0024] n can mean an integer between 6 and 9,

[0025] X can mean a leaving group,

[0026] are reacted in the presence of an organic base with compounds of general formula III



[0027] in which

[0028] R¹ and R², independently of one another, can mean a hydrogen atom, an unbranched C₁-C₄-alkyl group, a branched C₃-C₅-alkyl group, an unbranched hydroxy-C₁-C₄-alkyl group, a branched hydroxy-C₃-C₅-alkyl group, an unbranched C₁-C₄-alkoxy-C₁-C₄-alkyl group, a branched C₃-C₅-alkoxy-C₃-C₅-alkyl group, an unbranched C₁-C₄-alkanoyloxy-C₁-C₄-alkyl group, a branched C₃-C₅-alkanoyloxy-C₃-C₅-alkyl group, a (pyrrolidin-1-yl)methyl group, a carboxy group, a C₁-C₄-alkoxycarbonyl group or an aminocarbonyl group,

[0029] or

[0030] R¹ and R² together can mean a 2-hydroxypropane-1,3-diyl bridge;

[0031] R³ can mean a hydrogen atom or a hydroxy group.

[0032] Alkenyl chain —CH₂—_n is the n-hexylene, n-heptylene, n-octylene or n-nonylene group.

[0033] The unbranched hydroxy-C₁-C₄-alkyl group can be a hydroxymethyl-(HOCH₂—), 2-hydroxyethyl-(HOCH₂CH₂—), 1-hydroxyethyl-[CH₃CH(OH)—], 3-hydroxypropyl-(HOCH₂CH₂CH₂—), 2-hydroxypropyl-[CH₃CH(OH)CH₂CH₂—], 1-hydroxypropyl-[CH₃CH₂CH(OH)—], 4-hydroxybutyl-(HOCH₂CH₂CH₂CH₂—), 3-hydroxybutyl-[CH₃CH(OH)CH₂CH₂—], 2-hydroxybutyl-[CH₃CH₂CH(OH)CH₂—] or 1-hydroxybutyl-[CH₃CH₂CH₂CH(OH)] group.

[0034] The branched hydroxy-C₃-C₅-alkyl group can be a 1-hydroxy-1-methylethyl-[(CH₃)₂C(OH)—], 2-hydroxy-1-methylethyl-[HOCH₂CH(CH₃)—], 1-hydroxy-1-methylpropyl-[CH₃CH₂C(CH₃)(OH)—], 2-hydroxy-1-methylpropyl-[CH₃CH(OH)CH(CH₃)—], 3-hydroxy-1-methylpropyl-[HOCH₂CH₂CH(CH₃)—], 1-(hydroxymethyl)propyl-[CH₃CH₂C(CH₂OH)—], 1-hydroxy-2-methylpropyl-[(CH₃)₂CHCH(OH)—], 2-hydroxy-2-methylpropyl-[CH₃C(OH)(CH₃)CH₂—], 3-hydroxy-2-methylpropyl-[HOCH₂CH(CH₃)CH₂—], 1-(hydroxymethyl)butyl-[CH₃CH₂CH₂CH(CH₂OH)—], 1-hydroxy-1-methylbutyl-[CH₃CH₂CH₂C(CH₃)(OH)—], 2-hydroxy-1-methylbutyl-[CH₃CH₂CH(OH)CH(CH₃)—], 3-hydroxy-1-methylbutyl-[CH₃CH(OH)CH₂CH(CH₃)—], 4-hydroxy-1-methylbutyl-

[HOCH₂CH₂CH₂CH(CH₃)—], 3-hydroxy-1-ethylpropyl-
 [HOCH₂CH₂CH(CH₂CH₃)—], 2-hydroxy-1-ethylpropyl-
 [CH₃CH(OH)CH(CH₂CH₃)—], 1-hydroxy-1-ethylpropyl-
 [CH₃CH₂C(CH₂CH₃)(OH)—], 1-hydroxy-3-methylbutyl-
 [CH₃CH(CH₃)CH₂CH(OH)—], 2-hydroxy-3-methylbutyl-
 [(CH₃)₂CHCH(OH)CH₂—], 3-hydroxy-3-methylbutyl-
 [CH₃C(CH₃)(OH)CH₂CH₂—], 4-hydroxy-3-methylbutyl-
 [HOCH₂CH(CH₃)CH₂CH₂—], 1-(hydroxymethyl)-1-
 methylpropyl-[CH₃CH₂C(CH₃)(CH₂OH)—], 2-hydroxy-1-
 1-dimethylpropyl-[CH₃CH(OH)C(CH₃)₂—], 3-hydroxy-1,1-
 dimethylpropyl-[HOCH₂CH₂C(CH₃)₂—],
 1-(hydroxymethyl)-2-methylpropyl-
 [(CH₃)₂CHCH(CH₂OH)—], 1-hydroxy-1,2-dimethylpro-
 pyl-[(CH₃)₂CHC(OH)(CH₃)—], 2-hydroxy-1,2-dimethyl-
 propyl-[(CH₃)₂C(OH)CH(CH₃)—], 3-hydroxy-1,2-
 dimethylpropyl-[HOCH₂CH(CH₃)CH(CH₃)—] 9
 1-hydroxy-2,2-dimethylpropyl-[(CH₃)₃CCH(OH)—], 3-hy-
 droxy-2,2-dimethylpropyl-[HOCH₂C(CH₃)₂CH₂—], 1-hy-
 droxy-2-methylbutyl-[CH₃CH₂CH(CH₃)CH(OH)—], 2-hy-
 droxy-2-methylbutyl-[CH₃CH₂C(CH₃)(OH)CH₂—],
 3-hydroxy-2-methylbutyl-[CH₃CH(OH)CH(CH₃)CH₂—],
 4-hydroxy-2-methylbutyl-[HOCH₂CH₂CH(CH₃)CH₂—] or
 3-hydroxy-2-ethylpropyl-[HOCH₂CH(CH₂CH₃)CH₂—] group.

[0035] The unbranched C₁-C₄-alkoxy-C₁-C₄-alkylene group can be an alkoxyethyl-(AlkOCH₂—), 2-alkoxyethyl-(AlkOCH₂CH₂—), 1-alkoxyethyl-(CH₃CH(OAlk)—), 3-alkoxypropyl-(AlkOCH₂CH₂CH₂—), 2-alkoxypropyl-[CH₃CH(OAlk)CH₂CH₂—], 1-alkoxypropyl-[CH₃CH₂CH(OAlk)—], 4-alkoxybutyl-(AlkOCH₂CH₂CH₂CH₂—), 3-alkoxybutyl-[CH₃CH(OAlk)CH₂CH₂—], 2-alkoxybutyl-[CH₃CH₂CH(OAlk)CH₂—] or 1-alkoxybutyl-[CH₃CH₂CH₂CH(OAlk)—] group.

[0036] The branched C₁-C₄-alkoxy-C₃-C₅-alkyl group can be a 1-alkoxy-1-methylethyl-[(CH₃)₂C(OAlk)—], 2-alkoxy-1-methylethyl-[AlkOCH₂CH(CH₃)—], 1-alkoxy-1-methylpropyl-[CH₃CH₂C(CH₃)(OAlk)—], 2-alkoxy-1-methylpropyl-[CH₃CH(OAlk)CH(CH₃)—], 3-alkoxy-1-methylpropyl-[AlkOCH₂CH₂CH(CH₃)—], 1-(alkoxymethyl)propyl-[CH₃CH₂C(CH₂OAlk)—], 1-alkoxy-2-methylpropyl-[(CH₃)₂CHCH(OAlk)—], 2-alkoxy-2-methylpropyl-[CH₃C(OAlk)(CH₃)CH₂—], 3-alkoxy-2-methylpropyl-[AlkOCH₂CH(CH₃)CH₂—], 1-(alkoxymethyl)butyl-[CH₃CH₂CH(CH₂OAlk)—], 1-alkoxy-1-methylbutyl-[CH₃CH₂CH₂C(CH₃)(OAlk)—], 2-alkoxy-1-methylbutyl-[CH₃CH₂CH(OAlk)CH(CH₃)—], 3-alkoxy-1-methylbutyl-[CH₃CH(OAlk)CH₂CH(CH₃)—], 4-alkoxy-1-methylbutyl-[AlkOCH₂CH₂CH₂CH(CH₃)—], 3-alkoxy-1-ethylpropyl-[AlkOCH₂CH₂CH(CH₂CH₃)—], 2-alkoxy-1-ethylpropyl-[CH₃CH(OAlk)CH(CH₂CH₃)—], 1-alkoxy-1-ethylpropyl-[CH₃CH₂C(CH₂CH₃)(OAlk)—], 1-alkoxy-3-methylbutyl-[CH₃CH(CH₃)CH₂CH(OAlk)—], 2-alkoxy-3-methylbutyl-[(CH₃)₂CHCH(OAlk)CH₂—], 3-alkoxy-3-methylbutyl-[CH₃C(CH₃)(OAlk)CH₂CH₂—], 4-alkoxy-3-methylbutyl-[AlkOCH₂CH(CH₃)CH₂CH₂—], 1-(alkoxymethyl)-1-methylpropyl-[CH₃CH₂C(CH₃)(CH₂OAlk)—], 2-alkoxy-1,1-dimethylpropyl-[CH₃CH(OAlk)C(CH₃)₂—], 3-alkoxy-1,1-dimethylpropyl-[AlkOCH₂CH₂C(CH₃)₂—], 1-(alkoxymethyl)-2-methylpropyl-[(CH₃)₂CHCH(CH₂OAlk)—], 1-alkoxy-1,2-dimethylpropyl-[(CH₃)₂CHC(OAlk)(CH₃)—], 2-alkoxy-1,2-dimethylpropyl-[(CH₃)₂C(OAlk)CH(CH₃)—], 3-alkoxy-1,2-dimethylpropyl-[AlkOCH₂CH(CH₃)—CH(CH₃)—], 1-alkoxy-2,2-dimethylpropyl-[(CH₃)₃CCH(OAlk)—], 3-alkoxy-2,2-dimethylpropyl-[AlkOCH₂C(CH₃)₂CH₂—],

1-alkoxy-2-methylbutyl-[CH₃CH₂CH(CH₃)CH(OAlk)—], 2-alkoxy-2-methylbutyl-[CH₃CH₂C(CH₃)(OAlk)CH₂—], 3-alkoxy-2-methylbutyl-[CH₃CH(OAlk)CH(CH₃)CH₂—], 4-alkoxy-2-methylbutyl-[AlkOCH₂CH₂CH(CH₃)CH₂—] or 3-alkoxy-2-ethylpropyl-[AlkOCH₂CH(CH₂CH₃)CH₂—] group.

[0037] The C₁-C₄-alkoxy group can be, for example, a methoxy-, ethoxy-, n-propoxy-, iso-propoxy-, n-butoxy-, sec-butoxy-, iso-butoxy- or tert-butoxy group.

[0038] The C₁-C₄-alkanoyl group can be, for example, a formyl-, acetyl-, propanoyl-, butanoyl- or iso-butanoyl group.

[0039] The unbranched C₁-C₄-alkanoyloxy-C₁-C₄-alkylene group can be an alkanoyloxymethyl-(AlkCOOCH₂—), 2-alkanoyloxyethyl-(AlkCOOCH₂CH₂—), 1-alkanoyloxyethyl-[CH₃CH(OCOAlk)—], 3-alkanoyloxypropyl-(AlkCOOCH₂CH₂CH₂—), 2-alkanoyloxypropyl-[CH₃CH(OCOAlk)CH₂CH₂—], 1-alkanoyloxypropyl-[CH₃CH₂CH(OCOAlk)—], 4-alkanoyloxybutyl-(AlkCOOCH₂CH₂CH₂CH₂—), 3-alkanoyloxybutyl-[CH₃CH(OCOAlk)CH₂CH₂—], 2-alkanoyloxybutyl-[CH₃CH₂CH(OCOAlk)CH₂—] or 1-alkanoyloxybutyl-[CH₃CH₂CH₂CH(OCOAlk)—] group.

[0040] The branched C₃-C₄-alkanoyloxy-C₃-C₅-alkyl group can be a 1-alkanoyloxy-1-methylethyl-[(CH₃)₂C(OCOAlk)—], 2-alkanoyloxy-1-methylethyl-[AlkCOOCH₂CH(CH₃)—], 1-alkanoyloxy-1-methylpropyl-[CH₃CH₂C(CH₃)(OCOAlk)—], 2-alkanoyloxy-1-methylpropyl-[CH₃CH(OCOAlk)CH(CH₃)—], 3-alkanoyloxy-1-methylpropyl-[AlkCOOCH₂CH₂CH(CH₃)—], 1-(alkanoyloxymethyl)propyl-[CH₃CH₂C(CH₂OCOAlk)—], 1-alkanoyloxy-2-methylpropyl-[(CH₃)₂CHCH(OCOAlk)—], 2-alkanoyloxy-2-methylpropyl-[CH₃C(OCOAlk)(CH₃)CH₂—], 3-alkanoyloxy-2-methylpropyl-[AlkCOOCH₂CH(CH₃)CH₂—], 1-(alkanoyloxymethyl)butyl-[CH₃CH₂CH₂CH(CH₂OCOAlk)—], 1-alkanoyloxy-1-methylbutyl-[CH₃CH₂CH₂C(CH₃)(OCOAlk)—], 2-alkanoyloxy-1-methylbutyl-[CH₃CH₂CH(OCOAlk)CH(CH₃)—], 3-alkanoyloxy-1-methylbutyl-[CH₃CH(OCOAlk)CH₂CH(CH₃)—], 4-alkanoyloxy-1-methylbutyl-[AlkCOOCH₂CH₂CH₂CH(CH₃)—], 3-alkanoyloxy-1-ethylpropyl-[AlkCOOCH₂CH₂CH(CH₂CH₃)—], 2-alkanoyloxy-1-ethylpropyl-[CH₃CH(OCOAlk)CH(CH₂CH₃)—], 1-alkanoyloxy-1-ethylpropyl-[CH₃CH₂C(CH₂CH₃)(OCOAlk)—], 1-alkanoyloxy-3-methylbutyl-[CH₃CH(CH₃)CH₂CH(OCOAlk)—], 2-alkanoyloxy-3-methylbutyl-[(CH₃)₂CHCH(OCOAlk)CH₂—], 3-alkanoyloxy-3-methylbutyl-[CH₃C(CH₃)(OCOAlk)CH₂CH₂—], 4-alkanoyloxy-3-methylbutyl-[AlkCOOCH₂CH(CH₃)CH₂CH₂—], 1-(alkanoyloxymethyl)-1-methylpropyl-[CH₃CH₂C(CH₃)(CH₂OCOAlk)—], 2-alkanoyloxy-1,1-dimethylpropyl-[CH₃CH(OCOAlk)C(CH₃)₂—], 3-alkanoyloxy-1,1-dimethylpropyl-[AlkCOOCH₂CH₂C(CH₃)₂—], 1-(alkanoyloxymethyl)-2-methylpropyl-[(CH₃)₂CHCH(CH₂OCOAlk)—], 1-alkanoyloxy-1,2-dimethylpropyl-[(CH₃)₂CHC(OCOAlk)(CH₃)—], 2-alkanoyloxy-1,2-dimethylpropyl-[(CH₃)₂C(OCOAlk)CH(CH₃)—], 3-alkanoyloxy-1,2-dimethylpropyl-[AlkCOOCH₂CH(CH₃)CH(CH₃)—], 1-alkanoyloxy-2,2-dimethylpropyl-[(CH₃)₃CCH(OCOAlk)—], 3-alkanoyloxy-2,2-dimethylpropyl-[AlkCOOCH₂C(CH₃)₂CH₂—], 1-alkanoyloxy-2-methylbutyl-

[CH₃CH₂CH(CH₃)CH(OCOAlk)-], 2-alkanoyloxy-2-methylbutyl-[CH₃CH₂C(CH₃)(OCOAlk)CH₂-], 3-alkanoyloxy-2-methylbutyl-[CH₃CH(OCOAlk)CH(CH₃)CH₂-], 4-alkanoyloxy-2-methylbutyl-[Alk-COOCH₂CH₂CH(CH₃)CH₂-] or 3-alkanoyloxy-2-ethylpropyl [Alk-COOCH₂CH(CH₂CH₃)CH₂-] group.

[0041] Leaving group X can be a halogen or a sulfonic ester group.

[0042] Halogen can be chlorine, bromine or iodine, whereby iodine is preferred.

[0043] The sulfonic ester group can be, for example, a mesylate, benzenesulfonate, tosylate, brosylate, triflate or nonaflate group.

[0044] The organic base is a tertiary amine or amide base, such as, for example, triethylamine or ethyl diisopropylamine.

[0045] For the formation of pharmaceutically compatible salts of the compounds of general formula I according to the invention, according to the methods that are known to one skilled in the art, i.a., hydrochloric acid, hydrobromic acid, sulfuric acid and phosphoric acid and nitric acid can be considered as inorganic acids; i.a., acetic acid, propionic acid, hexanoic acid, octanoic acid, decanoic acid, oleic acid, stearic acid, maleic acid, fumaric acid, succinic acid, benzoic acid, ascorbic acid, oxalic acid, salicylic acid, tartaric acid, citric acid, lactic acid, glycolic acid, malic acid, mandelic acid, cinnamic acid, glutaminic acid, and asparaginic acid can be considered as carboxylic acids; and i.a., methanesulfonic acid, ethanesulfonic acid, toluenesulfonic acid, benzenesulfonic acid as well as naphthalenesulfonic acid can be considered as sulfonic acids.

[0046] Examples 1 to 58 of the compounds according to the invention that are mentioned below under the Chapter "Production Process" are especially preferred.

[0047] Pharmacological Studies

[0048] The compounds according to the invention were tested in various models. The compounds of general formula I according to the invention are distinguished in that in this case, these are compounds with anti-androgenic action that inhibit prostate tumor growth, simultaneously have a high, optionally oral bioavailability and optionally destabilize the androgen receptor.

[0049] The in vitro tests on the influences on the activities of the androgen receptor were performed as follows:

[0050] In the diagrams presented here, the following abbreviations were used:

[0051] Bicalutamide: N-[4-Cyano-3-(trifluoromethyl)phenyl]-3-[(4-fluorophenyl)sulfonyl]-2-hydroxy-2-methylpropanamide

[0052] R1881: Methyltrienolone, 17 β -hydroxy-17 α -methylene-4,9,11-trien-3-one

[0053] CPA: Cyproterone acetate, 17-(acetyloxy)-6-chloro-1 β ,2 β -dihydro-3'H-cyclopropa[1,2]pregna-1,4,6-triene-3,20-dione

[0054] Model 1: Inhibition of the Proliferation of LNCaP Cells

[0055] For the proliferation assay, 6000 LNCaP cells/well (Horoszewicz et al. *Cancer Res.* 1983. 43. 1809-1817) in a microtiter plate (96-well) in 50 μ l of RPMI1640 medium are grown with 5% CCS and cultivated as in Model 1. After 24 hours, the cells receive 50 μ l of 2 \times -concentrated test substance, diluted in culture medium. The solvent concentration is 0.5% DMSO. After 4 days, the cells receive another 100 μ l of 1 \times -concentrated test substance, diluted in culture medium. After 7 to 8 days, the proliferation rate of the cells is determined by means of crystal violet assay (Gillies et al. *Anal. Biochem.* 1986, 159, 109-113). To determine the antagonism, the substance treatment is performed in the presence of 0.1 nmol of R1881 (1:1000 dilution of ethanolic solution). Control cells receive only 0.5% DMSO. For the agonism, the cells are treated only with test substance (without R1881).

[0056] Table 1 shows the inhibitory action of test substances on the proliferation of the human androgen-dependent prostate cell line LNCaP. The inhibition of the cell proliferation is an important requirement for the therapeutic use of the substances in the treatment of prostatic cancer. The selected test substances according to the invention inhibit the cell proliferation in the presence of 0.1 nmol of the synthetic androgen R1881 with a considerably lower IC₅₀ (about 50 \times 10⁻⁹ M), such as the approved nonsteroidal anti-androgen bicalutamide (380 \times 10⁻⁹ M). At a substance concentration of 1 μ mol, the proliferation compared to the cell growth in the presence of 0.1 nmol of R1881 is reduced by at least 80%. Up to a tested concentration of 10 μ mol, a proliferation-stimulating action was observed in none of the test substances

[0057] It has been found, surprisingly enough, that the extent of the proliferation-inhibiting action of N-[ω -{3-[4-cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl}alkyl] derivatives is simultaneously dependent on chain substituents and the chain length. Especially for the pyrrolidine-substituted compounds that are claimed here, the strongest antiproliferative action is observed in chain lengths between C₆ and C₉. Analogous compounds with shorter chain lengths n=4 (comparison 1) and n=5 (comparison 2) show a considerably reduced antiproliferative activity. For the tested examples from U.S. Pat. No. Re. 35,956 [Examples 71 and 77] of the chain length n=2 and 4, little or no antiproliferative action could be detected.

TABLE 1

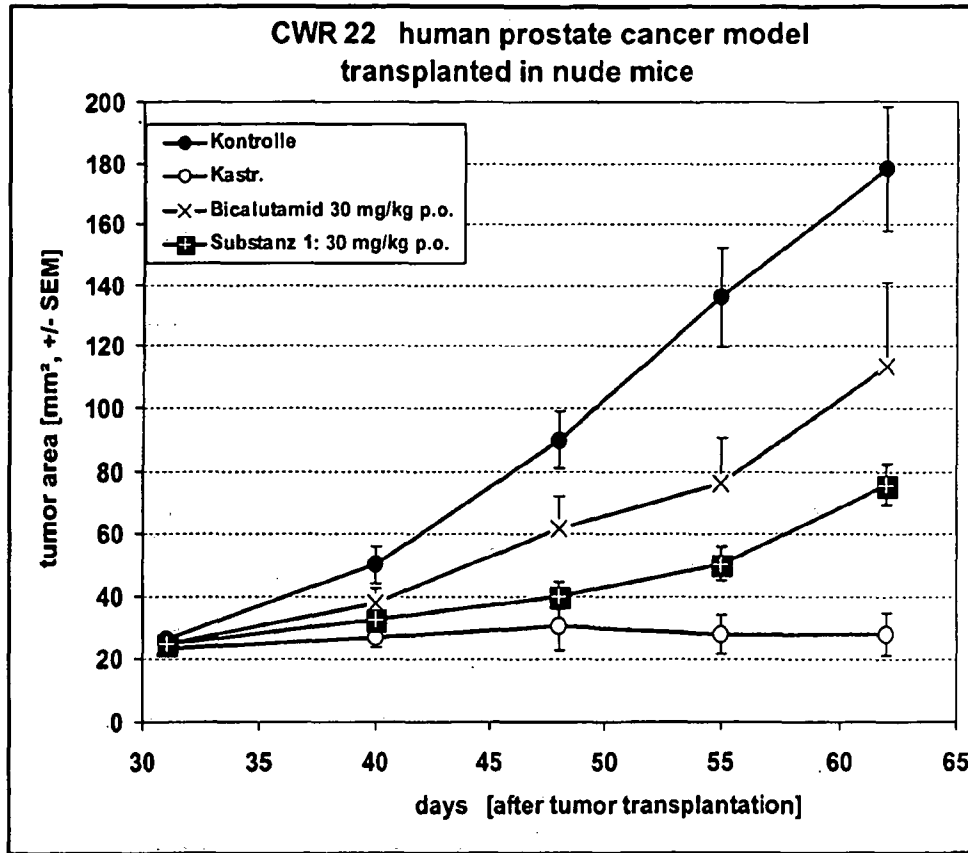
Example	Test Substance	IC ₅₀ [10 ⁻⁹ M]	% Inhibition at 1 μ M
Inhibition of the Proliferation of LNCaP Cells by Test Substances.			
Bicalutamide	N-[4-Cyano-3-(trifluoromethyl)phenyl]-3-[(4-fluorophenyl)sulfonyl]-2-hydroxy-2-methylpropanamide	380	85

TABLE 1-continued

Example	Test Substance	IC50 [10 ⁻⁹ M]	% Inhibition at 1 μM
Comparison 1	4-[4,4-Dimethyl-5-oxo-3-[4-(pyrrolidin-1-yl)butyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	4300	20
Comparison 2	4-[4,4-Dimethyl-5-oxo-3-[5-(pyrrolidin-1-yl)pentyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	2100	38
Example 71 USRe 35956	4-[3-(2-Hydroxyethyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	>10000	9
Example 77 USRe 35956	4-[3-(4-Hydroxybutyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	>10000	34
1	4-[4,4-Dimethyl-5-oxo-3-[8-(pyrrolidin-1-yl)octyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	45	98
2	4-[4,4-Dimethyl-5-oxo-3-[8-(pyrrolidin-1-yl)octyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile hydrochloride	41	100
4	4-[4,4-Dimethyl-5-oxo-3-[7-(pyrrolidin-1-yl)heptyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	45	98
6	4-[3-[6-[(2R)-2-(Hydroxymethyl)pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	55	102
8	4-[3-[7-[(2R)-2-(Hydroxymethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	11	101
9	4-[3-[7-[(2S)-2-(Hydroxymethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	26	92
10	4-[3-[8-[(2R)-2-(Hydroxymethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	<10	101
11	4-[3-[8-[(2S)-2-(Hydroxymethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	13	102
12	4-[3-[9-[(2R)-2-(Hydroxymethyl)pyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	24	93
13	4-[3-[9-[(2S)-2-(Hydroxymethyl)pyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	16	88
16	4-[3-[7-[(2R)-2-(Methoxymethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	18	98
17	4-[3-[7-[(2S)-2-(Methoxymethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	32	99
18	4-[3-[8-[(2R)-2-(Methoxymethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	11	103
19	4-[3-[8-[(2S)-2-(Methoxymethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	25	97
20	4-[3-[9-[(2R)-2-(Methoxymethyl)pyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	29	101
21	4-[3-[9-[(2S)-2-(Methoxymethyl)pyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	47	94
23	4-[3-[7-(3-Hydroxy-8-azabicyclo[3.2.1]oct-8-yl)heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	48	79
24	4-[3-[8-(3-Hydroxy-8-azabicyclo[3.2.1]oct-8-yl)octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	13	102

TABLE 1-continued

Example	Test Substance	IC50 [10 ⁻⁹ M]	% Inhibition at 1 μM
<u>Inhibition of the Proliferation of LNCaP Cells through Test Substances.</u>			
27	4-[3-[7-[(R)-3-Hydroxypyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	57	95
28	4-[3-[8-[(R)-3-Hydroxypyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	11	102
29	4-[3-[9-[(R)-3-Hydroxypyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	35	99
31	4-[3-[7-[(S)-3-Hydroxypyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	20	104
32	4-[3-[8-[(S)-3-Hydroxypyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	12	101
35	4-[4,4-Dimethyl-3-[7-(2-methylpyrrolidin-1-yl)heptyl]-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	30	92
36	4-[4,4-Dimethyl-3-[8-(2-methylpyrrolidin-1-yl)octyl]-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	15	96
37	4-[3-[8-[(2R,5S)-rel-2,5-Dimethylpyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	11	99
39	4-[3-[7-[(2S)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	41	108
40	4-[3-[8-[(2S)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	11	118
41	4-[3-[9-[(2S)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	48	89
<u>Inhibition of the Proliferation of LNCaP Cells by Test Substances.</u>			
43	4-[3-[7-[(2R)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	26	113
44	4-[3-[8-[(2R)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	11	115
47	4-[3-[8-[(2S)-2-(1-Hydroxy-1-ethylpropyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	27	99
49	(S)-1-[6-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]hexyl]pyrrolidine-4-carboxamide	32	110
50	(S)-1-[7-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]heptyl]pyrrolidine-4-carboxamide	15	110
51	(S)-1-[8-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]octyl]pyrrolidine-4-carboxamide	20	97
52	(R)-1-[6-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]hexyl]pyrrolidine-4-carboxamide	12	100
53	(R)-1-[7-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]heptyl]pyrrolidine-4-carboxamide	49	97
54	(R)-1-[8-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]octyl]pyrrolidine-4-carboxamide	64	93



[0067] While the tumor grows quickly in the untreated control animals, treatment with the compounds according to the invention results in a considerable growth inhibition of the prostate tumors, which is more strongly pronounced than

in the animals that are treated with bicalutamide. Both in the LNCaP tumor and in the CWR22 tumor, this growth inhibition is comparable to the effects of castration (Diagrams 1-2).

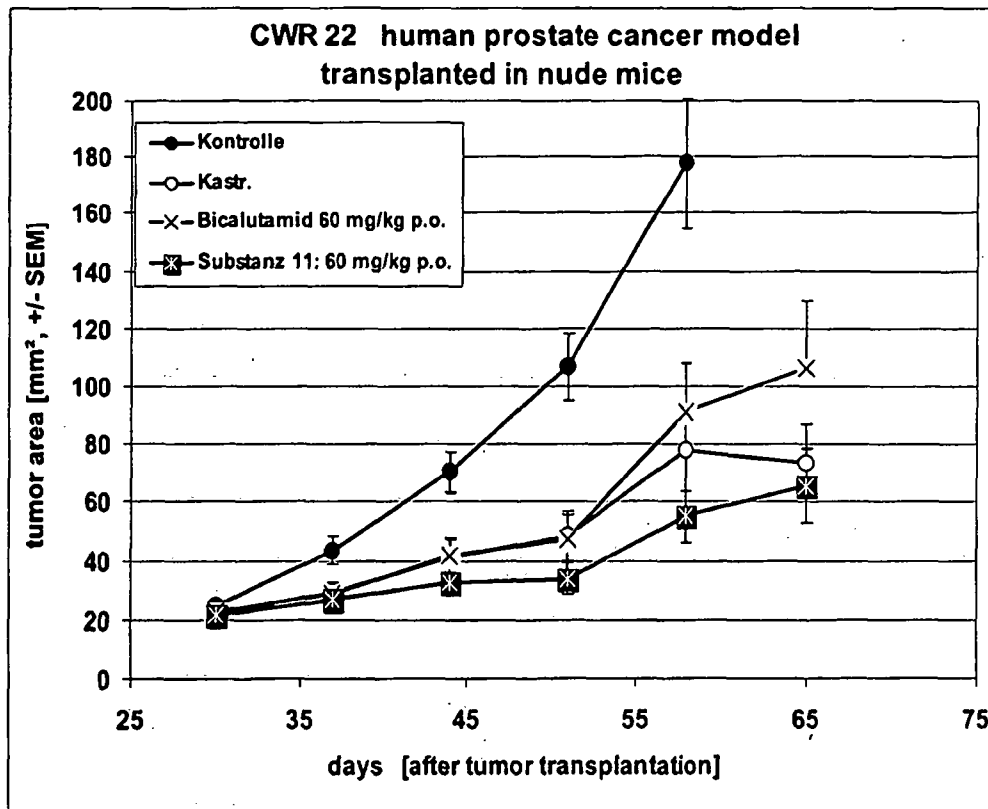
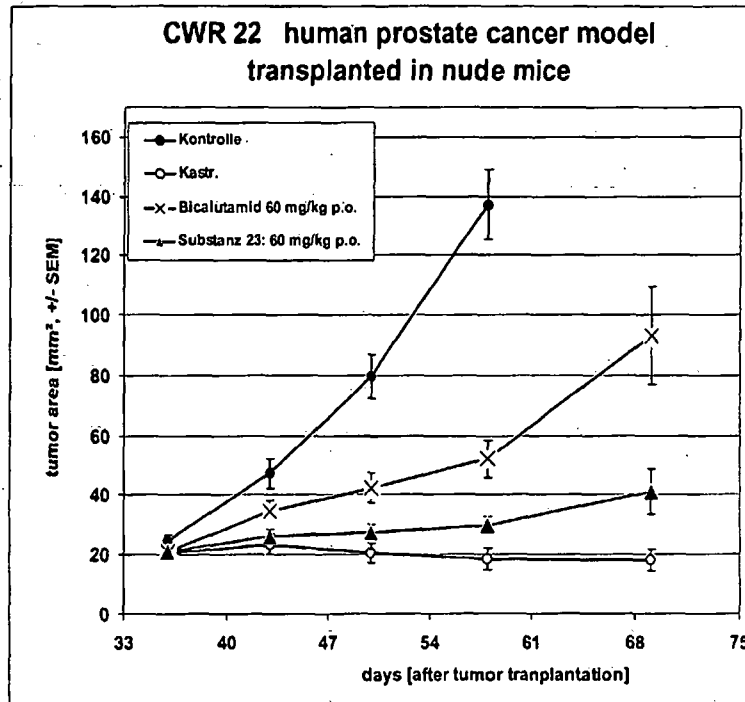


Diagram 4. Growth inhibition of CWR22 prostate cancers by substance according to Example 23. The treatment was carried out 1 x daily p.o. with 60 mg/kg. For comparison, bicalutamide as a reference substance was administered 1 x daily at 60 mg/kg.



[Key:]

Kontrolle = Control

Kastr. = Castr.

Bicalutamid = Bicalutamide

Substanz = Substance

[0069] While the tumor grows quickly in the untreated control animals, treatment with the compounds according to the invention results in a significant growth inhibition of the prostate tumors. In this experiment, the growth inhibition of the CWR22-prostate cancer is comparable to the effects of castration and superior to the comparison substance bicalutamide (Diagram 4).

[0070] In this invention, the action of the compounds according to the invention on tumor growth *in vivo* was studied by means of two different mouse-xenograft models, in which the compounds according to the invention were orally administered 1x daily over the entire treatment period. In comparison to the untreated control animals, an inhibition of the tumor growth resulted. Retardation of the tumor growth was shown as significant in castrated mice. The treatment was well tolerated.

[0071] In both models (CWR22; LNCaP), the inhibition of the tumor growth by the compounds according to the invention is superior to the treatment with the anti-androgen bicalutamide.

[0072] Model 4: Pharmacokinetics in Rats After Intravenous and Peroral Administration

[0073] The pharmacokinetic properties of the compounds according to the invention were studied in Examples 1: 4-[4,4-dimethyl-5-oxo-3-[8-(pyrrolidin-1-yl)octyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile and 11: 4-[3-[8-[(2S)-2-(hydroxymethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile.

TABLE 3

Pharmacokinetic Properties of Selected Examples		
Determined Parameters	Example 1	Example 11
Distribution Volume V_{dss} [l/kg]	37	11
Systemic Serum Clearance CL [ml/min/kg]	76	20
Half-Life <i>i.v.</i> $T_{1/2}$ [h]	5.6	5.7
Half-Life <i>p.o.</i> $T_{1/2}$ [h]	6.1	8.0
Absorption T_{max} [h]	5	7
Oral Bioavailability [%]	72	65

[0074] The determined pharmacokinetic data show very advantageous pharmacokinetic properties of the compounds according to the invention in rats, which result in high oral bioavailability and a long half-life (>5 h). The data indicate a high intestinal absorption and a relatively low liver-first-pass-effect (increased metabolic stability).

[0075] Model 5: Destabilization of AR in LNCaP Cells by Test Substances

[0076] In a 25 cm² cell culture flask, 2x10⁶ LNCaP cells in 6 ml of RPMI 1640 without phenol red are grown with 4

mmol of glutamine and 5% activated carbon-treated serum (CCS) and cultivated overnight at 37° C., 5% CO₂ in a humid atmosphere. On the next day, the cells are treated with the test substance at a concentration of 10 or 1 μmol, whereby the final concentration of the solvent is 0.5% DMSO. As a control, cells are treated only with 0.5% DMSO. After an incubation time of 24 hours, the medium is changed with renewed administration of substance, and another 24 hours of incubation. After 48 hours, the cells are washed with PBS, dissolved with PBS/20 mmol of EDTA, washed again with PBS-Ca²⁺/Mg²⁺ and then frozen for at least 2 hours as a cell pellet at -80° C. Then, the cell pellet is resuspended in 200 μl of lysis buffer (50 mmol of tris/HCl, pH 7.5; 150 mmol of NaCl, 1.5 mmol of MgCl₂, 0.2% SDS, 10% glycerol, 1 mmol of DTT, 0.01x complete-EDTA protease inhibitors (Roche, Mannheim)) and treated with 10 U benzonase (Merck, Darmstadt) for 10 minutes at 4° C. After 5 mmol of EDTA is added, insoluble material is pelletized and 25 μg of the cell extract is separated in a 4-12% SDS-polyacrylamide gel (invitrogen). Then, the proteins are transferred to nitrocellulose (HyBondECL, Amersham) and incubated with monoclonal antibodies against the androgen receptor (AR441; Santa Cruz Biotechnologies; 1:400 dilution) and actin (ICN, 1:5000-1:20000 dilution). After incubation with the secondary antibody (anti mouse IgG-HRP, Amersham or -AP, invitrogen), the Western Blot is developed by means of chemiluminescence (ECL, Amersham; Western Breeze, invitrogen), and the light signals are quantified with a Chemilmager™ (Kodak). The amount of androgen receptor is calculated in a ratio to actin as a percentage of the DMSO control.

[0077] Table 4 shows the action of selected test substances at concentrations of 10 or 1 μmol on the content of androgen receptor protein in the human prostate cell line LNCaP. The data correspond to the proportion, in percent, of the AR content of cells that were treated only with the solvent DMSO (=control). The treatment of the cells with the cited test substances results, as in Example 56 (4-[3-[7-[(2S)-4,4-dimethyl-5-oxo-2-(pyrrolidin-1-ylmethyl)pyrrolidin-1-yl]heptyl]-2-thioxoimidazolidin-1-yl]-2-trifluoromethyl)benzotrile) at a treatment concentration of 1 μmol, in a reduction of the AR content to up to one-fourth of the control (24%). The comparison substance bicalutamide does not influence the AR content, while the synthetic androgen R1881 stabilizes the AR protein. The latter is known from the literature (J. A. Kempainen et. al. J. Biol. Chem. 1992, 267, 968-974).

[0078] By the reduction of the AR content, which presumably is carried out by a destabilization of the AR protein, the inhibitory action of the antihormones on cell proliferation is to be enhanced.

TABLE 4

AR Content (%) in LNCaP Cells After Treatment with Selected Test Substances			
Example	Test Substance	AR Content [%]	
		at 10 μM	at 1 μM
1	4-[4,4-Dimethyl-5-oxo-3-[8-(pyrrolidin-1-yl)octyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	63	

TABLE 4-continued

Example	Test Substance	AR Content [%]	
		at 10 μ M	at 1 μ M
2	4-[4,4-Dimethyl-5-oxo-3-[8-(pyrrolidin-1-yl)octyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzotrile hydrochloride	50	
9	4-[3-[7-[(2S)-2-(Hydroxymethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile		63
11	4-[3-[8-[(2S)-2-(Hydroxymethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile	50	
37	4-[3-[8-[(2R,5S)-rel-2,5-Dimethylpyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile		57
40	4-[3-[8-[(2S)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile		40
52	(R)-1-[6-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]hexyl]pyrrolidine-4-carboxamide	36	74
56	4-[3-[7-[(2S)-4,4-Dimethyl-5-oxo-2-(pyrrolidin-1-ylmethyl)pyrrolidin-1-yl]heptyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile		24
Comparison US Re. 35956 Example 71	4-[3-(2-Hydroxyethyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzotrile	191	164
Comparison US Re. 35956 Example 77	4-[3-(4-Hydroxybutyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzotrile	180	230
Bicalutamide R1881 (10 nM)	N-[4-Cyano-3-(trifluoromethyl)phenyl]-3-[(4-fluorophenyl)sulfonyl]-2-hydroxy-2-methylpropanamide	100	
	17 β -Hydroxy-17 α -methylene-4,9,11-trien-3-one	250	

[0079] The compounds according to the invention are suitable for an extended effective treatment or prophylaxis of androgen-dependent diseases of the human or animal body. The compounds according to the invention are suitable especially for the treatment or prophylaxis of androgen-dependent proliferative diseases, in particular prostate cancers and benign prostate hyperplasia (BHP).

[0080] This invention comprises pharmaceutical preparations that contain one or more compounds of general formula I or their pharmaceutically compatible salts, optionally together with pharmaceutically compatible adjuvants and/or vehicles, as well as the use of compounds of general formula I for the production of a pharmaceutical agent for treatment or prophylaxis of diseases of the human or animal body, which can be influenced by the inhibition of the androgen receptor.

[0081] In this sense, the compounds of general formula I according to the invention as well as pharmaceutical preparations that contain the latter can also be used for prophylaxis and/or therapy of other androgen-dependent images of disease or symptoms, which have a non-proliferative nature (e.g., androgenetic alopecia, hirsutism or androgen-dependent acne).

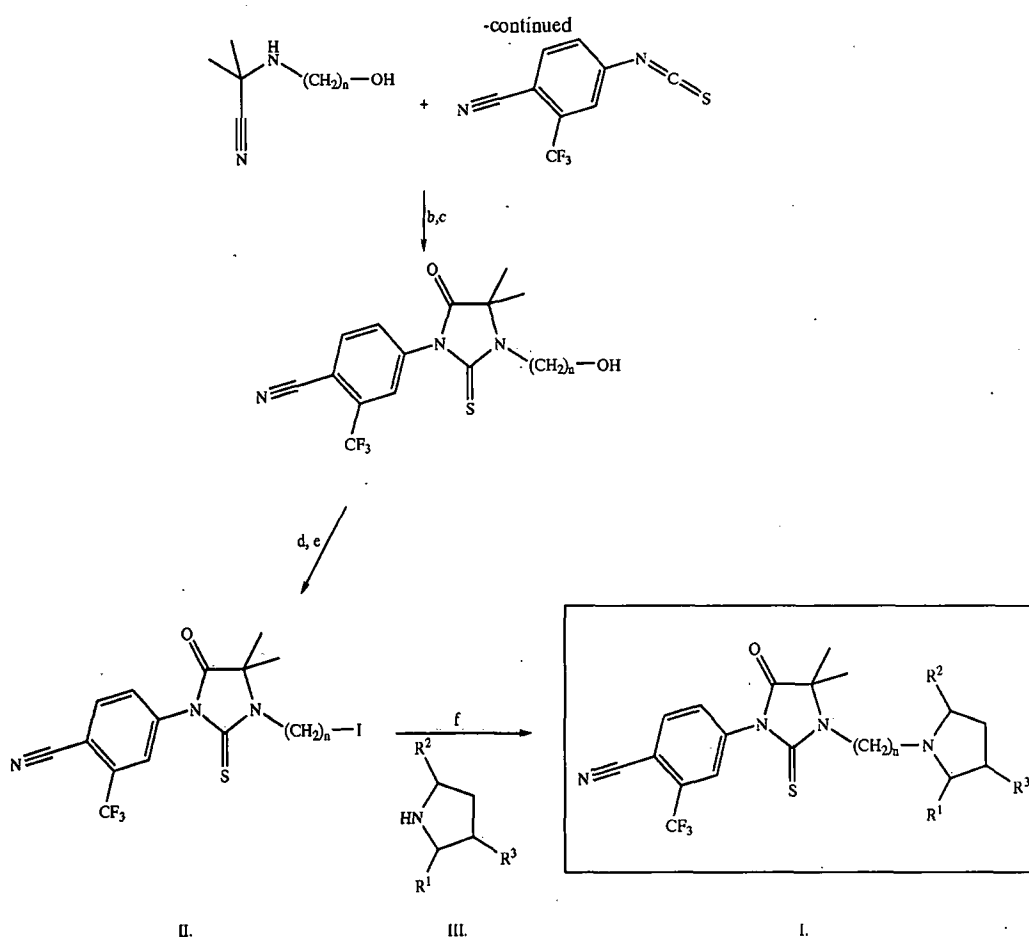
[0082] Dosage

[0083] In general, satisfactory results can be expected when the daily doses comprise a range of 5 μ g to 50 mg of the compound according to the invention per kg of body

weight. In the case of larger mammals, for example humans, a recommended daily dose is in the range of 10 μ g to 30 mg per kg of body weight. Suitable dosages for the compounds according to the invention are from 0.005 to 50 mg per day per kg of body weight, depending on age and constitution of the patient, whereby the necessary daily dose can be administered one or more times.

[0084] The formulation of the pharmaceutical preparations based on the new compounds is carried out in a way that is known in the art by the active ingredient being administered with the vehicles, fillers, substances that influence decomposition, binding agents, moisturizers, lubricants, absorbents, diluents, flavoring correctives, coloring agents, etc., that are commonly used in galenicals and being converted into the desired form of administration. In this case, reference is made to Remington's Pharmaceutical Science, 15th ed. Mack Publishing Company, East Pennsylvania (1980).

[0085] For oral administration, in particular tablets, coated tablets, capsules, pills, powders, granulates, pastilles, suspensions, emulsions or solutions are suitable. For parenteral administration, injection and infusion preparations are possible. For intraarticular injection, correspondingly prepared crystal suspensions can be used. For the intramuscular injection, aqueous and oily injection solutions or suspensions, and correspondingly depot preparations can be used. For rectal administration, new compounds in the form of suppositories, capsules, solutions (e.g., in the form of



[0109] Reagents: (a) CSCl_2 , DMF; (b) N^iEt_3 , THF; (c) 4M HCl; (d) RSO_2Cl , NEt_3 , CH_2Cl_2 ; (e) NaI, acetone; (f) NEt_3 , THF.

[0111] $^1\text{H-NMR}$ (300 MHz, CDCl_3): δ [ppm]=3.64 t (J=6.8 Hz, 2H, CH_2OH); 3.41 t (J=6.8 Hz, 2H, CH_2Br); 1.85 t (J=7.3 Hz/6.8 Hz, 2H, CH_2); 1.56 m (2H, CH_2); 149-1.27 m (8H, CH_2).

Production Processes

EXAMPLE 1

4-[4,4-Dimethyl-5-oxo-3-[8-(pyrrolidin-1-yl)octyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzothionitrile

1 a) 8-Bromooctan-1-ol

[0110] 25 g of octane-1,8-diol was boiled in 250 ml of cyclohexane with 22.6 ml of 47% aqueous hydrobromic acid for six hours in a water separator. The reaction mixture was then poured onto saturated aqueous sodium bicarbonate solution and extracted with ethyl acetate. The organic phase was washed with saturated, aqueous sodium chloride solution, dried on sodium sulfate, filtered and concentrated by evaporation in a vacuum. Column chromatography on silica gel with a mixture that consists of hexane/ethyl acetate yielded 21.6 g of the title compound as a yellowish oil.

1b) 2-(8-Hydroxyoctyl)-1H-isindolc-1,3(2H)-dione

[0112] A solution of 15.12 g of phthalic acid imide in 480 ml of N,N-dimethylformamide was mixed in portions at room temperature with 5.04 g of 50% sodium hydride as a dispersion in mineral oil. The reaction mixture was stirred for one hour at room temperature. Then, a solution of 20 g of the compound, produced under 1a), in 480 ml of N,N-dimethylformamide was added in drops, and the reaction mixture was stirred for three hours at room temperature. The mixture was then poured onto saturated aqueous sodium bicarbonate solution and extracted with ethyl acetate. The organic phase was washed with saturated aqueous sodium chloride solution, dried on sodium sulfate, filtered and concentrated by evaporation in a vacuum. Column chromatography on silica gel with a mixture that consists of hexane/ethyl acetate yielded 23.0 g of the title compound as a colorless foam.

[0113] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=7.84 m (2H, aryl); 7.71 m (2H, aryl); 3.68 t (J=7 Hz, 2H, CH₂N); 3.62 t (J=6 Hz, 2H, CH₂OH); 1.67 m (2H, CH₂); 1.56 m (2H, CH₂); 1.32 m (8H, CH₂).

1c) 8-Aminooctan-1-ol

[0114] 15.2 ml of 80% aqueous hydrazinium hydroxide was added in drops to a solution of 23 g of the compound, produced under 1b), in 400 ml of ethanol. The reaction mixture was boiled for four hours. The white precipitate was filtered off and rewashed with ethanol. The filtrate was concentrated by evaporation in a vacuum. The residue was taken up in ethyl acetate and irradiated for 30 minutes in an ultrasound bath. The white precipitate was in turn filtered off and rewashed with ethyl acetate. The filtrate was concentrated by evaporation in a vacuum. 8.88 g of the title compound was obtained.

[0115] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=3.62 t (J=7 Hz, 2H, CH₂OH); 2.68 t (J=6 Hz, 2H, CH₂NH₂); 1.56 m (2H, CH₂); 1.32 m (10H, CH₂).

1d) 4-[4,4-Dimethyl-5-oxo-3-(8-hydroxyoctyl)-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

[0116] While being cooled in an ice bath and under nitrogen atmosphere, 2.3 ml of thiophosgene was added in drops to a solution of 4.99 g of 4-amino-2-(trifluoromethyl)benzotrile in 30 ml of N,N-dimethylformamide. The reaction mixture was stirred for one hour at room temperature and then mixed with water. The aqueous phase was extracted with ethyl acetate. The combined organic phases were washed with saturated aqueous sodium chloride solution, dried on sodium sulfate, filtered and concentrated by evaporation in a vacuum. The thus obtained crude isothiocyanate was combined with the cyanamine produced by two hours of stirring of 5.4 ml of acetone cyanohydrin with 4.29 g of the compound produced under 1c) in the presence of 3 g of molecular sieve 3 Å at room temperature and boiled for one hour with 7.47 ml of triethylamine in 134 ml of tetrahydrofuran. The crude iminothiohydantoin obtained after concentration by evaporation in a vacuum was stirred with 26.8 ml of 4 molar aqueous hydrochloric acid in 134 ml of methanol overnight at room temperature. The reaction mixture was then poured onto saturated aqueous sodium bicarbonate solution and extracted with ethyl acetate. The organic phase was washed with saturated aqueous sodium chloride solution, dried on sodium sulfate, filtered and concentrated by evaporation in a vacuum. Column chromatography on silica gel with a mixture that consists of hexane/ethyl acetate yielded 8.1 g of the title compound as a colorless foam.

[0117] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=7.94 d (J=8 Hz, 1H, aryl); 7.89 d (J=2.1 Hz, 1H, aryl); 7.77 dd (J=8 Hz/2.1 Hz, 1H, aryl); 3.67 m (2H, CH₂N); 3.65 t (J=6.5 Hz, 2H, CH₂OH); 1.83 m (2H, CH₂); 1.55 m (2H, CH₂); 1.58 s (6H, CH₃); 1.37 m (8H, CH₂).

1e) 8-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]octyl 4-methylbenzenesulfonate

[0118] 8.1 g of the compound produced under 1d) was stirred for one hour at room temperature with 21.0 g of

p-toluenesulfonic acid chloride and 25.5 ml of triethylamine in 92 ml of dichloromethane. The reaction mixture was poured into saturated, aqueous sodium bicarbonate solution and extracted with dichloromethane. The organic phase was washed with saturated, aqueous sodium chloride solution, dried on sodium sulfate, filtered and concentrated by evaporation in a vacuum. Column chromatography on silica gel with a mixture that consists of hexane/ethyl acetate yielded 9.21 g of the title compound as a colorless foam.

[0119] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=7.94 d (J=8 Hz, 1H, aryl); 7.89 d (J=2.1 Hz, 1H, aryl); 7.78 d (J=8.2 Hz, 2H, aryl); 7.77 dd (J=8 Hz/2.1 Hz, 1H, aryl); 7.34 d (J=8.2 Hz, 2H, aryl); 4.03 t (J=6.5 Hz, 2H, CH₂O); 3.66 m (2H, CH₂N); 2.45 s (3H, CH₃); 1.81 m (2H, CH₂); 1.65 m (2H, CH₂); 1.58 s (6H, CH₃); 1.33 m (8H, CH₂).

1f) 4-[4,4-Dimethyl-5-oxo-3-(8-iodooctyl)-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

[0120] 9.21 g of the compound produced under 1e) was boiled for one hour with 9.3 g of sodium iodide in 150 ml of acetone. The reaction mixture was filtered at room temperature and concentrated by evaporation in a vacuum. Column chromatography on silica gel with a mixture that consists of hexane/ethyl acetate yielded 8.55 g of the title compound as a yellowish foam.

[0121] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=7.95 d (J=8 Hz, 1H, aryl); 7.89 d (J=2.1 Hz, 1H, aryl); 7.77 dd (J=8 Hz/2.1 Hz, 1H, aryl); 3.67 m (2H, CH₂N); 3.20 t (J=7 Hz, 2H, CH₂I); 1.82 m (2H, CH₂); 1.80 m (2H, CH₂); 1.58 s (6H, CH₃); 1.37 m (8H, CH₂).

1g) 4-[4,4-Dimethyl-5-oxo-3-[8-(pyrrolidin-1-yl)octyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

[0122] 200 mg of the compound produced under 1f) was boiled for four hours with 60 μl of pyrrolidine and 101 μl of triethylamine in 5 ml of tetrahydrofuran. The reaction mixture was concentrated by evaporation in a vacuum. Column chromatography on silica gel with a mixture that consists of hexane/ethyl acetate yielded 130 mg of the title compound as a colorless oil.

[0123] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=7.94 d (J=8 Hz, 1H, aryl); 7.89 d (J=2.1 Hz, 1H, aryl); 7.77 dd (J=8 Hz/2.1 Hz, 1H, aryl); 3.67 m (2H, CH₂N); 2.64 m (4H, CH₂N); 2.53 m (2H, CH₂N); 1.85 m (4H, CH₂); 1.82 m (2H, CH₂); 1.59 m (2H, CH₂); 1.58 s (6H, CH₃); 1.36 m (8H, CH₂).

EXAMPLE 2

4-[4,4-Dimethyl-5-oxo-3-[8-(pyrrolidin-1-yl)octyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile hydrochloride

[0124] 20 mg of the compound, produced under 1g), was dissolved in 1 ml of tetrahydrofuran and stirred for one hour at room temperature with 67 μl of a 1.2 molar solution of hydrochloric acid in diethyl ether. The reaction mixture was concentrated by evaporation in a vacuum. 21 mg of the title compound was obtained.

[0125] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=7.95 d (J=8 Hz, 1H, aryl); 7.89 d (J=2.1 Hz, 1H, aryl); 7.78 dd (J=8

Hz/2.1 Hz, 1H, aryl); 3.79 m (2H, CH₂N); 3.67 m (2H, CH₂N); 3.00 m (2H, CH₂N); 2.75 m (2H, CH₂N); 2.23 m (2H, CH₂); 2.06 m (2H, CH₂); 1.90 m (2H, CH₂); 1.82 m (2H, CH₂); 1.59 s (6H, CH₃); 1.38 m (8H, CH₂).

EXAMPLE 3

4-14,4-Dimethyl-5-oxo-3-[6-(pyrrolidin-1-yl)hexyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzoxonitrile

3a) 4-[3-(6-Iodoheptyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzoxonitrile

[0126] The production of the title compound was carried out analogously to the process described in Examples 1d) to 1f). 6-Aminohexan-1-ol was used as a chain component.

[0127] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=7.95 d (J=8 Hz, 1H, aryl); 7.89 d (J=2.1 Hz, 1H, aryl); 7.77 dd (J=8 Hz/2.1 Hz, 1H, aryl); 3.68 m (2H, CH₂N); 3.21 t (J=6.8 Hz, 2H, CH₂I); 1.86 m (2H, CH₂); 1.83 m (2H, CH₂); 1.59 s (6H, CH₃); 1.46 m (4H, CH₂).

3b) 4-[4,4-Dimethyl-5-oxo-3-[6-(pyrrolidin-1-yl)hexyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzoxonitrile

[0128] 20 mg of the compound, produced under 3a), was boiled for four hours with 6.3 μl of pyrrolidine and 10.6 μl of triethylamine in 1 ml of tetrahydrofuran. The reaction mixture was concentrated by evaporation in a vacuum. Column chromatography on silica gel with a mixture that consists of hexane/ethyl acetate yielded 15 mg of the title compound as a colorless oil.

[0129] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=7.95 d (J=8 Hz, 1H, aryl); 7.89 d (J=2.1 Hz, 1H, aryl); 7.77 dd (J=8 Hz/2.1 Hz, 1H, aryl); 3.68 m (2H, CH₂N); 2.75 (4H, CH₂N); 2.64 m (2H, CH₂N); 1.90 m (4H, CH₂); 1.84 m (2H, CH₂); 1.68 m (2H, CH₂); 1.59 s (6H, CH₃); 1.43 m (4H, CH₂).

EXAMPLE 4

4-[4,4-Dimethyl-5-oxo-3-[7-(pyrrolidin-1-yl)heptyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzoxonitrile

4a) 4-[3-(7-Iodoheptyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzoxonitrile

[0130] The production of the title compound was carried out analogously to the process described in Examples 1a) to 1f). As a chain component, heptane-1,7-diol was used.

[0131] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=7.95 d (J=8 Hz, 1H, aryl); 7.90 d (J=2.1 Hz, 1H, aryl); 7.78 dd (J=8 Hz/2.1 Hz, 1H, aryl); 3.67 m (2H, CH₂N); 3.20 t (J=7 Hz, 2H, CH₂I); 1.84 m (4H, CH₂); 1.59 s (6H, CH₃); 1.41 m (6H, CH₂).

4b) 4-[4,4-Dimethyl-5-oxo-3-[7-(pyrrolidin-1-yl)heptyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzoxonitrile

[0132] 13 mg of the compound produced under 4a) was boiled for four hours with 4 μl of pyrrolidine and 6.7 μl of triethylamine in 1 ml of tetrahydrofuran. The reaction mixture was concentrated by evaporation in a vacuum. Column chromatography on silica gel with a mixture that consists of hexane/ethyl acetate yielded 7 mg of the title compound as a colorless oil.

[0133] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=7.95 d (J=8 Hz, 1H, aryl); 7.89 d (J=2.1 Hz, 1H, aryl); 7.78 dd (J=8 Hz/2.1 Hz, 1H, aryl); 3.67 m (2H, CH₂N); 2.72 m (4H, CH₂N); 2.60 m (2H, CH₂N); 1.89 m (4H, CH₂); 1.83 m (2H, CH₂); 1.65 m (2H, CH₂); 1.58 s (6H, CH₃); 1.40 m (6H, CH₂).

EXAMPLE 5

4-[4,4-Dimethyl-5-oxo-3-[9-(pyrrolidin-1-yl)nonyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzoxonitrile

5a) 4-[3-(9-Iodononyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzoxonitrile

[0134] The production of the title compound was carried out analogously to the process described in Examples 1a) to 1f). Nonane-1,9-diol was used as a chain component.

[0135] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=7.95 d (J=8 Hz, 1H, aryl); 7.89 d (J=2.1 Hz, 1H, aryl); 7.77 dd (J=8 Hz/2.1 Hz, 1H, aryl); 3.67 m (2H, CH₂N); 3.19 t (J=7 Hz, 2H, CH₂I); 1.84 m (2H, CH₂); 1.82 m (2H, CH₂); 1.58 s (6H, CH₃); 1.36 m (10H, CH₂).

5b) 4-[4,4-Dimethyl-5-oxo-3-[9-(pyrrolidin-1-yl)nonyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzoxonitrile

[0136] 20 mg of the compound produced under 5a) was boiled for four hours with 5.9 μl of pyrrolidine and 9.9 μl of triethylamine in 1 ml of tetrahydrofuran. The reaction mixture was concentrated by evaporation in a vacuum. Column chromatography on silica gel with a mixture that consists of hexane/ethyl acetate yielded 10 mg of the title compound as a colorless oil.

[0137] ¹H-NMR (300 MHz, CDCl₃): δ[ppm]=7.95 d (J=8 Hz, 1H, aryl); 7.89 d (J=2.1 Hz, 1H, aryl); 7.77 dd (J=8 Hz/2.1 Hz, 1H, aryl); 3.66 m (2H, CH₂N); 2.47 m (4H, CH₂N); 2.40 m (2H, CH₂N); 1.77 m (4H, CH₂); 1.82 m (2H, CH₂); 1.58 s (6H, CH₃); 1.51 m (2H, CH₂); 1.31 m (10H, CH₂).

[0138] Analogously to the production instructions, described in detail, in Examples 1 to 5, the following compounds were obtained:

Example	Product/ Reagent	Instructions analogous to δ [ppm]	¹ H-NMR(300 MHz, CDCl ₃)
6	4-[3-[6-((2R)-2-(Hydroxymethyl)-	3	7.95d(J=8Hz, 1H, aryl); 7.90d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz,

-continued

Example	Product/ Reagent	Instructions analogous to	¹ H-NMR(300 MHz, CDCl ₃) δ [ppm]
	pyrrolidin-1-yl]hexyl]- 4,4-dimethyl-5-oxo-2- thioxoimidazolidin-1- yl]-2-(trifluoromethyl)- benzonitrile (2R)-Pyrrolidine- methanol		1H, aryl); 3.67m(2H, CH ₂ N); 3.62dd (J=10.5Hz/3.8Hz, 1H, CH ₂ OH); 3.39 dd(J=10.5Hz/2.1Hz, 1H, CH ₂ OH); 3.19m(1H, CH ₂ N); 2.73ddbr(J=11.8Hz/ 7.6Hz/7.6Hz, 1H, CH ₂ N); 2.58 m(1H, CH ₂ N); 2.25m(2H, CH ₂ N); 1.96-1.67m(4H, CH ₂); 1.84m(2H, CH ₂); 1.58s(6H, CH ₃); 1.54m(2H, CH ₂); 1.41m(4H, CH ₂)
7	4-[3-[6-[(2S)-2- (Hydroxymethyl)- pyrrolidin-1-yl]hexyl]- 4,4-dimethyl-5-oxo-2- thioxoimidazolidin-1- yl]-2-(trifluoromethyl)- benzonitrile (2S)-Pyrrolidinemethanol	3	7.95d(J=8Hz, 1H, aryl); 7.90d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.67m(2H, CH ₂ N); 3.62dd (J=10.5Hz/3.8Hz, 1H, CH ₂ OH); 3.39 dd(J=10.5Hz/2.1Hz, 1H, CH ₂ OH); 3.19m(1H, CH ₂ N); 2.73ddbr(J=11.8Hz/ 7.6Hz/7.6Hz, 1H, CH ₂ N); 2.58 m(1H, CH ₂ N); 2.25m(2H, CH ₂ N); 1.96-1.67m(4H, CH ₂); 1.84m(2H, CH ₂); 1.58s(6H, CH ₃); 1.54m(2H, CH ₂); 1.41m(4H, CH ₂)
8	4-[3-[7-[(2R)-2- (Hydroxymethyl)- pyrrolidin-1-yl]heptyl]- 4,4-dimethyl-5-oxo-2- thioxoimidazolidin-1- yl]-2-(trifluoromethyl)- benzonitrile (2R)-Pyrrolidinemethanol	4	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.67m(2H, CH ₂ N); 3.62dd (J=10.5Hz/3.8Hz, 1H, CH ₂ OH); 3.39 dd(J=10.5Hz/2.5Hz, 1H, CH ₂ OH); 3.18m(1H, CH ₂ N); 2.71ddbr(J=11.8Hz/ 8Hz/8Hz, 1H, CH ₂ N); 2.57m (1H, CH ₂ N); 2.25m(1H, CH ₂ N); 2.24 m(1H, CH ₂ N); 1.95-1.66m(4H, CH ₂); 1.80m(2H, CH ₂); 1.58s(6H, CH ₃); 1.51m(2H, CH ₂); 1.39m(6H, CH ₂)
9	4-[3-[7-[(2S)-2- (Hydroxymethyl)- pyrrolidin-1-yl]heptyl]- 4,4-dimethyl-5-oxo-2- thioxoimidazolidin-1- yl]-2-(trifluoromethyl)- benzonitrile (2S)-Pyrrolidinemethanol	4	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.67m(2H, CH ₂ N); 3.62dd (J=10.5Hz/3.8Hz, 1H, CH ₂ OH); 3.39 dd(J=10.5Hz/2.5Hz, 1H, CH ₂ OH); 3.18m(1H, CH ₂ N); 2.71ddbr(J=11.8Hz/ 8Hz/8Hz, 1H, CH ₂ N); 2.57m (1H, CH ₂ N); 2.25m(1H, CH ₂ N); 2.24 m(1H, CH ₂ N); 1.95-1.66m(4H, CH ₂); 1.80m(2H, CH ₂); 1.58s(6H, CH ₃); 1.51m(2H, CH ₂); 1.39m(6H, CH ₂)
10	4-[3-[8-[(2R)-2- (Hydroxymethyl)- pyrrolidin-1-yl]octyl]- 4,4-dimethyl-5-oxo-2- thioxoimidazolidin-1- yl]-2-(trifluoromethyl)- benzonitrile (2R)-Pyrrolidinemethanol	1	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.78dd(J=8Hz/2.1Hz, 1H, aryl); 3.67m(2H, CH ₂ N); 3.63dd (J=10.5Hz/3.8Hz, 1H, CH ₂ OH); 3.39 dd(J=10.5Hz/2.5Hz, 1H, CH ₂ OH); 3.18m(1H, CH ₂ N); 2.71ddbr(J=11.8Hz/ 8Hz/8Hz, 1H, CH ₂ N); 2.58m (1H, CH ₂ N); 2.25m(2H, CH ₂ N); 1.96-1.67 m(4H, CH ₂); 1.82m(2H, CH ₂); 1.58s(6H, CH ₃); 1.50m(2H, CH ₂); 1.36m(8H, CH ₂)
11	4-[3-[8-[(2S)-2- (Hydroxymethyl)- pyrrolidin-1-yl]octyl]- 4,4-dimethyl-5-oxo-2- thioxoimidazolidin-1- yl]-2-(trifluoromethyl)- benzonitrile (2S)-Pyrrolidinemethanol	1	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.78dd(J=8Hz/2.1Hz, 1H, aryl); 3.67m(2H, CH ₂ N); 3.63dd (J=10.5Hz/3.8Hz, 1H, CH ₂ OH); 3.39 dd(J=10.5Hz/2.5Hz, 1H, CH ₂ OH); 3.18m(1H, CH ₂ N); 2.71ddbr(J=11.8Hz/ 8Hz/8Hz, 1H, CH ₂ N); 2.58m (1H, CH ₂ N); 2.25m(2H, CH ₂ N); 1.96-1.67 m(4H, CH ₂); 1.82m(2H, CH ₂); 1.58s(6H, CH ₃); 1.50m(2H, CH ₂); 1.36 m(8H, CH ₂)
12	4-[3-[9-[(2R)-2- (Hydroxymethyl)- pyrrolidin-1-yl]nonyl]- 4,4-dimethyl-5-oxo-2- thioxoimidazolidin-1- yl]-2-(trifluoromethyl)- benzonitrile (2R)-Pyrrolidinemethanol	5	7.95d(J=8Hz, 1H, aryl); 7.90d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.67m(2H, CH ₂ N); 3.62dd (J=10.5Hz/3.8Hz, 1H, CH ₂ OH); 3.38 dd(J=10.5Hz/2.5Hz, 1H, CH ₂ OH); 3.17m(1H, CH ₂ N); 2.70ddbr(J=11.8Hz/ 8.4Hz/8Hz, 1H, CH ₂ N); 2.56m (1H, CH ₂ N); 2.24m(1H, CH ₂ N); 2.21 m(1H, CH ₂ N); 1.92-1.67m(4H, CH ₂);

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Example	Product/ Reagent	Instructions analogous to	¹ H-NMR(300 MHz, CDCl ₃) δ [ppm]
13	4-[3-[9-[(2S)-2-(Hydroxymethyl)-pyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzoxirone (2S)-Pyrrolidinemethanol	5	1.82m(2H, CH ₂); 1.58s(6H, CH ₃); 1.48m(2H, CH ₂); 1.31m(10H, CH ₂); 7.95d(J=8Hz, 1H, aryl); 7.90d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.67m(2H, CH ₂ N); 3.62dd(J=10.5Hz/3.8Hz, 1H, CH ₂ OH); 3.38dd(J=10.5Hz/2.5Hz, 1H, CH ₂ OH); 3.17m(1H, CH ₂ N); 2.70ddbr(J=11.8Hz/8.4Hz/8Hz, 1H, CH ₂ N); 2.56m(1H, CH ₂ N); 2.24m(1H, CH ₂ N); 2.21m(1H, CH ₂ N); 1.92-1.67m(4H, CH ₂); 1.82m(2H, CH ₂); 1.58s(6H, CH ₃); 1.48m(2H, CH ₂); 1.31m(10H, CH ₂)
14	4-[3-[6-[(2R)-2-(Methoxymethyl)-pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzoxirone (2R)-2-(Methoxymethyl)pyrrolidine	3	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.67m(2H, CH ₂ N); 3.42dd(J=9.3Hz/5.1Hz, 1H, CH ₂ O); 3.35s(3H, CH ₃ O); 3.28dd(J=9.3Hz/5.9Hz, 1H, CH ₂ O); 3.16m(1H, CH ₂ N); 2.85dddbr(J=11.8Hz/8.4Hz/8Hz, 1H, CH ₂ N); 2.56m(1H, CH ₂ N); 2.26dddbr(J=11.8Hz/7.6Hz/7.6Hz, 1H, CH ₂ N); 2.17m(1H, CH ₂ N); 1.92-1.67m(4H, CH ₂); 1.83m(2H, CH ₂); 1.57s(6H, CH ₃); 1.54m(2H, CH ₂); 1.40m(4H, CH ₂)
15	4-[3-[6-[(2S)-2-(Methoxymethyl)-pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzoxirone (2S)-2-(Methoxymethyl)pyrrolidine	3	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.67m(2H, CH ₂ N); 3.42dd(J=9.3Hz/5.1Hz, 1H, CH ₂ O); 3.35s(3H, CH ₃ O); 3.28dd(J=9.3Hz/5.9Hz, 1H, CH ₂ O); 3.16m(1H, CH ₂ N); 2.85dddbr(J=11.8Hz/8.4Hz/8Hz, 1H, CH ₂ N); 2.56m(1H, CH ₂ N); 2.26dddbr(J=11.8Hz/7.6Hz/7.6Hz, 1H, CH ₂ N); 2.17m(1H, CH ₂ N); 1.92-1.67m(4H, CH ₂); 1.83m(2H, CH ₂); 1.57s(6H, CH ₃); 1.54m(2H, CH ₂); 1.40m(4H, CH ₂)
16	4-[3-[7-[(2R)-2-(Methoxymethyl)-pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzoxirone (2R)-2-(Methoxymethyl)pyrrolidine	4	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.66m(2H, CH ₂ N); 3.42dd(J=9.3Hz/5.1Hz, 1H, CH ₂ O); 3.35s(3H, CH ₃ O); 3.27dd(J=9.3Hz/5.9Hz, 1H, CH ₂ O); 3.14m(1H, CH ₂ N); 2.83dddbr(J=11.8Hz/8.4Hz/8Hz, 1H, CH ₂ N); 2.55m(1H, CH ₂ N); 2.24dddbr(J=11.8Hz/7.6Hz/7.6Hz, 1H, CH ₂ N); 2.16m(1H, CH ₂ N); 1.92-1.67m(4H, CH ₂); 1.83m(2H, CH ₂); 1.58s(6H, CH ₃); 1.51m(2H, CH ₂); 1.38m(6H, CH ₂)
17	4-[3-[7-[(2S)-2-(Methoxymethyl)-pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzoxirone (2S)-2-(Methoxymethyl)pyrrolidine	4	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.66m(2H, CH ₂ N); 3.42dd(J=9.3Hz/5.1Hz, 1H, CH ₂ O); 3.35s(3H, CH ₃ O); 3.27dd(J=9.3Hz/5.9Hz, 1H, CH ₂ O); 3.14m(1H, CH ₂ N); 2.83dddbr(J=11.8Hz/8.4Hz/8Hz, 1H, CH ₂ N); 2.55m(1H, CH ₂ N); 2.24dddbr(J=11.8Hz/7.6Hz/7.6Hz, 1H, CH ₂ N); 2.16m(1H, CH ₂ N); 1.92-1.67m(4H, CH ₂); 1.83m(2H, CH ₂); 1.58s(6H, CH ₃); 1.51m(2H, CH ₂); 1.38m(6H, CH ₂)
18	4-[3-[8-[(2R)-2-(Methoxymethyl)-pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzoxirone	1	7.94d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.66m(2H, CH ₂ N); 3.43dd(J=9.3Hz/5.1Hz, 1H, CH ₂ O); 3.35s(3H, CH ₃ O); 3.27dd(J=9.3Hz/5.9Hz, 1H, CH ₂ O); 3.15m(1H, CH ₂ N); 2.82dddbr(J=11.8Hz/8.4Hz/8Hz, 1H,

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Example	Product/ Reagent	Instructions analogous to	¹ H-NMR(300 MHz, CDCl ₃) δ [ppm]
	(2R)-2-(Methoxymethyl) pyrrolidine		CH ₂ N; 2.56m(1H, CH ₂ N); 2.24dddb (J=11.8Hz/7.6Hz/7.6Hz, 1H, CH ₂ N); 2.17m(1H, CH ₂ N); 1.92-1.67 m(4H, CH ₂); 1.82m(2H, CH ₂); 1.58s (6H, CH ₃); 1.51m(2H, CH ₂); 1.36m (8H, CH ₂)
19	4-[3-[8-[(2S)-2- (Methoxymethyl)- pyrrolidin-1-yl]octyl]- 4,4-dimethyl-5-oxo-2- thioxoimidazolidin-1- yl]-2-(trifluoromethyl)- benzotrile (2S)-2-(Methoxymethyl) pyrrolidine	1	7.94d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.66m(2H, CH ₂ N); 3.43dd (J=9.3Hz/5.1Hz, 1H, CH ₂ O); 3.35s (3H, CH ₃ O); 3.27dd(J=9.3Hz/5.9Hz, 1H, CH ₂ O); 3.15m(1H, CH ₂ N); 2.82 dddb(J=11.8Hz/8.4Hz/8Hz, 1H, CH ₂ N); 2.56m(1H, CH ₂ N); 2.24dddb (J=11.8Hz/7.6Hz/7.6Hz, 1H, CH ₂ N); 2.17m(1H, CH ₂ N); 1.92-1.67 m(4H, CH ₂); 1.82m(2H, CH ₂); 1.58s (6H, CH ₃); 1.51m(2H, CH ₂); 1.36m (8H, CH ₂)
20	4-[3-[9-[(2R)-2- (Methoxymethyl)- pyrrolidin-1-yl]nonyl]- 4,4-dimethyl-5-oxo-2- thioxoimidazolidin-1- yl]-2-(trifluoromethyl)- benzotrile (2R)-2-(Methoxymethyl) pyrrolidine	5	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.66m(2H, CH ₂ N); 3.41dd (J=9.3Hz/5.1Hz, 1H, CH ₂ O); 3.35s (3H, CH ₃ O); 3.27dd(J=9.3Hz/5.9Hz, 1H, CH ₂ O); 3.13m(1H, CH ₂ N); 2.80 dddb(J=11.8Hz/8.4Hz/8Hz, 1H, CH ₂ N); 2.53m(1H, CH ₂ N); 2.22dddb (J=11.8Hz/7.6Hz/7.6Hz, 1H, CH ₂ N); 2.15m(1H, CH ₂ N); 1.92-1.67 m(4H, CH ₂); 1.83m(2H, CH ₂); 1.58s (6H, CH ₃); 1.49m(2H, CH ₂); 1.31m (10H, CH ₂)
21	4-[3-[9-[(2S)-2- (Methoxymethyl)- pyrrolidin-1-yl]nonyl]- 4,4-dimethyl-5-oxo-2- thioxoimidazolidin-1- yl]-2-(trifluoromethyl)- benzotrile (2S)-2-(Methoxymethyl) pyrrolidine	5	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.66m(2H, CH ₂ N); 3.41dd (J=9.3Hz/5.1Hz, 1H, CH ₂ O); 3.35s (3H, CH ₃ O); 3.27dd(J=9.3Hz/5.9Hz, 1H, CH ₂ O); 3.13m(1H, CH ₂ N); 2.80 dddb(J=11.8Hz/8.4Hz/8Hz, 1H, CH ₂ N); 2.53m(1H, CH ₂ N); 2.22dddb (J=11.8Hz/7.6Hz/7.6Hz, 1H, CH ₂ N); 2.15m(1H, CH ₂ N); 1.92-1.67 m(4H, CH ₂); 1.83m(2H, CH ₂); 1.58s (6H, CH ₃); 1.49m(2H, CH ₂); 1.31m (10H, CH ₂)
22	4-[3-[6-(3-Hydroxy-8- azabicyclo[3.2.1]oct-8- yl)hexyl]-4,4-dimethyl- 5-oxo-2-thioxoimidazolidin- 1-yl]-2- (trifluoromethyl)- benzotrile 8-Azabicyclo[3.2.1]- octan-3-ol	3	7.95d(J=8Hz, 1H, aryl); 7.90d(J=2.1Hz, 1H, aryl); 7.79dd(J=8Hz/2.1Hz, 1H, aryl); 4.04m(1H, CHOH); 3.69m (2H, CH ₂ N); 3.19m(2H, CHN); 2.35m (2H, CH ₂ N); 2.05m(4H, CH ₂); 1.91m (2H, CH ₂); 1.86m(2H, CH ₂); 1.62m (2H, CH ₂); 1.58s(6H, CH ₃); 1.53m (2H, CH ₂); 1.40m(4H, CH ₂)
23	4-[3-[7-(3-Hydroxy-8- azabicyclo[3.2.1]oct-8- yl)heptyl]-4,4-dimethyl- 5-oxo-2-thioxoimidazolidin- 1-yl]-2- (trifluoromethyl)- benzotrile 8-Azabicyclo[3.2.1]- octan-3-ol	4	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 4.05m(1H, CHOH); 3.66m (2H, CH ₂ N); 3.19m(2H, CHN); 2.33m (2H, CH ₂ N); 2.07m(4H, CH ₂); 1.92m (2H, CH ₂); 1.82m(2H, CH ₂); 1.62m (2H, CH ₂); 1.58s(6H, CH ₃); 1.49m (2H, CH ₂); 1.38m(6H, CH ₂)
24	4-[3-[8-(3-Hydroxy-8- azabicyclo[3.2.1]oct-8- yl)octyl]-4,4-dimethyl-5- oxo-2-thioxoimidazolidin- 1-yl]-2- (trifluoromethyl)- benzotrile 8-Azabicyclo[3.2.1]- octan-3-ol	1	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.78dd(J=8Hz/2.1Hz, 1H, aryl); 4.05m(1H, CHOH); 3.66m (2H, CH ₂ N); 3.19m(2H, CHN); 2.33m (2H, CH ₂ N); 2.07m(4H, CH ₂); 1.92m (2H, CH ₂); 1.81m(2H, CH ₂); 1.62m (2H, CH ₂); 1.59s(6H, CH ₃); 1.48m (2H, CH ₂); 1.38m(8H, CH ₂)

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Example	Product/ Reagent	Instructions analogous to	¹ H-NMR(300 MHz, CDCl ₃) δ [ppm]
25	4-[3-[9-(3-Hydroxy-8-azabicyclo[3.2.1]oct-8-yl)nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzonitrile 8-Azabicyclo[3.2.1]octan-3-ol	5	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 4.04m(1H, CHOH); 3.66m(2H, CH ₂ N); 3.19m(2H, CHN); 2.33m(2H, CH ₂ N); 2.08m(4H, CH ₂); 1.91m(2H, CH ₂); 1.82m(2H, CH ₂); 1.62m(2H, CH ₂); 1.58s(6H, CH ₃); 1.48m(2H, CH ₂); 1.31m(10H, CH ₂)
26	4-[3-[6-[(R)-3-Hydroxypyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzonitrile (R)-Pyrrolidin-3-ol	3	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 4.34m(1H, CHOH); 3.67m(2H, CH ₂ N); 2.89m(1H, CH ₂ N); 2.69m(1H, CH ₂ N); 2.50m(1H, CH ₂ N); 2.46m(2H, CH ₂ N); 2.28m(1H, CH ₂ N); 2.20m(1H, CH ₂); 1.83m(2H, CH ₂); 1.74m(1H, CH ₂); 1.57s(6H, CH ₃); 1.56m(2H, CH ₂); 1.41m(4H, CH ₂)
27	4-[3-[7-[(R)-3-Hydroxypyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzonitrile (R)-Pyrrolidin-3-ol	4	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 4.34m(1H, CHOH); 3.66m(2H, CH ₂ N); 2.89m(1H, CH ₂ N); 2.69m(1H, CH ₂ N); 2.52m(1H, CH ₂ N); 2.45m(2H, CH ₂ N); 2.30m(1H, CH ₂ N); 2.17m(1H, CH ₂); 1.83m(2H, CH ₂); 1.74m(1H, CH ₂); 1.58s(6H, CH ₃); 1.52m(2H, CH ₂); 1.39m(6H, CH ₂)
28	4-[3-[8-[(R)-3-Hydroxypyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzonitrile (R)-Pyrrolidin-3-ol	1	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 4.34m(1H, CHOH); 3.66m(2H, CH ₂ N); 2.89m(1H, CH ₂ N); 2.70m(1H, CH ₂ N); 2.53m(1H, CH ₂ N); 2.45m(2H, CH ₂ N); 2.30m(1H, CH ₂ N); 2.19m(1H, CH ₂); 1.82m(2H, CH ₂); 1.75m(1H, CH ₂); 1.58s(6H, CH ₃); 1.51m(2H, CH ₂); 1.35m(8H, CH ₂)
29	4-[3-[9-[(R)-3-Hydroxypyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzonitrile (R)-Pyrrolidin-3-ol	5	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 4.32m(1H, CHOH); 3.66m(2H, CH ₂ N); 2.87m(1H, CH ₂ N); 2.66m(1H, CH ₂ N); 2.48m(1H, CH ₂ N); 2.41m(2H, CH ₂ N); 2.25m(1H, CH ₂ N); 2.17m(1H, CH ₂); 1.82m(2H, CH ₂); 1.72m(1H, CH ₂); 1.58s(6H, CH ₃); 1.49m(2H, CH ₂); 1.31m(10H, CH ₂)
30	4-[3-[6-[(S)-3-Hydroxypyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzonitrile (S)-Pyrrolidin-3-ol	3	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 4.37m(1H, CHOH); 3.67m(2H, CH ₂ N); 2.94m(1H, CH ₂ N); 2.75m(1H, CH ₂ N); 2.59m(1H, CH ₂ N); 2.51m(2H, CH ₂ N); 2.37m(1H, CH ₂ N); 2.21m(1H, CH ₂); 1.83m(2H, CH ₂); 1.81m(1H, CH ₂); 1.58s(6H, CH ₃); 1.57m(2H, CH ₂); 1.42m(4H, CH ₂)
31	4-[3-[7-[(S)-3-Hydroxypyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzonitrile (S)-Pyrrolidin-3-ol	4	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 4.37m(1H, CHOH); 3.66m(2H, CH ₂ N); 2.95m(1H, CH ₂ N); 2.75m(1H, CH ₂ N); 2.61m(1H, CH ₂ N); 2.52m(2H, CH ₂ N); 2.40m(1H, CH ₂ N); 2.21m(1H, CH ₂); 1.83m(2H, CH ₂); 1.81m(1H, CH ₂); 1.58s(6H, CH ₃); 1.56m(2H, CH ₂); 1.39m(6H, CH ₂)
32	4-[3-[8-[(S)-3-Hydroxypyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzonitrile (S)-Pyrrolidin-3-ol	1	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 4.34m(1H, CHOH); 3.67m(2H, CH ₂ N); 2.89m(1H, CH ₂ N); 2.70

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Example	Product/ Reagent	Instructions analogous to	¹ H-NMR(300 MHz, CDCl ₃) δ [ppm]
	1-yl]-2-(trifluoromethyl)benzotrile (S)-Pyrrolidin-3-ol		m(1H, CH ₂ N); 2.53m(1H, CH ₂ N); 2.45m(2H, CH ₂ N); 2.30m(1H, CH ₂ N); 2.19m(1H, CH ₂); 1.82m(2H, CH ₂); 1.75m(1H, CH ₂); 1.58s(6H, CH ₃); 1.51m(2H, CH ₂); 1.36m(8H, CH ₂)
33	4-[3-[9-[(S)-3-Hydroxypyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile (S)-Pyrrolidin-3-ol	5	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 4.33m(1H, CHOH); 3.66m(2H, CH ₂ N); 2.87m(1H, CH ₂ N); 2.67m(1H, CH ₂ N); 2.49m(1H, CH ₂ N); 2.42m(2H, CH ₂ N); 2.26m(1H, CH ₂ N); 2.17m(1H, CH ₂); 1.82m(2H, CH ₂); 1.72m(1H, CH ₂); 1.58s(6H, CH ₃); 1.49m(2H, CH ₂); 1.31m(10H, CH ₂)
34	4-[4,4-Dimethyl-3-[6-(2-methylpyrrolidin-1-yl)hexyl]-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile 2-Methylpyrrolidine	3	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.67m(2H, CH ₂ N); 3.17m(1H, CH ₂ N); 2.79m(1H, CH ₂ N); 2.27m(1H, CHN); 2.06m(1H, CH ₂ N); 2.03m(1H, CH ₂ N); 2.00-1.35m(4H, CH ₂); 1.83m(2H, CH ₂); 1.57s(6H, CH ₃); 1.57m(2H, CH ₂); 1.41m(4H, CH ₂); 1.10d(J=6Hz, 3H, CH ₃)
35	4-[4,4-Dimethyl-3-[7-(2-methylpyrrolidin-1-yl)heptyl]-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile 2-Methylpyrrolidine	4	7.94d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.66m(2H, CH ₂ N); 3.16m(1H, CH ₂ N); 2.78m(1H, CH ₂ N); 2.24m(1H, CHN); 2.06m(1H, CH ₂ N); 2.02m(1H, CH ₂ N); 2.00-1.35m(4H, CH ₂); 1.82m(2H, CH ₂); 1.57s(6H, CH ₃); 1.52m(2H, CH ₂); 1.38m(6H, CH ₂); 1.10d(J=6Hz, 3H, CH ₃)
36	4-[4,4-Dimethyl-3-[8-(2-methylpyrrolidin-1-yl)octyl]-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile 2-Methylpyrrolidine	1	7.94d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.66m(2H, CH ₂ N); 3.16m(1H, CH ₂ N); 2.77m(1H, CH ₂ N); 2.25m(1H, CHN); 2.05m(1H, CH ₂ N); 2.03m(1H, CH ₂ N); 2.00-1.35m(4H, CH ₂); 1.82m(2H, CH ₂); 1.58s(6H, CH ₃); 1.51m(2H, CH ₂); 1.35m(8H, CH ₂); 1.10d(J=6Hz, 3H, CH ₃)
37	4-[3-[8-[(2R,5S)-rel-2,5-Dimethylpyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile (2R,5S)-rel-2,5-Dimethylpyrrolidine	1	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.78dd(J=8Hz/2.1Hz, 1H, aryl); 3.67m(2H, CH ₂ N); 2.57m(2H, CHN); 2.55m(2H, CH ₂ N); 1.80m(2H, CH ₂); 1.82m(2H, CH ₂); 1.58s(6H, CH ₃); 1.46m(2H, CH ₂); 1.30m(2H, CH ₂); 1.35m(8H, CH ₂); 1.11d(J=6Hz, 6H, CH ₃)
38	4-[3-[6-[(2S)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile (2S)-α,α-Dimethylpyrrolidinemethanol, produced from L-prolinemethyl ester according to K. Nakayama, W. J. Thompson, J. Am. Chem. Soc. 112, 6936-6942 (1990)	3	1.12(s, 3H); 1.21(s, b, 3H); 1.30-1.48(b, 4H); 1.58(s, 6H); 1.49-1.95(b, 9H); 2.33-2.72(b, 3H); 2.73-2.96(b, 1H); 2.99-3.28(b, 1H); 3.65-3.70(m, 2H); 7.77(dd, J=2.11Hz, 8.43Hz, 1H); 7.89(d, J=1.68Hz, 1H); 7.95(d, J=8.43Hz, 1H)
39	4-[3-[7-[(2S)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]heptyl]-4,4-	4	1.11(s, 3H); 1.20(s, b, 3H); 1.30-1.47(b, 6H); 1.58(s, 6H); 1.50-1.92(b, 9H); 2.35-2.68(b, 3H); 2.70-2.91(b, 1H); 3.00-3.22(b, 1H); 3.64-3.69(m, 2H);

-continued

Example	Product/ Reagent	Instructions analogous to	¹ H-NMR(300 MHz, CDCl ₃) δ [ppm]
	dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzotrile (2S)-α,α-Dimethylpyrrolidinemethanol		7.77(dd, J=1.90Hz, 8.26Hz; 1H); 7.89(d, J=2.10Hz; 1H); 7.95(d, J=8.43Hz; 1H)
40	4-[3-[8-[(2S)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzotrile (2S)-α,α-Dimethylpyrrolidinemethanol	1	1.12(s, 3H); 1.22(s, b, 3H); 1.27-1.44(b, 8H); 1.58(s, 6H); 1.50-1.92(b, 10H); 2.36-2.70(b, 2H); 2.71-2.95(b, 1H); 3.00-3.23(b, 1H); 3.64-3.70(m, 2H); 7.77(dd, J=2.11Hz, 8.43Hz; 1H); 7.89(d, J=2.11Hz; 1H); 7.95(d, J=8.43Hz; 1H)
41	4-[3-[9-[(2S)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzotrile (2S)-α,α-Dimethylpyrrolidinemethanol	5	1.11(s, 3H); 1.18(s, b, 3H); 1.24-1.44(b, 10H); 1.45-1.55(b, 2H); 1.58(s, 6H); 1.63-1.90(b, 7H); 2.30-3.20(b, 5H); 3.64-3.69(m, 2H); 7.78(dd, J=2.10Hz, 8.43Hz; 1H); 7.90(d, J=2.53Hz; 1H); 7.95(d, J=8.01Hz; 1H)
42	4-[3-[6-[(2R)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzotrile (2R)-α,α-Dimethylpyrrolidinemethanol, produced from D-prolinemethyl ester according to K. Nakayama, W. J. Thompson, J. Am. Chem. Soc. 112, 6936-6942 (1990)	3	1.10(s, 3H); 1.18(s, b, 3H); 1.29-1.47(b, 4H); 1.58(s, 6H); 1.48-1.91(b, 9H); 2.30-2.65(b, 3H); 2.68-2.92(b, 1H); 2.95-3.18(b, 1H); 3.64-3.70(m, 2H); 7.77(dd, J=1.90Hz, 8.01Hz; 1H); 7.89(d, J=1.68Hz; 1H); 7.95(d, J=8.43Hz; 1H)
43	4-[3-[7-[(2R)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzotrile (2R)-α,α-Dimethylpyrrolidinemethanol	4	1.09(s, 3H); 1.17(s, b, 3H); 1.29-1.44(b, 6H); 1.58(s, 6H); 1.46-1.90(b, 9H); 2.33-2.64(b, 3H); 2.68-2.86(b, 1H); 3.97-3.15(b, 1H); 3.64-3.69(m, 2H); 7.77(dd, J=2.11Hz, 8.01Hz; 1H); 7.89(d, J=2.10Hz; 1H); 7.95(d, J=8.01Hz; 1H)
44	4-[3-[8-[(2R)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzotrile (2R)-α,α-Dimethylpyrrolidinemethanol	1	1.11(s, 3H); 1.18(s, b, 3H); 1.23-1.44(b, 8H); 1.58(s, 6H); 1.45-1.90(b, 9H); 2.30-3.15(b, 5H); 3.64-3.69(m, 2H); 7.78(dd, J=2.10Hz, 8.43Hz; 1H); 7.90(d, J=2.11Hz; 1H); 7.95(d, J=8.43Hz; 1H)
45	4-[3-[6-[(2S)-2-(1-Hydroxy-1-ethyl-propyl)pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzotrile (2S)-α,α-Diethylpyrrolidinemethanol, produced from L-proline methyl ester according to K. Nakayama, W. J. Thompson,	3	0.83(t, J=8.22Hz; 6H); 1.25-1.46(b, 6H); 1.59(s, 6H); 1.47-1.94(b, 11H); 2.36-2.56(b, 2H); 2.58-2.77(b, 2H); 2.87-3.03(b, 1H); 3.64-3.70(m, 2H); 7.77(dd, J=2.11Hz, 8.01Hz; 1H); 7.90(d, J=2.11Hz; 1H); 7.95(d, J=8.00Hz; 1H)

-continued

Example	Product/ Reagent	Instructions analogous to	¹ H-NMR(300 MHz, CDCl ₃) δ [ppm]
	J. Am. Chem. Soc. 112, 6936- 6942(1990)		
46	4-[3-[7-[(2S)-2-(1-Hydroxy-1-ethylpropyl)-pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzotrile (2E) α,α-Diethylpyrrolidinemethanol	4	0.80-0.91(m, 6H); 1.22-1.47(b, 8H); 1.46-1.95(b, 11H); 1.58(s, 6H); 2.30-3.30(b, 5H); 3.64-3.70(m, 2H); 7.77(dd, J=2.11Hz, 8.01Hz; 1H); 7.89(d, J=1.60Hz; 1H); 7.95(d, J=8.01Hz; 1H)
47	4-[3-[8-[(2S)-2-(1-Hydroxy-1-ethylpropyl)-pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)-benzotrile (2S)-α,α-Diethylpyrrolidine-methanol	1	0.84(t, J=7.80Hz; 6H); 1.20-1.43(b, 10H); 1.44-1.62(b, 4H); 1.59(s, 6H); 1.62-1.90(b, 7H); 2.35-2.56(b, 2H); 2.57-2.76(b, 2H); 2.85-3.04(b, 1H); 3.64-3.70(m, 2H); 7.78(dd, J=2.32Hz, 8.43Hz; 1H); 7.90(d, J=1.68Hz; 1H); 7.95(d, J=8.43Hz; 1H)
48	(S)-1-[7-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]heptyl]pyrrolidine-2-carboxylic acidmethyl ester L-Prolinemethyl ester, Hydrochloride	4	7.93d(J=8Hz, 1H, aryl); 7.88d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 3.70s(3H, OCH ₃); 3.65m(2H, CH ₂ N); 3.17m(1H, CH ₂ N); 3.11dd(J=9Hz/6Hz, 1H, CHN); 2.64m(1H, CH ₂ N); 2.33m(1H, CH ₂ N); 2.31m(1H, CH ₂ N); 2.09m(1H, CH ₂); 1.90m(2H, CH ₂); 1.82m(2H, CH ₂); 1.80m(1H, CH ₂); 1.56s(6H, CH ₃); 1.49m(2H, CH ₂); 1.35m(6H, CH ₃)
49	(S)-1-[6-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]hexyl]pyrrolidine-4-carboxamide L-Prolinamide	3	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 7.22m(1H, CONH ₂); 5.61m(1H, CONH ₂); 3.66m(2H, CH ₂ N); 3.18m(1H); 2.99m(1H); 2.64m(1H); 2.44m(1H); 2.28m(1H); 2.17m(1H); 1.83m(2H, CH ₂); 1.80m(3H, CH ₂); 1.58s(6H, CH ₃); 1.51m(2H, CH ₂); 1.40m(4H, CH ₂)
50	(S)-1-[7-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]heptyl]pyrrolidine-4-carboxamide L-Prolinamide	4	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 7.26m(1H, CONH ₂); 5.39m(1H, CONH ₂); 3.66m(2H, CH ₂ N); 3.18m(1H); 3.00m(1H); 2.63m(1H); 2.45m(1H); 2.30m(1H); 2.18m(1H); 1.89m(1H, CH ₂); 1.81m(2H, CH ₂); 1.80m(2H, CH ₂); 1.58s(6H, CH ₃); 1.52m(2H, CH ₂); 1.38m(6H, CH ₃)
51	(S)-1-[8-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]octyl]pyrrolidine-4-carboxamide L-Prolinamide	1	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 7.27m(1H, CONH ₂); 5.46m(1H, CONH ₂); 3.66m(2H, CH ₂ N); 3.19m(1H); 3.00m(1H); 2.63m(1H); 2.44m(1H); 2.30m(1H); 2.18m(1H); 1.90m(1H, CH ₂); 1.81m(3H, CH ₂); 1.66m(1H, CH ₂); 1.58s(6H, CH ₃); 1.49m(2H, CH ₂); 1.35m(8H, CH ₂)
52	(S)-1-[6-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]hexyl]pyrrolidine-4-carboxamide D-Prolinamide	1	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 7.77m(1H, CONH ₂); 5.57m(1H, CONH ₂); 3.66m(2H, CH ₂ N); 3.18m(1H); 2.99m(1H); 2.64m(1H); 2.44m(1H); 2.28m(1H); 2.17m(1H); 1.83m(2H, CH ₂); 1.80m(3H, CH ₂); 1.58s(6H, CH ₃); 1.51m(2H, CH ₂); 1.40m(4H, CH ₂)
53	(R)-1-[7-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-	4	7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 7.26m(1H, CONH ₂); 5.36m(1H, CONH ₂); 3.66m(2H, CH ₂ N); 3.18m(1H); 3.00m(1H); 2.63m(1H); 2.45

-continued

Example	Product/ Reagent	Instructions analogous to	¹ H-NMR(300 MHz, CDCl ₃) δ [ppm]
54	yl]heptyl]pyrrolidine-4-carboxamide D-Prolinamide (R)-1-[8-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]octyl]pyrrolidine-4-carboxamide D-Prolinamide	1	m(1H); 2.30m(1H); 2.18m(1H); 1.89m(1H, CH ₂); 1.81m(2H, CH ₂); 1.80m(2H, CH ₂); 1.58s(6H, CH ₃); 1.52m(2H, CH ₂); 1.38m(6H, CH ₃) 7.95d(J=8Hz, 1H, aryl); 7.89d(J=2.1Hz, 1H, aryl); 7.77dd(J=8Hz/2.1Hz, 1H, aryl); 7.24m(1H, CONH ₂); 5.53m(1H, CONH ₂); 3.66m(2H, CH ₂ N); 3.19m(1H); 3.00m(1H); 2.63m(1H); 2.44m(1H); 2.30m(1H); 2.18m(1H); 1.90m(1H, CH ₂); 1.81m(3H, CH ₃); 1.66m(1H, CH ₂); 1.58s(6H, CH ₃); 1.49m(2H, CH ₂); 1.35m(8H, CH ₂)
55	4-[3-[6-[(2S)-4,4-Dimethyl-5-oxo-2-(pyrrolidin-1-ylmethyl)pyrrolidin-1-yl]hexyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile (2S)-2-(Pyrrolidin-1-ylmethyl)pyrrolidine, produced from L-proline according to M. Amedjkouh, P. Ahlberg, Tetrahedron: Asymmetry 13, 2229-2234(2002)	3	1.34-1.49(b, 6H); 1.58(s, 6H); 1.65-1.89(b, 8H); 1.90-2.09(b, 2H); 2.10-2.37(b, 2H); 2.38-2.78(b, 7H); 2.84-3.01(b, 1H); 3.15-3.26(b, 1H); 3.64-3.70(m, 2H); 7.77(dd, J=2.1Hz, 8.43Hz; 1H); 7.89(d, J=1.68Hz; 1H); 7.95(d, J=8.01Hz; 1H)
56	4-[3-[7-[(2S)-4,4-Dimethyl-5-oxo-2-(pyrrolidin-1-ylmethyl)pyrrolidin-1-yl]heptyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile (2S)-2-(Pyrrolidin-1-ylmethyl)pyrrolidine	4	1.28-1.44(b, 6H); 1.44-1.88(b, 10H); 1.58(s, 6H); 1.89-2.07(b, 2H); 2.08-2.25(b, 2H); 2.31-2.70(b, 7H); 2.81-2.93(b, 1H); 3.11-3.21(b, 1H); 3.64-3.69(m, 2H); 7.77(dd, J=2.1Hz, 8.43Hz; 1H); 7.89(d, J=1.68Hz; 1H); 7.95(d, J=8.43Hz; 1H)
57	4-[3-[8-[(2S)-4,4-Dimethyl-5-oxo-2-(pyrrolidin-1-ylmethyl)pyrrolidin-1-yl]octyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile (2S)-2-(Pyrrolidin-1-ylmethyl)pyrrolidine	1	1.22-1.43(b, 8H); 1.48-1.60(b, 2H); 1.58(s, 6H); 1.65-1.87(b, 8H); 1.92-2.08(b, 2H); 2.09-2.29(b, 2H); 2.33-2.62(b, 6H); 2.62-2.76(b, 1H); 2.80-2.97(b, 1H); 3.11-3.25(b, 1H); 3.64-3.69(m, 2H); 7.77(dd, J=1.69Hz, 8.01Hz; 1H); 7.89(d, J=1.26Hz; 1H); 7.95(d, J=8.43Hz; 1H)

EXAMPLE 58

(S)-1-[7-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]heptyl]pyrrolidine-2-carboxylic acid hydrochloride

[0139] 20 mg of the compound that is produced under 48 is stirred with a spatula tip full of potassium carbonate in 1 ml of methanol overnight at room temperature. The reaction mixture is acidified with 4 molar aqueous hydrochloric acid and extracted with ethyl acetate. The organic phase is washed with saturated sodium chloride solution and concentrated by evaporation in a vacuum. 12 mg of the title compound is obtained.

[0140] ¹H-NMR (300 MHz, CD₃OD): δ[ppm]=8.15 d (J=8 Hz, 1H, aryl); 8.10 d (J=2.1 Hz, 1H, aryl); 7.93 dd (J=8 Hz/2.1 Hz, 1H, aryl); 3.90 dd (J=9 Hz/6 Hz, 1H, CHN); 3.77 m (3H, CH₂N); 3.14 m (1H, CH₂N); 3.26 m (1H, CH₂N); 3.14 m (2H, CH₂N); 2.45 m (1H, CH₂); 2.14 m (2H, CH₂); 1.98 m (1H, CH₂); 1.89 m (2H, CH₂); 1.77 m (2H, CH₂); 1.60 s (6H, CH₃); 1.47 m (6H, CH₂).

[0141] Without further elaboration, it is believed that one skilled in the art can, using the preceding description, utilize the present invention to its fullest extent. The preceding preferred specific embodiments are, therefore, to be construed as merely illustrative, and not limitative of the remainder of the disclosure in any way whatsoever.

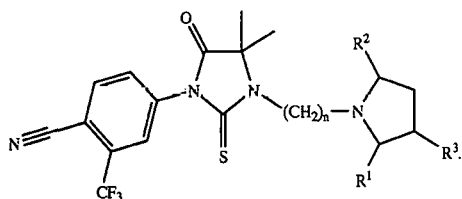
[0142] In the foregoing and in the examples, all temperatures are set forth uncorrected in degrees Celsius and, all parts and percentages are by weight, unless otherwise indicated.

[0143] The entire disclosures of all applications, patents and publications, cited herein and of corresponding Germany application No. 103 22 108.5, filed May 9, 2003, and U.S. Provisional Application Ser. No. 60/470,182, filed May 14, 2003, are incorporated by reference herein.

[0144] The preceding examples can be repeated with similar success by substituting the generically or specifically described reactants and/or operating conditions of this invention for those used in the preceding examples.

[0145] From the foregoing description, one skilled in the art can easily ascertain the essential characteristics of this invention and, without departing from the spirit and scope thereof, can make various changes and modifications of the invention to adapt it to various usages and conditions.

1. Compounds of general formula 1:



in which

n means an integer between 6 and 9,

R₁ and R₂, independently of one another, mean a hydrogen atom, an unbranched C₁-C₄-alkyl group, a branched C₃-C₅-alkyl group, an unbranched hydroxy-C₁-C₄-alkyl group, a branched hydroxy-C₃-C₅-alkyl group, an unbranched C₁-C₄-alkoxy-C₁-C₄-alkyl group, a branched C₁-C₄-alkoxy-C₃-C₅-alkyl group, an unbranched C₁-C₄-alkanoyloxy-C₁-C₄-alkyl group, a branched C₁-C₄-alkanoyloxy-C₃-C₅-alkyl group, a (pyrrolidin-1-yl)methyl group, a carboxy group, a C₁-C₄-alkoxycarbonyl group or an aminocarbonyl group,

or

R₁ and R₂ together mean a 2-hydroxypropane-1,3-diyl bridge;

R₃ means a hydrogen atom or a hydroxy group, as well as their pharmacologically compatible salts.

2. Compounds of general formula I according to claim 1, characterized in that R₁ represents a hydrogen atom, a hydroxymethyl group, an aminocarbonyl group or a methoxymethyl group; R₂ and R₃ in each case represent a hydrogen atom.

3. Compounds of general formula I according to claim 1, wherein R₁ represents a hydrogen atom, or a methyl group; and R₂ and R₃ in each case represent a hydrogen atom.

4. Compounds of general formula I according to claim 1, wherein R₁ and R₂ together represent a 2-hydroxypropane-1,3-diyl bridge; R₃ represents a hydrogen atom.

5. Compounds according to claim 1, namely

4-[4,4-Dimethyl-5-oxo-3-[8-(pyrrolidin-1-yl)octyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[4,4-Dimethyl-5-oxo-3-[8-(pyrrolidin-1-yl)octyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile hydrochloride

4-[4,4-Dimethyl-5-oxo-3-[6-(pyrrolidin-1-yl)hexyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[4,4-Dimethyl-5-oxo-3-[7-(pyrrolidin-1-yl)heptyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[4,4-Dimethyl-5-oxo-3-[9-(pyrrolidin-1-yl)nonyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[6-[(2R)-2-(Hydroxymethyl)pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[6-[(2S)-2-(Hydroxymethyl)pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[7-[(2R)-2-(Hydroxymethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[7-[(2S)-2-(Hydroxymethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[8-[(2R)-2-(Hydroxymethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[8-[(2S)-2-(Hydroxymethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[9-[(2R)-2-(Hydroxymethyl)pyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[9-[(2S)-2-(Hydroxymethyl)pyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[6-[(2R)-2-(Methoxymethyl)pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[6-[(2S)-2-(Methoxymethyl)pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[7-[(2R)-2-(Methoxymethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[7-[(2S)-2-(Methoxymethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[8-[(2R)-2-(Methoxymethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[8-[(2S)-2-(Methoxymethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[9-[(2R)-2-(Methoxymethyl)pyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

4-[3-[9-[(2S)-2-(Methoxymethyl)pyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

- 4-[3-[6-(3-Hydroxy-8-azabicyclo[3.2.1]oct-8-yl)hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[7-(3-Hydroxy-8-azabicyclo[3.2.1]oct-8-yl)heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[8-(3-Hydroxy-8-azabicyclo[3.2.1]oct-8-yl)octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[9-(3-Hydroxy-8-azabicyclo[3.2.1]oct-8-yl)nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[6-[(R)-3-Hydroxypyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[7-[(R)-3-Hydroxypyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[8-[(R)-3-Hydroxypyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[9-[(R)-3-Hydroxypyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[6-[(S)-3-Hydroxypyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[7-[(S)-3-Hydroxypyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[8-[(S)-3-Hydroxypyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[9-[(S)-3-Hydroxypyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[4,4-Dimethyl-3-[6-(2-methylpyrrolidin-1-yl)hexyl]-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[4,4-Dimethyl-3-[7-(2-methylpyrrolidin-1-yl)heptyl]-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[4,4-Dimethyl-3-[8-(2-methylpyrrolidin-1-yl)octyl]-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[8-[(2R,5S)-rel-2,5-Dimethylpyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[6-[(2S)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[7-[(2S)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[8-[(2S)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[9-[(2S)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]nonyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[6-[(2R)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[7-[(2R)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[8-[(2R)-2-(1-Hydroxy-1-methylethyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[6-[(2S)-2-(1-Hydroxy-1-ethylpropyl)pyrrolidin-1-yl]hexyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[7-[(2S)-2-(1-Hydroxy-1-ethylpropyl)pyrrolidin-1-yl]heptyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[8-[(2S)-2-(1-Hydroxy-1-ethylpropyl)pyrrolidin-1-yl]octyl]-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- (S)-1-[7-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]heptyl]pyrrolidine-2-carboxylic acid methyl ester
- (S)-1-[6-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]hexyl]pyrrolidine-4-carboxamide
- (S)-1-[7-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]heptyl]pyrrolidine-4-carboxamide
- (S)-1-[8-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]octyl]pyrrolidine-4-carboxamide
- (R)-1-[6-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]hexyl]pyrrolidine-4-carboxamide
- (R)-1-[7-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]heptyl]pyrrolidine-4-carboxamide
- (R)-1-[8-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]octyl]pyrrolidine-4-carboxamide
- 4-[3-[6-[(2S)-4,4-Dimethyl-5-oxo-2-(pyrrolidin-1-yl)methyl]pyrrolidin-1-yl]hexyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[7-[(2S)-4,4-Dimethyl-5-oxo-2-(pyrrolidin-1-yl)methyl]pyrrolidin-1-yl]heptyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile
- 4-[3-[8-[(2S)-4,4-Dimethyl-5-oxo-2-(pyrrolidin-1-yl)methyl]pyrrolidin-1-yl]octyl]-2-thioxoimidazolidin-1-yl]-2-(trifluoromethyl)benzotrile

(S)-1-[7-[3-[4-Cyano-3-(trifluoromethyl)phenyl]-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]heptyl]pyrrolidine-2-carboxylic acid hydrochloride

6. Pharmaceutical compositions that contain at least one compound of general formula I or their pharmacologically compatible salts according to claim 1, together with pharmaceutically compatible adjuvants and/or vehicles.

7. Use of the compounds of general formula I according to claim 1 for the production of a pharmaceutical agent for treatment or prophylaxis of diseases of the human or animal body, which can be influenced by the inhibition of the androgen receptor.

8. Use according to claim 7, wherein the diseases to be prevented or treated are androgen-dependent proliferative diseases.

9. Use according to claim 7, wherein the diseases to be treated are tumor diseases.

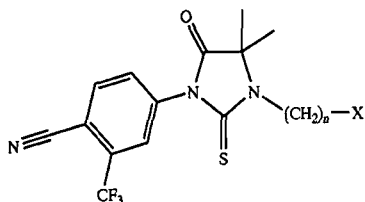
10. Use according to claim 7, wherein the disease to be treated is prostate cancer.

11. Use according to claim 7, wherein the diseases to be prevented or treated are androgen-dependent, non-proliferative diseases.

12. Use according to claim 11, wherein the diseases to be prevented or treated are androgenetic alopecia, hirsutism or acne.

13. Use according to claim 7, wherein the disease is benign prostate hyperplasia.

14. Process for the production of the compounds of general formula I according to the invention according to claim 1, wherein compounds of general formula II



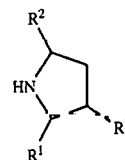
II

in which

n can mean an integer between 6 and 9,

X can mean a leaving group,

are reacted in the presence of an organic base with compounds of general formula III



III

in which

R1 and R2, independently of one another, can mean a hydrogen atom, an unbranched C₁-C₄-alkyl group, a branched C₃-C₅-alkyl group, an unbranched hydroxy-C₁-C₄-alkyl group, a branched hydroxy-C₃-C₅-alkyl group, an unbranched C₁-C₄-alkoxy-C₁-C₄-alkyl group, a branched C₁-C₄-alkoxy-C₃-C₅-alkyl group, an unbranched C₁-C₄-alkanoyloxy-C₁-C₄-alkyl group, a branched C₁-C₄-alkanoyloxy-C₃-C₅-alkyl group, a (pyrrolidin-1-yl)methyl group, a carboxy group, a C₁-C₄-alkoxycarbonyl group or an aminocarbonyl group,

or

R1 and R2 together can mean a 2-hydroxypropano-1,3-diyl bridge;

R3 can mean a hydrogen atom or a hydroxy group.

* * * * *



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(57) **ABSTRACT**

The present invention provides a compound represented by formula (I):

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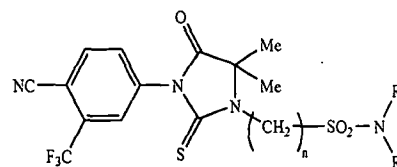
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Jun. 12, 2003 (JP) 2003-168267



wherein n is an integer selected from 1 to 20, and R¹ and R², which may be the same or different, each represent a hydrogen atom, or a linear or branched C₁-C₆ alkyl group, or a salt, a prodrug or a solvate thereof, as well as a drug, a pharmaceutical composition containing the compound, and the like.

IMIDAZOLIDINE DERIVATIVES

FIELD OF THE INVENTION

[0001] The present invention relates to imidazolidine derivatives which have a substituted alkyl group in 3-position, and a drug containing these imidazolidine derivatives as an active ingredient.

BACKGROUND ART

[0002] It has been made clear in the past that the male hormone androgen plays an important role in prostate cancer; benign prostatic hypertrophy, male pattern baldness, sexual precociousness, common acne, seborrhea and hypertrichosis. For example, it is known that persons who have been castrated and persons suffering from sexual gland failure almost never develop prostate cancer or benign prostatic hypertrophy.

[0003] For example, cyproterone acetate, chlormadinone acetate, flutamide, bicalutamide and the like are used as anti-androgen agents, i.e., androgen receptor antagonists. These anti-androgen agents show an effect in many cases such as drug therapy in prostate cancer, and constitute important treatment drugs in this area. Furthermore, it is known that cyproterone acetate suppresses the occurrence of baldness and the progression of acne in teenagers. Furthermore, in females, cyproterone acetate is used in the treatment of androgenization and hair loss. Flutamide and bicalutamide are used as prostate cancer treatment agents.

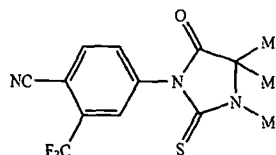
[0004] However, as problems encountered in these anti-androgen agents, it is known that even if the anti-androgen agents are effective, the disease recurs in almost all cases in two to five years, and in such cases, androgen resistance appears.

[0005] Furthermore, it has been reported that hydroxyflutamide, which is the active form of flutamide, causes an increase in androgen receptor transcription activity at a concentration of 10 $\mu\text{mol/L}$. Moreover, the hydroxyflutamide concentration in the blood in prostate cancer patients treated with flutamide is several $\mu\text{mol/L}$. However, it has been reported that this concentration reaches a concentration at which hydroxyflutamide shows an agonist effect (see Non-patent Document 1).

[0006] Furthermore, it has been reported that there is an increase in the weight of the prostate gland when cyproterone acetate and chlormadinone acetate are continuously administered to castrated rats for two weeks (see Non-patent Document 2). Moreover, in regard to flutamide and bicalutamide, there are also reports of side effects such as liver-toxicity and the like. Accordingly, there is a demand for an anti-androgen agent which has a sufficient antagonistic effect, and in which these problems have been solved.

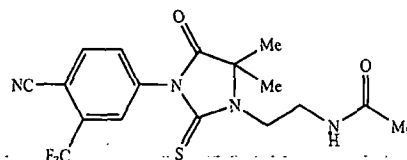
[0007] Meanwhile, the compounds represented by the following formula described in Japanese Patent Application No. 4-308579 A (Patent Document 1) and the corresponding European Patent Application No 494819 A (Patent Document 2) are known as phenylimidazolidines that show anti-androgen activity.

[Formula 1]



[0008] Furthermore, the compounds represented by the following formula described in Japanese Patent Application No. 10-510845 A (Patent Document 3) and the corresponding International Patent Publication WO 97/00071 (Patent Document 4) are known as substituted phenylimidazolidines that show anti-androgen activity.

[Formula 2]



[0009] However, the compounds likewise do not constitute means for solving the problems of existing anti-androgen agents.

[Patent Document 1]

[0010] Japanese Patent Application No. 4-308579 A

[Patent Document 2]

[0011] European Patent Application No 494819 A

[Patent Document 3]

[0012] Japanese Patent Application No. 10-510845 A

[Patent Document 4]

[0013] International Patent Publication WO 97/00071

[Non-Patent Document 1]

[0014] J. Biol. Chem., Vol. 270, pp. 19998-20003, 1995

[Non-Patent Document 2]

[0015] Journal of the Endocrine Society of Japan, Vol. 66, pp. 597-606, 1990

DISCLOSURE OF THE INVENTION

Problems to be Solved

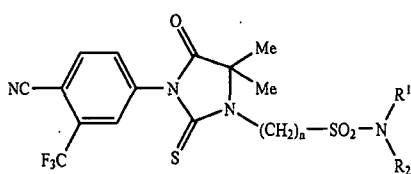
[0016] It is one object of the present invention to provide imidazolidine derivatives which have a substituted alkyl group in 3-position, and which show a useful activity as drugs, especially an anti-androgen activity, and salts, prodrugs or solvates thereof.

[0017] It is another object of the present invention to provide drugs containing the abovementioned imidazolidine derivatives.

Means for Solving the Problems

[0018] The present inventors conducted diligent research with the aim of solving the abovementioned problems. As a result of this research, the inventors found that imidazolidine derivatives having a sulfonamide group represented by Formula (I) show anti-androgen activity, and show no or almost no agonist activity, and then completed the present invention.

[0019] Specifically, the present invention provides a compound represented by formula (I):



wherein n is an integer selected from 1 to 20, and R^1 and R^2 , which may be the same or different, each represent a hydrogen atom or a linear or branched C_1 - C_6 alkyl group, or a salt, a prodrug or a solvate thereof. Also, the present invention provides a compound represented by formula (I) wherein n is an integer selected from 1 to 10, or a salt, a prodrug or a solvate thereof. Also, the present invention provides a compound represented by formula (I) wherein R^1 and R^2 are each a hydrogen atom, or a salt, a prodrug or a solvate thereof. Also, the present invention provides a compound represented by formula (I) wherein at least one of R^1 and R^2 is a methyl group, or a salt, a prodrug or a solvate thereof. Further, the present invention provides a compound represented by formula (I), which is selected from the group consisting of:

[0020] 4-[3'-(3"-aminosulfonylpropyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0021] 4-[3'-(4"-aminosulfonylbutyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0022] 4-[3'-(6"-aminosulfonylhexyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0023] 4-[3'-(7"-aminosulfonylheptyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0024] 4-[3'-(8"-aminosulfonyloctyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0025] 4-[3'-(9"-aminosulfonylnonyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0026] 4-[3'-(5"-aminosulfonylpentyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0027] 4-[3'-(4"-N,N-dimethylaminosulfonylbutyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0028] 4-[3'-(3"-N,N-dimethylaminosulfonylpropyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0029] 4-[3'-(5"-N,N-dimethylaminosulfonylpentyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0030] 4-[3'-(6"-N,N-dimethylaminosulfonylhexyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

(I) [0031] 4-[3'-(7"-N,N-dimethylaminosulfonylheptyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0032] 4-[3'-(8"-N,N-dimethylaminosulfonyloctyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0033] 4-[3'-(9"-N,N-dimethylaminosulfonylnonyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0034] 4-[3'-(3"-N-methylaminosulfonylpropyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0035] 4-[3'-(4"-N-methylaminosulfonylbutyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole;

[0036] 4-[3'-(5"-N-methylaminosulfonylpentyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole; and

[0037] 4-[3'-(2"-aminosulfonylethyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole or a salt, a prodrug or a solvate thereof.

[0038] According to another aspect of the present invention, there provides a drug comprising a compound represented by formula (I) or a salt, a prodrug or a solvate thereof as an active ingredient.

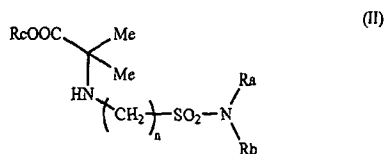
[0039] According to still another aspect of the present invention, there provides a pharmaceutical composition comprising a compound represented by formula (I) or a salt, a prodrug or a solvate thereof as an active ingredient. Also, the present invention provides an anti-androgen agent comprising a compound represented by formula (I) or a salt, a prodrug or a solvate thereof as an active ingredient. Also, the present invention provides a prophylactic or therapeutic agent for a disease selected from prostate cancer, benign prostatic hypertrophy, male pattern baldness, sexual precociousness, common acne, seborrhea and hypertrichosis, which comprises a compound represented by formula (I) or a salt, a prodrug or a solvate thereof as an active ingredient.

[0040] According to still another aspect of the present invention, there provides the use of a compound represented by formula (I) or a salt, a prodrug or a solvate thereof in manufacturing a medicament that acts as an androgen receptor antagonist.

[0041] According to still another aspect of the present invention, there provides a process for preparing a compound represented by formula (I), which comprises the steps of:

[0042] reacting a compound represented by formula (II):

[Formula 4]



wherein

[0043] n is an integer selected from 1 to 20;

[0044] Ra and Rb, which may be the same or different, are each selected from the group consisting of a linear or branched C₁-C₆ alkyl group substituted with one or more W¹, a linear or branched C₁-C₆ alkylcarbonyl group which may be substituted with one or more W¹, an arylcarbonyl group which may be substituted with one or more W², a linear or branched C₁-C₆ alkoxycarbonyl group which may be substituted with one or more W¹, an aryloxycarbonyl group which may be substituted with one or more W², a linear or branched C₁-C₆ alkylaminocarbonyl group which may be substituted with one or more W¹, a linear or branched C₁-C₆ dialkylaminocarbonyl group which may be substituted with one or more W¹, a linear or branched C₁-C₆ alkylsulfonyl group which may be substituted with one or more W¹, an arylsulfonyl group which may be substituted with one or more W², and R¹ and R²;

or

[0045] Ra and Rb may be joined together to form a group =CH-W³;

[0046] W¹ is a linear or branched C₁-C₆ alkoxy group, a linear or branched C₁-C₆ alkylthio group, a linear or branched C₁-C₆ alkylsulfinyl group, a linear or branched C₁-C₆ alkylsulfonyl group, an aryl group which may be substituted with one or more W², an aryloxy group which may be substituted with one or more W², or a C₁-C₃ aralkyloxy group which may be substituted with one or more W²;

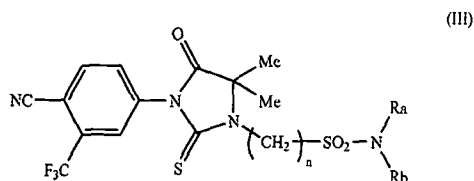
[0047] W² is a linear or branched C₁-C₆ alkyl group, a linear or branched C₁-C₆ alkoxy group, a linear or branched C₁-C₆ haloalkyl group, a halogen atom, a cyano group, or a nitro group;

[0048] W³ is a linear or branched C₁-C₆ alkyl group, a linear or branched C₁-C₆ alkoxy group, a linear or branched C₁-C₆ alkylamino group, or a linear or branched C₁-C₆ dialkylamino group;

[0049] R¹ and R² are as defined above; and

[0050] Rc is a linear or branched C₁-C₆ alkyl group with 4-cyano-3-trifluoromethylphenyl isothiocyanate to obtain a compound represented by formula (III):

[Formula 5]

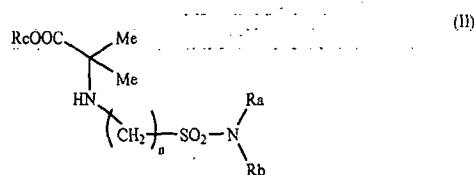


wherein n, Ra and Rb are as defined above; and

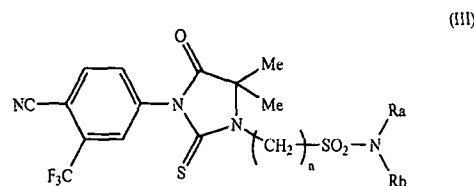
[0051] a deprotection in cases where at least one of the groups Ra and Rb is other than R¹ and R².

[0052] According to still another aspect of the present invention, there also provides, as a synthetic intermediate for a compound represented by formula (I), a compound represented by formula (II):

[Formula 7]



wherein n, Ra, Rb and Rc are as defined herein above] and a compound represented by formula (III):



wherein n, Ra and Rb are as defined herein above, or a salt, a prodrug or a solvate thereof.

[0053] According to still another aspect of the present invention, there provides a method for preventing or treating a disease, which comprises administering a compound represented by formula (I) or a salt, a prodrug or a solvate thereof.

EFFECT OF THE INVENTION

[0054] The present invention makes it possible to provide imidazolidine derivatives that can be an antiandrogen that shows no occurrence of androgen resistance as a result of long-term administration, and/or side effects such as liver toxicity or the like.

PREFERRED MODE FOR CARRYING OUT
THE INVENTION

[0055] In the present invention, the following terms includes the meanings described below unless specifically noted otherwise.

[0056] Examples of a linear or branched C_1 - C_6 alkyl group include methyl group, ethyl group, n-propyl group, i-propyl group, n-butyl group, s-butyl group, i-butyl group, t-butyl group, n-pentyl group, 3-methylbutyl group, 2-methylbutyl group, 1-methylbutyl group, 1-ethylpropyl group, n-hexyl group and the like. A linear or branched alkyl group with 1 to 3 carbon atoms is preferable, and methyl group is more preferable, as R^1 or R^2 in formula (I) of the present invention.

[0057] A linear or branched C_1 - C_6 alkoxy group is a group that has the already-defined alkyl group as alkyl moieties. Examples include methoxy group, ethoxy group, n-propoxy group, i-propoxy group, n-butoxy group, s-butoxy group, i-butoxy group, t-butoxy group, n-pentoxy group, 3-methylbutoxy group, 2-methylbutoxy group, 1-methylbutoxy group, 1-ethylpropoxy group, n-hexyloxy group and the like.

[0058] A linear or branched C_1 - C_6 alkylcarbonyl group is a group that has the already-defined alkyl group as alkyl moieties. Examples include acetyl group, propionyl group, 2-methylpropionyl group, 2,2-dimethylpropionyl group and the like.

[0059] The term aryl group refers to a single-ring or fused-ring arylhydrocarbon group with 6 to 14 carbon atoms. Examples of such a group include phenyl group, 1-naphthyl group, 2-naphthyl group, anthracenyl group and the like. Furthermore, the same is true in cases where an aryl group is included as parts of other substituents.

[0060] An aryloxy group is a group that has the already-defined aryl as aryl moieties. Examples include phenoxy group, 1-naphthoxy group, 2-naphthoxy group and the like.

[0061] Examples of arylcarbonyl group include benzoyl group, 1-naphthoyl group, 2-naphthoyl group and the like.

[0062] A linear or branched C_1 - C_6 alkoxy carbonyl group is a group that has the already-defined alkyl group as alkyl moieties. Examples include methoxycarbonyl group, ethoxycarbonyl group, t-butoxycarbonyl group and the like.

[0063] Examples of an aryloxy carbonyl group include phenoxy carbonyl group, 1-naphthoxy carbonyl group, 2-naphthoxy carbonyl group and the like.

[0064] A linear or branched C_1 - C_6 alkylaminocarbonyl group is a group that has the already-defined alkyl group as alkyl moieties. Examples include methylaminocarbonyl group, ethylaminocarbonyl group, t-butylaminocarbonyl group and the like.

[0065] A linear or branched C_1 - C_6 dialkylaminocarbonyl group is a group that has the already-defined alkyl group as alkyl moieties. Examples include dimethylaminocarbonyl group, diethylaminocarbonyl group, diisopropylaminocarbonyl group, methyl-t-butylaminocarbonyl group and the like.

[0066] A linear or branched C_1 - C_6 alkylthio group is a group that has the already-defined alkyl group as alkyl moieties. Examples include methylthio group, ethylthio group and the like.

[0067] A linear or branched C_1 - C_6 alkylsulfinyl group is a group that has the already-defined alkyl group as alkyl moieties. Examples include methylsulfinyl group, ethylsulfinyl group and the like.

[0068] A linear or branched C_1 - C_6 alkylsulfonyl group is a group that has the already-defined alkyl group as alkyl moieties. Examples include methanesulfonyl group, ethanesulfonyl group and the like.

[0069] Examples of an arylsulfonyl group include benzenesulfonyl group, 1-naphthalenesulfonyl group, 2-naphthalenesulfonyl group and the like.

[0070] A linear or branched C_1 - C_3 aralkyl group is a group that has the already-defined alkyl group as a linear or branched alkyl moiety with 1 to 3 carbon atoms. Examples include benzyl group, 1-phenethyl group, 2-phenethyl group and the like.

[0071] A linear or branched C_1 - C_3 aralkyloxy group is a group that has the already-defined aralkyl group as a linear or branched aralkyl moiety with 1 to 3 carbon atoms. Examples include benzyloxy group, 1-phenethyloxy group, 2-phenethyloxy group and the like.

[0072] Halogen atoms refer to fluorine atoms, chlorine atoms, bromine atoms, iodine atoms and the like.

[0073] A linear or branched C_1 - C_3 haloalkyl group is an alkyl group substituted with one or more of the halogen atoms defined above, which has the already-defined alkyl group as a linear or branched alkyl moiety with 1 to 3 carbon atoms. Examples of such a haloalkyl group include fluoromethyl group, chloromethyl group, bromomethyl group, difluoromethyl group, trifluoromethyl group, dichloromethyl group, trichloromethyl group, chlorodifluoromethyl group, 1,1,1-trifluoroethyl group, 1,1,1-trichloroethyl group, perfluoroethyl group, perfluoropropyl group and the like.

[0074] In addition to the abovementioned substituent group, examples of R_a and R_b include a C_1 - C_6 alkoxy C_1 - C_6 alkyl group such as methoxymethyl group, ethoxymethyl group, methoxyethyl group and the like; a C_1 - C_3 aralkyloxy C_1 - C_6 alkyl group such as benzyloxymethyl group and the like; a C_1 - C_3 aralkyl group such as benzyl group, 4-methoxybenzyl group and the like; a C_1 - C_3 aralkyloxy carbonyl group such as benzyloxy carbonyl group and the like; p-toluenesulfonyl group, and the like.

[0075] Examples of the abovementioned group $=CH-W^3$ include the group $=CH-CH_3$, the group $=CH-N(CH_3)_2$, the group $=CH-N(CH_2CH_3)_2$, the group $=CH-OCH_3$, the group $=CH-OCH_2CH_3$ and the like. These groups may be cis forms, trans forms or a mixture thereof.

[0076] Preferably, n is 1, 2, 3, 4, 5, 6, 7, 8, 9 or 10, more preferably 2 to 9, and even more preferably 2 to 6. Furthermore, in cases where n is 3 or 4, a significant separation of agonist activity and antagonist activity is recognized.

[0077] There are no particular restrictions on the deprotection process. However, examples of such processes include hydrolysis reactions performed in the presence of an acid or base, reduction reactions including hydrogenation using Pd/C or the like, dehydrogenation reactions using dichlorodicyanoquinone or the like, and other such reactions.

[0078] R^1 and R^2 may be the same or different, and these groups are preferably a hydrogen atom or a linear or branched alkyl group with 1 to 3 carbon atoms.

[0079] Salts of the compounds represented by formula (I) are pharmaceutically acceptable salts which are manufactured by contacting the abovementioned compounds with acids or bases that can be used in the manufacture of drugs. Examples of such salts include hydrochloric acid salts, hydrobromic acid salts, hydroiodic acid salts, sulfuric acid salts, sulfonic acid salts, phosphoric acid salts, phosphonic acid salts; carboxylic acid salts such as acetic acid salts, citric acid salts, malic acid salts, salicylic acid salts and the like; alkali metal salts such as sodium salts, potassium salts and the like; alkaline earth metal salts such as magnesium salts, calcium salts and the like; ammonium salts such as ammonium salts, alkylammonium salts, dialkylammonium salts, trialkylammonium salts, tetraalkylammonium salts and the like, and other such salts.

[0080] The term "a prodrug of a compound represented by formula (I)" includes a chemically modified compound that is designed to produce a compound represented by formula (I) in the body after being administered as drugs, by chemical reactions that take place in the body. Examples of such a prodrug include a compound obtained by subjecting a compound represented by formula (I) to a C_1 - C_6 alkylcarbonyl conversion, C_6 - C_{10} arylcarbonyl conversion, C_1 - C_6 alkoxy carbonyl conversion, C_1 - C_6 alkylaminocarbonyl conversion, C_1 - C_6 alkylsulfonyl conversion or the like, and a compound subjected to an imino conversion using a reagent such as N,N -dimethylformamide dimethylacetal or the like. Specific examples of a prodrug also include the compound represented by formula (III).

[0081] A solvate of a compound represented by formula (I) include a compound in which a molecule of a solvent that can be used in the manufacture of drugs is coordinated with the abovementioned compound. For example, such a solvate include a hydrate.

[0082] The compound of the present invention represented by general formula (I) is expected to act as anti-androgen agents that do not show any appearance of androgen resistance due to long-term administration, and/or side effects such as toxicity or the like, and are expected to be useful as therapeutic agents for the treatment of diseases such as prostate cancer, benign prostatic hypertrophy, male pattern baldness, sexual precociousness, common acne, seborrhea and hypertrichosis. Furthermore, if the compounds of the present invention represented by general formula (I) are administered beforehand, it is expected that the onset of diseases such as prostate cancer, benign prostatic hypertrophy, male pattern baldness, sexual precociousness, common acne, seborrhea and hypertrichosis will be prevented or delayed. Accordingly, it is expected that these compounds will also constitute prophylactic agents for such diseases.

[0083] The pharmaceutical composition of the present invention contains a compound represented by formula (I), or a salt, a prodrug or a solvate thereof, in amounts that is effective in treatment, and a pharmaceutically acceptable carrier. If necessary, this composition may contain other chemotherapeutic agents. For example, one or more agents selected from cell division inhibiting agents, alkylating agents, metabolism inhibiting agents, intercalating antibiotics, growth factor inhibiting agents, cell period inhibiting agents, enzymes, enzyme inhibitors, aromatase inhibitors, topoisomerase inhibitors, biological response modifiers, anti-hormone agents, anti-estrogen agents and anti-androgen agents.

[0084] The compound of the present invention represented by general formula (I) is expected to act as anti-androgen agents that do not show any appearance of androgen resistance due to long-term administration, and/or side effects such as toxicity or the like, and are expected to be useful as therapeutic agents for the treatment of diseases such as prostate cancer, benign prostatic hypertrophy, male pattern baldness, sexual precociousness, common acne, seborrhea and hypertrichosis. Furthermore, if the compounds of the present invention represented by general formula (I) are administered beforehand, it is expected that the onset of diseases such as prostate cancer, benign prostatic hypertrophy, male pattern baldness, sexual precociousness, common acne, seborrhea and hypertrichosis will be prevented or delayed. Accordingly, it is expected that these compounds will also constitute prophylactic agents for such diseases.

[0085] The compound of the present invention represented by general formula (I), as well as a salt, a prodrug and a solvate thereof, can be administered orally or parenterally in the form of pharmaceutical compositions which also contain pharmaceutically acceptable additive agents such as carriers, excipients, binders, diluents, stabilizing agents, lubricants, flavoring agents, disintegrating agents, coating agents, coloring agents, antioxidants, buffering agents, aqueous solvents, oily solvents, isotonic agents, dispersing agents, preservatives, solubilizing agents, fluidizing agents, analgesic agents, pH adjusting agents, antiseptic agents, base agents and the like. Examples of the abovementioned pharmaceutical composition include granular agents, powder-form agents, tablets, hard capsule agents, soft capsule agents, syrup agents, emulsions, suspensions and the like as orally administered agents. Examples of parenteral agents include injection agents such as subcutaneous injection agents, intravenous injection agents, intramuscular injection agents, intra-abdominal injection agents and the like; transdermal administration agents such as ointments, crèmes, lotions and the like; suppositories such as rectal suppositories, vaginal suppositories and the like; nasal administration formulations; and other agents. These formulations can be manufactured by universally known methods that are commonly used in formulation processes.

[0086] Examples of excipients that can be used in the present invention include sugars such as lactose, white sugar, glucose, D-mannitol, sorbit and the like; cellulose and cellulose derivatives such as crystalline cellulose, hydroxypropylcellulose, hydroxypropylmethyl cellulose, methylcellulose and the like; starch and starch derivatives such as

[0100] Examples of solubilizing agents that can be used include sodium benzoate, ethylenediamine, citric acid, sodium citrate, glycerol, sodium acetate, sodium salicylate, sorbitan sesquioleate, nicotinic acid amide, glucose, benzyl alcohol, polyvinylpyrrolidones, acetone, ethanol, isopropanol, D-sorbitol, sodium hydrogencarbonate, sodium carbonate, lactose, urea, white sugar and the like.

[0101] Examples of fluidizing agents that can be used include hydrated silicon dioxide, talc, anhydrous ethanol, crystalline cellulose, synthetic aluminum silicate, calcium hydrogenphosphate; stearic acid and salts of the same such as magnesium stearate and the like; and other agents.

[0102] Examples of analgesic agents that can be used include benzalkonium chloride, caproin hydrochloride, meprylcaine hydrochloride, lidocaine hydrochloride, lidocaine and the like.

[0103] Examples of pH adjusting agents that can be used include hydrochloric acid, citric acid, succinic acid, acetic acid, boric acid, maleic acid, sodium hydroxide and the like.

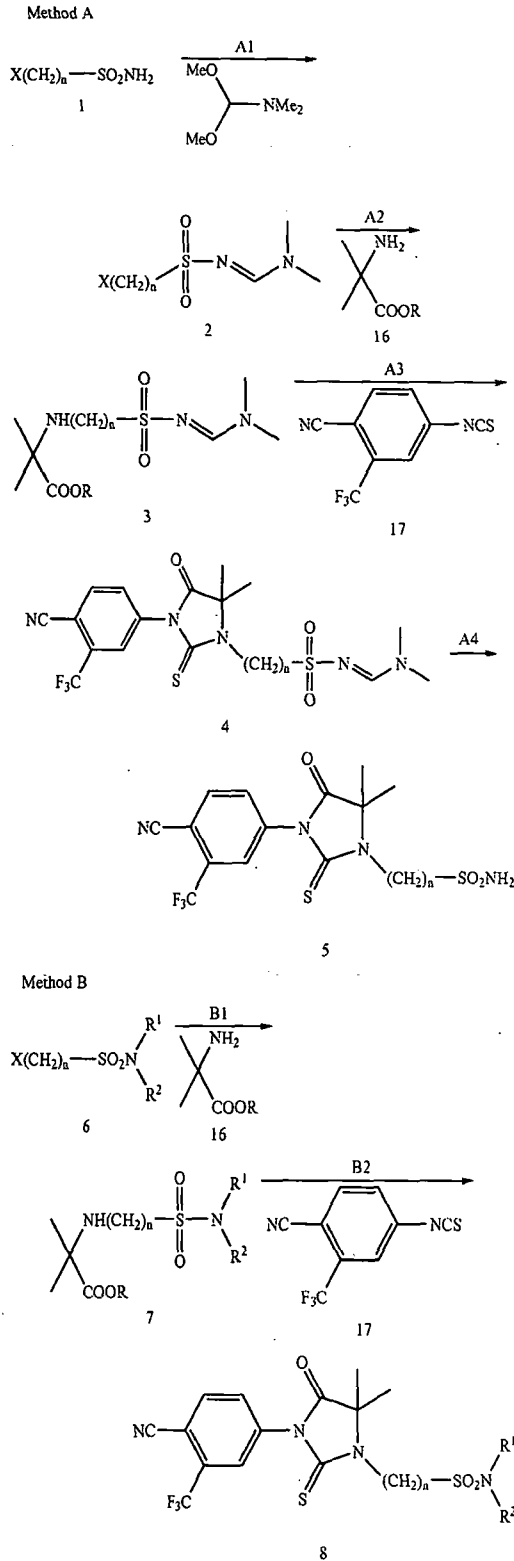
[0104] Examples of antiseptic agents that can be used include benzoic acid, sodium benzoate, cetylpyridinium chloride, salicylic acid, sodium salicylate, sorbic acid, potassium sorbate, thymol, methyl para-oxybenzoate, butyl para-oxybenzoate and the like.

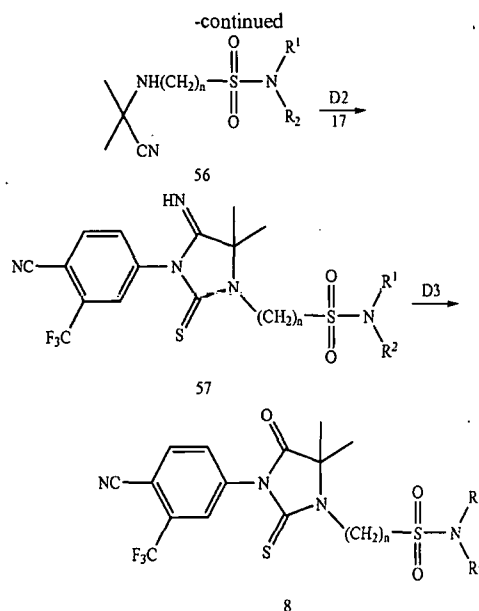
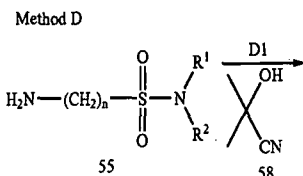
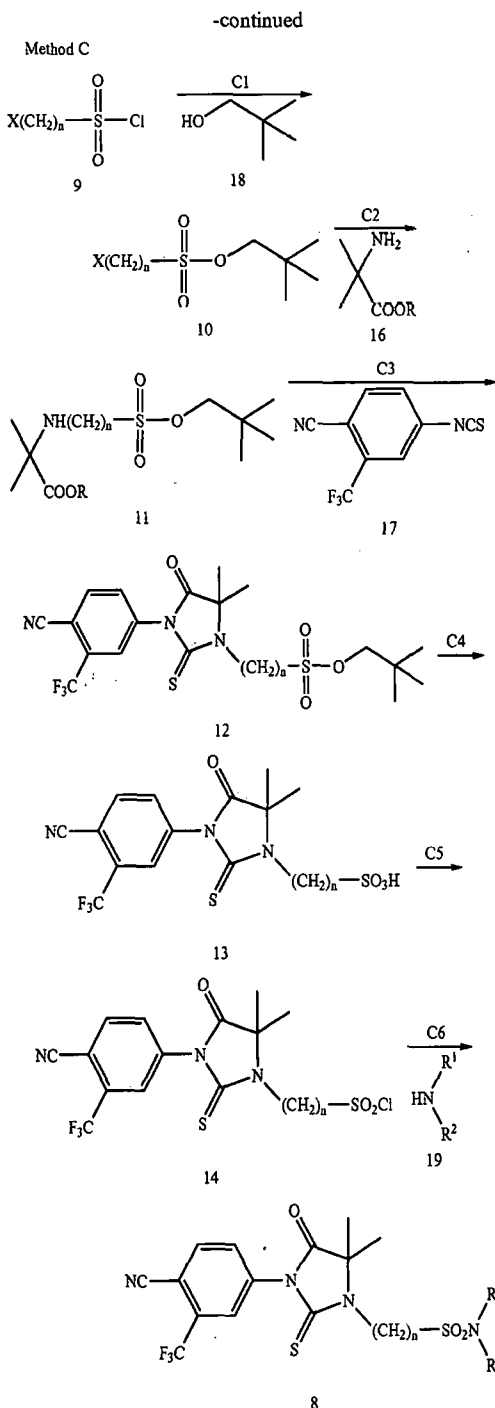
[0105] Examples of base agents that can be used include glycerol, stearyl alcohol, polyethylene glycols, propylene glycol, cetanol, lard, white Vaseline, paraffin, bentonite, lanoline fatty acid isopropyl ester, Vaseline, polysorbates, macrogols, lauryl alcohol, sodium laurylsulfate, ethyl linolate, sodium hydrogenphosphate, rosin; vegetable oils such as olive oil, sesame oil, wheat germ oil and the like; and other base agents.

[0106] The amount of compounds represented by general formula (I) in the pharmaceutical composition of the present invention varies according to the agent type, but is preferably approximately 0.1 to 100 wt % based on the total amount of the pharmaceutical composition. Furthermore, the amount of the pharmaceutical composition of the present invention that is administered may vary over a wide range depending on the subject of administration (warm-blooded animals such as humans), seriousness of the disease, age, sex, administration method, physician's diagnosis and the like. However, in regard to the amount of compounds represented by formula (I) administered to adults, it is preferable that this amount be approximately 0.1 to 500 mg/kg per day both in the case of oral administration and in the case of parenteral administration. Furthermore, the abovementioned administration amount is the value per unit weight of the object of administration. Furthermore, in the present invention, depending on the seriousness of the disease, judgment of the physician and the like, the abovementioned administration amount may be administered as one dose in a period ranging from one day to one month, or may be divided into several doses or more.

[General Procedures for Synthesis]

[0107] The compounds of the present invention represented by general formula (I) can be manufactured, e.g., according to the following methods A to D with or without modifications depending on the compounds to be manufactured.





[0108] In the chemical formulae shown in methods A to D, n , R^1 and R^2 are as defined above.

[0109] R represents a linear or branched C_1 - C_6 alkyl group. Examples include methyl group, ethyl group, n-propyl group, i-propyl group, n-butyl group, s-butyl group, i-butyl group, t-butyl group, n-pentyl group, 3-methylbutyl group, 2-methylbutyl group, 1-methylbutyl group, 1-ethylpropyl group, n-hexyl group and the like. Preferred is a linear or branched C_1 - C_3 alkyl group, and more preferred are methyl group and ethyl group.

[0110] X represents a leaving group such as a halogen atom (e.g., a chlorine atom, a bromine atom, an iodine atom), methanesulfonyloxy group, p-toluenesulfonyloxy group or the like. Preferred is a halogen atom such as a chlorine atom, a bromine atom, an iodine atom or the like.

[0111] Method A is a method for preparing compound 5, in which both R^1 and R^2 are a hydrogen atom among the compounds represented by general formula (I).

[0112] Step A1 is a step in which compound 2 is manufactured; this compound is manufactured by reacting compound 1 and compound 15 in an inert solvent.

[0113] There are no particular restrictions on the inert solvent that is used, as long as this solvent does not participate in the reaction. Examples of solvents that can be used include halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride; ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; aromatic solvents such as benzene, toluene, xylene, quinoline and chlorobenzene; and other solvents such as cyclohexane, dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like. Most suitable are dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like, and dimethylformamide and the like are especially preferable.

[0114] The reaction temperature varies depending on the type of solvent used and the like, but is ordinarily -30°C . to 100°C ., and is preferably 0°C . to 50°C .

[0115] The reaction time varies depending on the reaction temperature and the like, but is ordinarily 10 minutes to 48 hours, and is preferably 30 minutes to 24 hours.

[0116] Step A2 is a step in which compound 3 is manufactured; this is achieved by reacting compound 2 and compound 16 in the presence of a base with or without additives in an inert solvent.

[0117] There are no particular restrictions on the inert solvent used, as long as this solvent does not participate in the reaction; examples of such inert solvents include halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride; ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; aromatic solvents such as benzene, toluene, xylene, quinoline and chlorobenzene; and also cyclohexane, dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like. Especially suitable are dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like. These inert solvents may be used singly or in mixtures.

[0118] Examples of bases that can be used include carbonates such as potassium carbonate and sodium carbonate; metal hydrides such as sodium hydride, potassium hydride and calcium hydride; alkyl lithium compounds such as methyl lithium, ethyl lithium, n-butyl lithium and t-butyl lithium; metal hydroxides such as lithium hydroxide, sodium hydroxide, potassium hydroxide, potassium hydroxide, calcium hydroxide, barium hydroxide and cesium hydroxide; metal amides such as sodium amide, potassium bistrimethylsilylamide, sodium bistrimethylsilylamide and lithium diisopropylamide; amines such as triethylamine, diisopropylethylamine, 1,8-diazabicyclo[5.4.0]-7-undecene, pyridine, dimethylaminopyridine and pyrazine; and other compounds such as sodium tetraborate, sodium iodide, lithium hexamethyldisilazane, sodium hexamethyldisilazane, potassium hexamethyldisilazane and the like. Especially suitable are carbonates such as potassium carbonate and sodium carbonate.

[0119] There are no particular restrictions on additives used, as long as these additive accelerate the progress of the reaction; examples of additives that can be used include potassium iodide, sodium iodide, tetra-n-butylammonium iodide and the like.

[0120] The reaction temperature varies depending on the type of solvent used and the like, but is ordinarily 0°C . to 150°C ., and is preferably 30°C . to 100°C .

[0121] The reaction time varies depending on the reaction temperature and the like, but is ordinarily 10 minutes to 48 hours, and is preferably 30 minutes to 24 hours.

[0122] Step A3 is a step in which compound 4 is manufactured; this is achieved by reacting compound 3 and compound 17 in the presence of a base or without a base in an inert solvent.

[0123] There are no particular restrictions on the inert solvent used, as long as this solvent does not participate in the reaction. However, examples of such inert solvents

include halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride; ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; aromatic solvents such as benzene, toluene, xylene, quinoline and chlorobenzene; and also cyclohexane, dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like. Especially suitable are halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride, and ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane, and dichloromethane, tetrahydrofuran and the like are even more preferable.

[0124] Examples of bases that can be used include amines such as triethylamine, diisopropylethylamine, 1,8-diazabicyclo[5.4.0]-7-undecene, pyridine, dimethylaminopyridine and pyrazine. Preferably, the base used is triethylamine, dimethylaminopyridine or the like. Such a base may be used or omitted. However, the use of a base is preferable.

[0125] The reaction temperature varies depending on the type of solvent used and the like, but is ordinarily -30°C . to 100°C ., and is preferably 0°C . to 50°C .

[0126] The reaction time varies depending on the reaction temperature and the like, but is ordinarily 10 minutes to 48 hours, and is preferably 30 minutes to 24 hours.

[0127] Step A4 is a step in which compound 5 is manufactured; this is achieved by hydrolyzing compound 4 in an inert solvent.

[0128] There are no particular restrictions on the inert solvent that is used, as long as this solvent does not participate in the reaction. Examples of solvents that can be used include alcohol type solvents such as methanol, ethanol, n-propanol, i-propanol, n-butanol, s-butanol, t-butanol, pentanol, hexanol, cyclopropanol, cyclobutanol, cyclopentanol, cyclohexanol, ethylene glycol, 1,3-propanediol, 1,4-butanediol and 1,5-pentanediol; halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride; ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; aromatic solvents such as benzene, toluene, xylene, quinoline and chlorobenzene; and other solvents such as cyclohexane, dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like. Most suitable are alcohol type solvents such as methanol, ethanol, n-propanol, i-propanol, n-butanol, s-butanol, t-butanol, pentanol, hexanol, cyclopropanol, cyclobutanol, cyclopentanol, cyclohexanol, ethylene glycol, 1,3-propanediol, 1,4-butanediol and 1,5-pentanediol, and ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; furthermore, dioxane and the like are especially preferable.

[0129] There are no particular restrictions on the acid used. However, examples of acids that can be used include hydrochloric acid, sulfuric acid and the like. Here, hydrochloric acid and the like are especially suitable.

[0130] The reaction temperature varies depending on the type of solvent used and the like, but is ordinarily 0°C . to 200°C ., and is preferably 20°C . to 150°C .

[0131] The reaction time varies depending on the reaction temperature and the like, but is ordinarily 10 minutes to 48 hours, and is preferably 30 minutes to 24 hours.

[0132] Method B is a method for manufacturing compound 8, which is a compound represented by general formula (I) in which R¹ and R² may be the same or different, and are a hydrogen atom or a linear or branched C₁-C₆ alkyl group.

[0133] Step B1 is a step for manufacturing compound 7; this is accomplished by reacting compound 6 and compound 16 in the presence of a base, with or without additive, in an inert solvent, and is performed in the same manner as step A2 of method A.

[0134] Step B2 is a step for manufacturing compound 8; this is accomplished by reacting compound 7 and compound 17 in the presence of a base or without a base in an inert solvent, and is performed in the same manner as step A3 of method A.

[0135] Method C is another method for manufacturing compound 8, which is a compound represented by general formula (I) in which R¹ and R² may be the same or different, and are a hydrogen atom or a linear or branched C₁-C₆ alkyl group.

[0136] Step C1 is a step for manufacturing compound 10; this is accomplished by reacting compound 9 and compound 18 in the presence of a base in an inert solvent. The alcohol used in this step may be a linear or branched alkyl alcohol with 1 to 6 carbon atoms, or a linear or branched aralkyl alcohol with 1 to 3 carbon atoms or aryl alcohol. For example, methanol, ethanol, n-propanol, isopropanol, t-butanol, neopentyl alcohol (compound 18), benzyl alcohol or the like may be used.

[0137] There are no particular restrictions on the inert solvent that is used, as long as this solvent does not participate in the reaction. Examples of solvents that can be used include halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride; ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; aromatic solvents such as benzene, toluene, xylene, quinoline and chlorobenzene; and other solvents such as cyclohexane, dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like. Most suitable are halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride, and ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; and dichloromethane and the like are especially preferable.

[0138] Examples of bases that can be used include amines such as triethylamine, diisopropylethylamine, 1,8-diazabicyclo[5.4.0]-7-undecene, pyridine, dimethylaminopyridine and pyrazine. Preferably, the base used is triethylamine, dimethylaminopyridine or the like.

[0139] The reaction temperature varies depending on the type of solvent used and the like, but is ordinarily -30° C. to 100° C., and is preferably -10° C. to 30° C.

[0140] The reaction time varies depending on the reaction temperature and the like, but is ordinarily 10 minutes to 48 hours, and is preferably 30 minutes to 24 hours.

[0141] Step C2 is a step for manufacturing compound 11; this is accomplished by reacting compound 10 and compound 16 in the presence of a base, with or without additives, in an inert solvent. This step is performed in the same manner as step A2 of method A.

[0142] Step C3 is a step for manufacturing compound 12; this is accomplished by reacting compound 11 and compound 17 in the presence of a base or without a base in an inert solvent. This step is performed in the same manner as step A3 of method A.

[0143] Step C4 is a step for manufacturing compound 13; this is accomplished by reacting compound 12 with tetramethylammonium chloride or the like in an inert solvent.

[0144] There are no particular restrictions on the inert solvent that is used, as long as this solvent does not participate in the reaction. Examples of solvents that can be used include halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride; ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; aromatic solvents such as benzene, toluene, xylene, quinoline and chlorobenzene; and other solvents such as cyclohexane, dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like. Most suitable are dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like, and dimethylformamide and the like are especially preferable.

[0145] The reaction temperature varies depending on the type of solvent used and the like, but is ordinarily 30° C. to 250° C., and is preferably 80° C. to 230° C.

[0146] The reaction time varies depending on the reaction temperature and the like, but is ordinarily 10 minutes to 48 hours, and is preferably 30 minutes to 24 hours.

[0147] Step C5 is a step for manufacturing compound 14; this is accomplished by reacting a salt formed by compound 13 and a base such as triethylamine or the like with a reagent such as triphenylphosphine—thionyl chloride or the like in an inert solvent.

[0148] There are no particular restrictions on the inert solvent that is used, as long as this solvent does not participate in the reaction. Examples of solvents that can be used include halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride; ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; aromatic solvents such as benzene, toluene, xylene, quinoline and chlorobenzene; and other solvents such as cyclohexane, dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like. Most suitable are halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride, and ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; and dichloromethane and the like are especially preferable.

[0149] The reaction temperature varies depending on the type of solvent used and the like, but is ordinarily -30° C. to 50° C., and is preferably 0° C. to 30° C.

[0150] The reaction time varies depending on the reaction temperature and the like, but is ordinarily 10 minutes to 48 hours, and is preferably 30 minutes to 24 hours.

[0151] Step C6 is a step for manufacturing compound 8; this is accomplished by reacting compound 14 and compound 19 in an inert solvent.

[0152] There are no particular restrictions on the inert solvent that is used, as long as this solvent does not participate in the reaction. Examples of solvents that can be used include halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride; ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; aromatic solvents such as benzene, toluene, xylene, quinoline and chlorobenzene; and other solvents such as cyclohexane, dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like. Most suitable are halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride, and ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; and dichloromethane and the like are especially preferable.

[0153] The reaction temperature varies depending on the type of solvent used and the like, but is ordinarily -30°C . to 50°C ., and is preferably 0°C . to 30°C .

[0154] The reaction time varies depending on the reaction temperature and the like, but is ordinarily 10 minutes to 48 hours, and is preferably 30 minutes to 24 hours.

[0155] Method D is another method for manufacturing compound 8, which is a compound represented by general formula (I) in which R^1 and R^2 may be the same or different, and are a hydrogen atoms or a linear or branched C_1 - C_6 alkyl group.

[0156] Step D1 is a step for manufacturing compound 56, and is achieved by reacting compound 55 with compound 58 in the inert solvent.

[0157] There are no particular restrictions on the inert solvent that is used, as long as this solvent does not participate in the reaction. Examples of solvents that can be used include ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; alcohol type solvents such as methanol, ethanol, n-propanol, i-propanol, n-butanol, s-butanol, t-butanol, pentanol, hexanol, cyclopropanol, cyclobutanol, cyclopentanol, cyclohexanol, ethylene glycol, 1,3-propanediol, 1,4-butanediol and 1,5-pentanediol; and other solvents such as dimethylsulfoxide, dimethylacetamide and the like. Most suitable are methanol, ethanol, diethyl ether and the like, and methanol is especially preferable.

[0158] The reaction temperature varies depending on the type of solvent used and the like, but is ordinarily 0°C . to 200°C ., and is preferably 10°C . to 100°C .

[0159] The reaction time varies depending on the reaction temperature and the like, but is ordinarily 10 minutes to 48 hours, and is preferably 30 minutes to 24 hours.

[0160] Step D2 is a step for manufacturing compound 57; this is accomplished by reacting compound 56 and compound 17 in the presence of a base or without a base in an inert solvent.

[0161] There are no particular restrictions on the inert solvent used, as long as this solvent does not participate in the reaction. However, examples of such inert solvents include halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride; ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; aromatic solvents such as benzene, tolu-

ene, xylene, quinoline and chlorobenzene; and also cyclohexane, dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like. Especially suitable are halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride, and ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane, and dichloromethane, tetrahydrofuran and the like are even more preferable.

[0162] Examples of bases that can be used include amines such as triethylamine, diisopropylethylamine, 1,8-diazabicyclo[5.4.0]-7-undecene, pyridine, dimethylaminopyridine and pyrazine. Preferably, the base used is triethylamine, dimethylaminopyridine or the like. Such a base may be used or omitted. However, the use of a base is preferable.

[0163] The reaction temperature varies depending on the type of solvent used and the like, but is ordinarily -30°C . to 100°C ., and is preferably 0°C . to 50°C .

[0164] The reaction time varies depending on the reaction temperature and the like, but is ordinarily 10 minutes to 48 hours, and is preferably 30 minutes to 24 hours.

[0165] Step D3 is a step for manufacturing compound 8; this is accomplished by hydrolyzing compound 57 in an inert solvent.

[0166] There are no particular restrictions on the inert solvent that is used, as long as this solvent does not participate in the reaction. Examples of solvents that can be used include alcohol type solvents such as methanol, ethanol, n-propanol, i-propanol, n-butanol, s-butanol, t-butanol, pentanol, hexanol, cyclopropanol, cyclobutanol, cyclopentanol, cyclohexanol, ethylene glycol, 1,3-propanediol, 1,4-butanediol and 1,5-pentanediol; halogen type solvents such as dichloromethane, chloroform and carbon tetrachloride; ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; aromatic solvents such as benzene, toluene, xylene, quinoline and chlorobenzene; and other solvents such as cyclohexane, dimethylsulfoxide, dimethylacetamide, dimethylimidazolidinone, dimethylformamide, N-methylpyrrolidone, acetonitrile and the like. Most suitable are alcohol type solvents such as methanol, ethanol, n-propanol, i-propanol, n-butanol, s-butanol, t-butanol, pentanol, hexanol, cyclopropanol, cyclobutanol, cyclopentanol, cyclohexanol, ethylene glycol, 1,3-propanediol, 1,4-butanediol and 1,5-pentanediol, and ether type solvents such as diethyl ether, tetrahydrofuran, dioxane and dimethoxyethane; furthermore, dioxane and the like are especially preferable.

[0167] There are no particular restrictions on the acid used. However, examples of acids that can be used include hydrochloric acid, sulfuric acid and the like. Here, hydrochloric acid and the like are especially preferable.

[0168] The reaction temperature varies depending on the type of solvent used and the like, but is ordinarily 0°C . to 200°C ., and is preferably 20°C . to 150°C .

[0169] The reaction time varies depending on the reaction temperature and the like, but is ordinarily 10 minutes to 48 hours, and is preferably 30 minutes to 24 hours.

[0170] In cases where groups requiring protection and deprotection are present in the respective processes of the abovementioned methods A through D, these respective

groups can be subjected to protection and deprotection by method that are universally known to persons skilled in the art. For example, in such protection and deprotection, reference may be made to "Protective Groups in Organic Synthesis 2nd Edition", Theodora W. Green, John Wiley & Sons, Inc., 1991 or the like.

[0171] The above compound 1, compound 6 and compound 55, which are starting materials, are either universally known, or can easily be manufactured by universally known methods or methods similar to such universally known methods [see, e.g., The Journal of Organic Chemistry, 52(11), 2162-2166 (1987); The Journal of Organic Chemistry, 58(5), 1128-1135 (1993); Bioorganic & Medicinal Chemistry Letters, 8(13), 1607-1612 (1998)].

[0172] The abovementioned compound 9, compound 15, compound 16, compound 18 and compound 19, which are starting materials, are easily obtainable as commercially marketed products, or else are universally known or can easily be manufactured by universally known methods or methods similar to these universally known methods. Furthermore, compound 16 used in the present invention may be a salt such as a hydrochloride or the like. Hydrochloride salts are suitable for use.

[0173] The above compound 17, which is a starting material, is universally known and can easily be manufactured by universally known methods or methods similar to such universally known methods [see, e.g., The Journal of Steroid Biochemistry and Molecular Biology, 48(1), 111-119 (1994)].

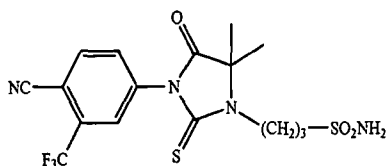
EXAMPLES

[0174] Preferred examples of the present invention will be described in detail below. However, the present invention is not limited to these examples:

[0175] NMR was measured using a nuclear magnetic resonance apparatus ARX 300 (manufactured by Bruker). Furthermore, mass analysis was performed using a mass analysis apparatus Q-micro, Triple Quadrupole Mass Spectrometer (manufactured by MICROMASS). Furthermore, Rf values in thin-layer chromatography were measured using a silica gel plate Silica gel 60 F₂₅₄ (manufactured by Merck).

Example 1

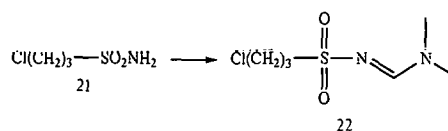
[0176] [Formula 9]



25

(First Step)

[Formula 10]



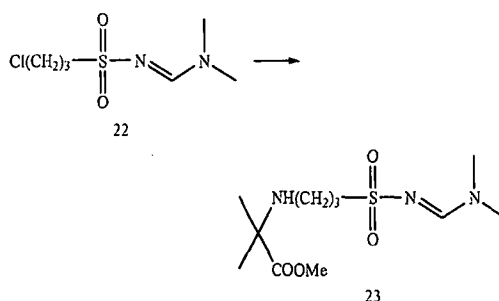
[0177] Compound 21 (4.0 g) was dissolved in N,N-dimethylformamide (20 ml). To this solution, N,N-dimethylformamide dimethylacetal (3.7 ml) was added and stirred at room temperature for 1 hour. After addition of ethyl acetate, the organic layer was washed with water and dried over magnesium sulfate. After filtration, the solvent was distilled off under reduced pressure to give the desired compound (compound 22) (3.05 g, yield 57%).

[0178] ¹H-NMR (300 MHz, CDCl₃) δ: 2.25-2.34(2H, m), 3.05(3H, s), 3.15(3H, s), 3.18(2H, t, J=7.2 Hz), 3.71(2H, t, J=6.0 Hz), 8.05(1H, s).

[0179] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=2:1): 0.31.

(Second Step)

[0180] [Formula 11]

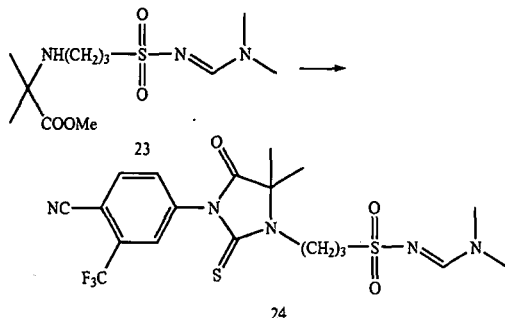


[0181] 2-Aminoisobutyric acid methyl ester hydrochloride (4.33 g) and potassium carbonate (7.8 g) were dissolved in N,N-dimethylformamide (30 ml) and stirred at room temperature for 30 minutes. To this solution, a solution of compound 22 (3.0 g) in N,N-dimethylformamide (20 ml) and sodium iodide (2.11 g) were added and stirred at 80° C. to 90° C. for 15 hours. After cooling, water was added and the reaction mixture was extracted with ethyl acetate. The organic layer was washed with water and dried over magnesium sulfate. After filtration, the solvent was distilled off under reduced pressure. The resulting residue was purified by silica gel column chromatography (NH silica, developing solvent: ethyl acetate:n-hexane=1:1 to 2:1 to 4:1) to give the desired compound (compound 23) (1.81 g, yield 44%).

[0182] ¹H-NMR (300 MHz, CDCl₃) δ: 1.29(6H, s), 1.89-1.94(2H, m), 2.57(2H, t, J=6.8 Hz), 3.04(3H, s), 3.07-3.11(2H, m), 3.13(3H, s), 3.70(3H, s), 8.03(1H, s).

[0183] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=3:1): 0.09.

(Third Step)

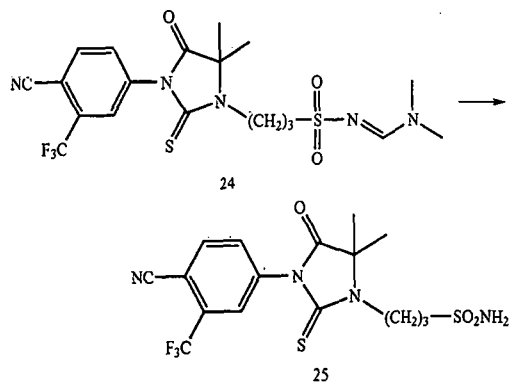


[0184] Compound 23 (2.2 g) was dissolved in tetrahydrofuran (34 ml). To this solution, triethylamine (0.21 ml) and 4-cyano-3-trifluoromethylphenyl isothiocyanate (1.71 g) were added and stirred at room temperature for 2 hours. The reaction solution was concentrated and the resulting residue was purified by silica gel column chromatography (NH silica, developing solvent: ethyl acetate:n-hexane=1:1 to 2:1 to 4:1) to give the desired compound (compound 24) (2.6 g, yield 71%).

[0185] $^1\text{H-NMR}$ (300 MHz, CDCl_3) δ : 1.62(6H, s), 2.35-2.42(2H, m), 3.06(3H, s), 3.09-3.14(2H, m), 3.15(3H, s), 3.92-3.97(2H, m), 7.77(1H, dd, $J=2.0, 8.2$ Hz), 7.90(1H, d, $J=2.0$ Hz), 7.95(1H, d, $J=8.2$ Hz), 8.07(1H, s).

[0186] Rf value (silica gel plate, developing solvent: ethyl acetate:methanol=3:1): 0.53.

(Fourth Step)



[0187] Compound 24 (2.6 g) was dissolved in 1,4-dioxane (25 ml), followed by addition of 6N-hydrochloric acid (25 ml). The resulting mixture was heated under reflux for 1 hour. After cooling, water was added and the reaction mixture was extracted with dichloromethane. The organic layer was washed with water and dried over magnesium sulfate. After filtration and concentration under reduced pressure, the resulting residue was purified by silica gel column chromatography (developing solvent: ethyl acetate:n-hexane=1:1 to 2:1 to 4:1) to give the desired compound (compound 25) (1.62 g, yield 70%).

[0188] $^1\text{H-NMR}$ (300 MHz, CDCl_3) δ : 1.62(6H, s), 2.36-2.46(2H, m), 3.28(2H, t, $J=7.1$ Hz), 3.90-3.95(2H, m), 4.85(2H, s), 7.77(1H, dd, $J=2.3, 8.4$ Hz), 7.90(1H, d, $J=2.3$ Hz), 7.97(1H, d, $J=8.4$ Hz).

[0189] Rf value (silica gel plate, developing solvent: ethyl acetate): 0.56.

[0190] MS(ESI $^-$): 433.3([M-H] $^-$).

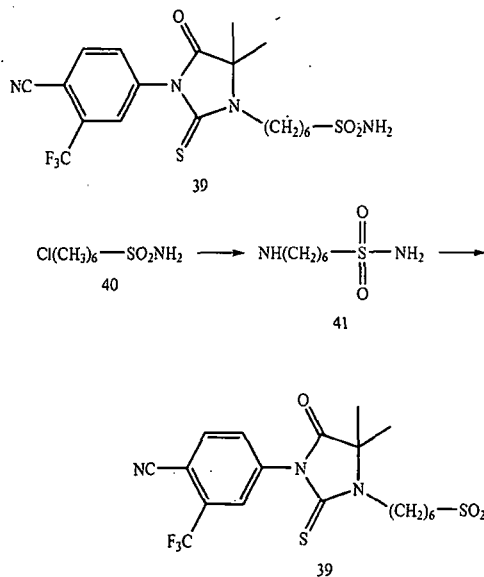
[0191] The following compound was synthesized by the same method as in Example 1.

TABLE 1

Example No.	n	Data
2	4	Rf: 0.18 (ethyl acetate:n-hexane = 2:1) MS(ESI $^-$): 447.2([M-H] $^-$)

Example 3

[0192]



[0193] 2-Aminoisobutyric acid methyl ester hydrochloride (215 mg) and potassium carbonate (406 mg) were dissolved in a mixed solvent of acetonitrile (2 ml) and dimethylformamide (0.4 ml), followed by stirring at room temperature for 1.5 hours. After addition of compound 40 (93 mg) and tetra-n-butylammonium iodide (172 mg), the reaction mixture was heated under reflux for 19 hours. After

cooling, water was added and the reaction mixture was extracted with ethyl acetate. The organic layer was washed with brine and dried over magnesium sulfate. After filtration, the solvent was distilled off under reduced pressure to give a crude product of compound 41 (94 mg). 4-Cyano-3-trifluoromethylphenyl isothiocyanate (54 mg) was dissolved in tetrahydrofuran (1 ml). To this solution, the above crude product of compound 41 (94 mg) and triethylamine (0.006 ml) were added and stirred at room temperature for 7.5 hours. The reaction solution was purified by silica gel column chromatography (developing solvent: ethyl acetate:n-hexane=1:1 to 2:1) and reversed-phase column chromatography (packing material: LiChroprep RP-18, developing solvent: methanol:water=2:3 to 1:1) to give the desired compound (compound 39) (12 mg, yield 5.4%).

[0194] $^1\text{H-NMR}$ (300 MHz, CDCl_3) δ : 1.43-1.56(4H, m), 1.61(6H, s), 1.84-1.93(4H, m), 3.11-3.16(2H, m), 3.66-3.71(2H, m), 4.68(2H, s), 7.77(1H, dd, $J=1.9, 8.5$ Hz), 7.90(1H, d, $J=1.9$ Hz), 7.95(1H, d, $J=8.5$ Hz).

[0195] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=1:1): 0.07.

[0196] MS(ESI): 477.5($[\text{M}+\text{H}]^+$).

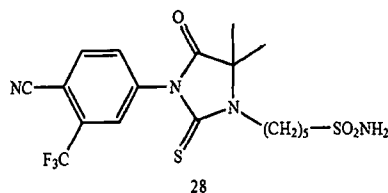
[0197] The following compounds were synthesized by the same method as in Example 3.

TABLE 2

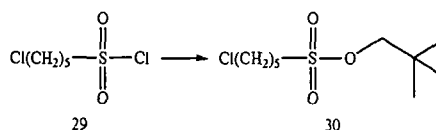
Example No.	n	Data
4	7	MS(ESI): 491.5($[\text{M}+\text{H}]^+$)
5	8	Rf: 0.20 (n-hexane:ethyl acetate = 1:2) MS(ESI): 505.6($[\text{M}+\text{H}]^+$)
6	9	Rf: 0.50 (n-hexane:ethyl acetate = 1:2) MS(ESI): 519.4($[\text{M}+\text{H}]^+$)

Example 7

[0198]



(First Step)

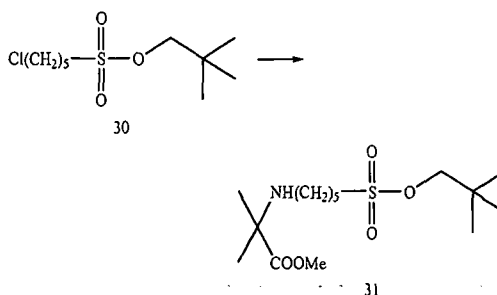


[0199] Compound 29 (2.05 g) and neopentyl alcohol (0.96 g) were dissolved in dichloromethane (20 ml) and cooled to 0° C. To this solution, triethylamine (4.6 ml) was added dropwise and stirred at 0° C. to 5° C. for 2.5 hours. The reaction solution was washed with saturated aqueous sodium hydrogencarbonate and the organic layer was dried over magnesium sulfate. After filtration and concentration under reduced pressure, the resulting residue was purified by silica gel column chromatography (developing solvent: ethyl acetate:n-hexane=1:10) to give the desired compound (compound 30) (743 mg, yield 29%).

[0200] $^1\text{H-NMR}$ (300 MHz, CDCl_3) δ : 0.99(9H, s), 1.58-1.67(2H, m), 1.78-1.96(4H, m), 3.06-3.16(2H, m), 3.55(2H, t, $J=6.5$ Hz), 3.87(2H, s).

[0201] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=1:9): 0.53.

(Second Step)

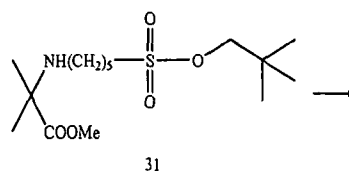


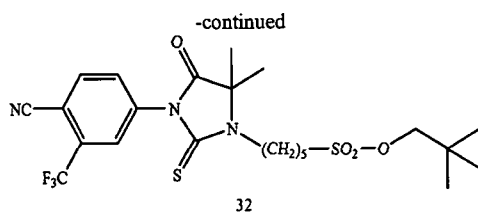
[0202] The desired compound (compound 31) (357 mg, yield 37%) was obtained from compound 30 (743 mg) by the same method as in the second step of Example 1.

[0203] $^1\text{H-NMR}$ (300 MHz, CDCl_3) δ : 0.99(9H, s), 1.30(6H, s), 1.48-1.60(4H, m), 1.85-1.91(2H, m), 2.43-2.48(2H, m), 3.07-3.12(2H, m), 3.70(3H, s), 3.86(2H, s).

[0204] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=1:1): 0.35.

(Third Step)



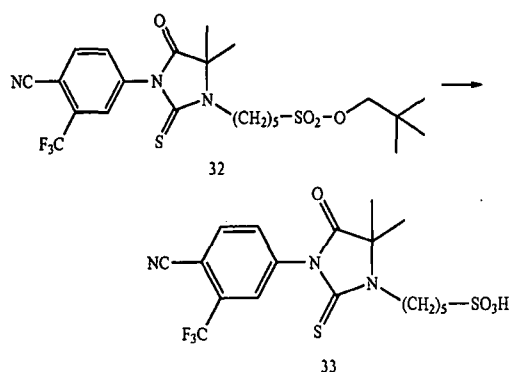


[0205] The desired compound (compound 32) (465 mg, yield 82%) was obtained from compound 31 (357 mg) by the same method as in the third step of Example 1.

[0206] $^1\text{H-NMR}$ (300 MHz, CDCl_3) δ : 0.99(9H, s), 1.59(6H, s), 1.50-1.62(2H, m), 1.87-2.00(4H, m), 3.15(2H, t, $J=7.6$ Hz), 3.67-3.73(2H, m), 3.88(2H, s), 7.77(1H, dd, $J=1.6, 8.5$ Hz), 7.89(1H, d, $J=1.6$ Hz), 7.96(1H, d, $J=8.5$ Hz).

[0207] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=1:1): 0.40.

(Fourth Step)



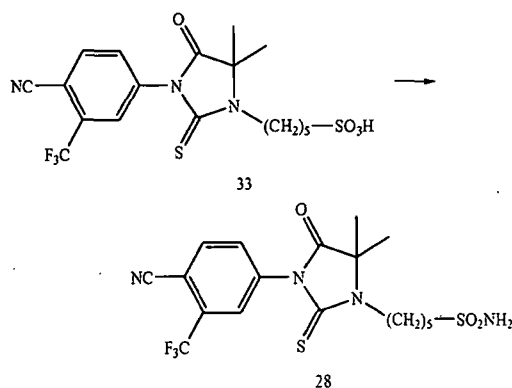
[0208] Compound 32 (460 mg) was dissolved in *N,N*-dimethylformamide, followed by addition of tetramethylammonium chloride (472 mg). The resulting mixture was heated under reflux for 6 hours. After cooling, water was added and the reaction mixture was extracted with dichloromethane. The organic layer was washed with water and brine, and then dried over magnesium sulfate. After filtration and evaporation of the solvent under reduced pressure, the resulting residue was purified by silica gel column chromatography to give the desired compound (compound 33) (220 mg, yield 55%).

[0209] $^1\text{H-NMR}$ (300 MHz, CD_3OD) δ : 1.60-1.70(2H, m), 1.72(6H, s), 1.98-2.06(4H, m), 2.97-3.02(2H, m), 3.87-3.92(2H, m), 8.05(1H, dd, $J=1.5, 8.2$ Hz), 8.21(1H, d, $J=1.5$ Hz), 8.26(1H, d, $J=8.2$ Hz).

[0210] Rf value (silica gel plate, developing solvent: ethyl acetate:methanol=3:1): 0.28.

[0211] MS(ESI): 464.5([$\text{M}+\text{H}$] $^+$).

(Fifth Step)



[0212] To compound 33 (80 mg), triethylamine (2.4 ml) was added and stirred at room temperature for 1 hour. This mixture was concentrated under reduced pressure to give a triethylammonium salt of compound 33 (86 mg). In a separate vessel, triphenylphosphine (93 mg) was dissolved in dichloromethane, followed by addition of thionyl chloride (0.0205 ml) at 0°C . To this reaction solution, the above triethylammonium salt of compound 33 (54 mg) in dichloromethane was added at 0°C and stirred at room temperature for 4 hours. To the reaction solution, a mixed solvent of pentane and diethyl ether (1:1, 5 ml) was added, and the supernatant was separated and concentrated under reduced pressure. The resulting residue was dissolved in dichloromethane and aqueous ammonia (0.5 ml) was added thereto at 0°C , followed by stirring at 0°C for 1 hour. After addition of water, the reaction mixture was extracted with dichloromethane and the organic layer was dried over magnesium sulfate. After filtration and concentration, the resulting residue was purified by thin-layer chromatography (ethyl acetate:n-hexane=1:1) to give the desired compound (compound 28) (7.6 mg).

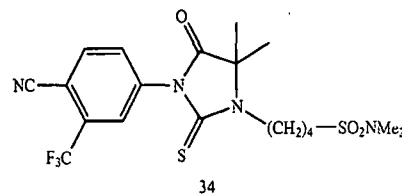
[0213] $^1\text{H-NMR}$ (300 MHz, CDCl_3) δ : 1.50-1.60(2H, m), 1.58(6H, s), 1.87-1.99(4H, m), 3.15-3.21(2H, m), 3.67-3.73(2H, m), 4.61(2H, brs), 7.77(1H, dd, $J=1.8, 8.1$ Hz), 7.89(1H, d, $J=1.8$ Hz), 7.95(1H, d, $J=8.1$ Hz).

[0214] Rf value (silica gel plate, developing solvent: ethyl acetate:methanol=1:1): 0.083.

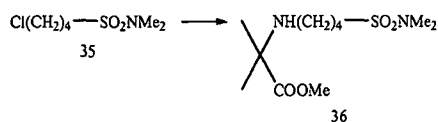
[0215] MS(ESI): 463.7([$\text{M}+\text{H}$] $^+$).

Example 8

[0216]



(First Step)

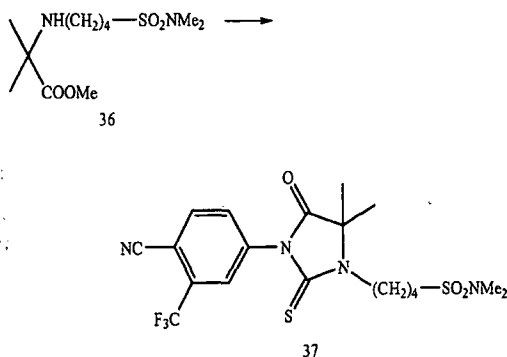


[0217] 2-Aminoisobutyric acid methyl ester hydrochloride (1.0 g) and potassium carbonate (1.8 g) were dissolved in *N,N*-dimethylformamide (5 ml). To this solution, compound 35 (350 mg) and potassium iodide (50 mg) were added and stirred at 80° C. for 36 hours. After addition of water, the reaction mixture was extracted with ethyl acetate and the organic layer was dried over magnesium sulfate. After filtration and concentration, the resulting residue was purified by silica gel column chromatography to give the desired compound (compound 36) (119 mg, yield 24%).

[0218] ¹H-NMR (300 MHz, CDCl₃) δ: 1.29(6H, s), 1.56-1.61(2H, m), 1.83-1.89(2H, m), 2.48(2H, t, J=7.1 Hz), 2.87(6H, s), 2.90-2.95(2H, m), 3.70(3H, s).

[0219] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=1:2): 0.13.

(Second Step)



[0220] Compound 36 (115 mg) was dissolved in tetrahydrofuran (3 ml). To this solution, 4-cyano-3-trifluoromethylphenyl isothiocyanate (125 mg) and triethylamine (2 drops) were added and stirred at room temperature for 3 hours. The reaction solution was concentrated and recrystallized from ethyl acetate/n-hexane (1:1) to give the desired compound (compound 37) (98 mg, yield 54%).

[0221] ¹H-NMR (300 MHz, CD₃OD) δ: 1.50(6H, s), 1.76-1.94(4H, m), 2.77(6H, s), 3.00-3.05(2H, m), 3.69-3.74(2H, m), 7.81(1H, dd, J=1.6, 8.4 Hz), 7.97(1H, d, J=1.6 Hz), 8.02(1H, d, J=8.4 Hz).

[0222] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=1:2): 0.48.

[0223] MS(ESI): 477.5([M+H]⁺).

[0224] The following compounds were synthesized by the same method as in Example 8, except that in the first steps of Examples 9 to 14, tetra-*n*-butylammonium iodide was

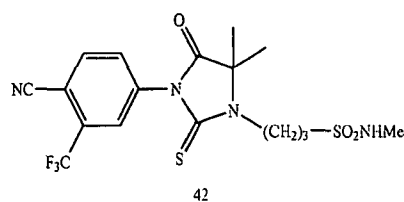
used instead of potassium iodide and a mixed solvent of acetonitrile and dimethylformamide was used as a solvent instead of dimethylformamide.

TABLE 3

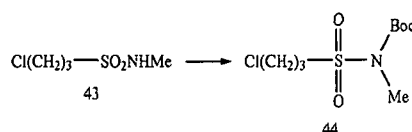
Example	n	Data
9	3	Rf: 0.77 (dichloromethane:acetone = 20:1) MS(ESI): 485.5([M+Na] ⁺)
10	5	Rf: 0.18 (ethyl acetate:n-hexane = 1:1) MS(ESI): 491.5([M+H] ⁺)
11	6	Rf: 0.22 (ethyl acetate:n-hexane = 1:1) MS(ESI): 505.5([M+H] ⁺)
12	7	Rf: 0.64 (ethyl acetate:n-hexane = 2:1) MS(ESI): 519.4([M+H] ⁺)
13	8	Rf: 0.43 (ethyl acetate:n-hexane = 2:1) MS(ESI): 555([M+Na] ⁺)
14	9	Rf: 0.76 (ethyl acetate:n-hexane = 2:1) MS(ESI): 547.5([M+H] ⁺)

Example 15

[0225]



(First Step)

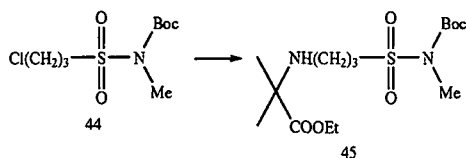


[0226] Compound 43 (1.08 g), di-*t*-butyl dicarbonate (2.06 g) and *N,N*-dimethylaminopyridine (77 mg) were dissolved in acetonitrile (12.6 ml) and stirred at room temperature for 17 hours. After addition of water, the reaction mixture was extracted with dichloromethane and the organic layer was dried over magnesium sulfate. After filtration, the solvent was distilled off under reduced pressure to give the desired compound (compound 44) (1.65 g, yield 96%).

[0227] ¹H-NMR (300 MHz, CDCl₃) δ: 1.55(9H, s), 2.23-2.32(2H, m), 3.21(3H, s), 3.62-3.69(4H, m).

[0228] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=1:2): 0.62.

(Second Step)

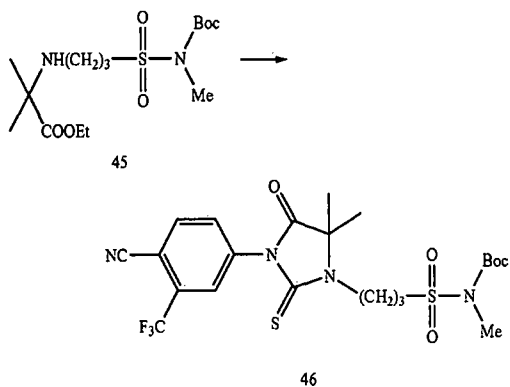


[0229] 2-Aminoisobutyric acid ethyl ester hydrochloride (592 mg) and potassium carbonate (1.02 g) were dissolved in a mixed solvent of acetonitrile (5 ml) and dimethylformamide (1 ml), followed by stirring at room temperature for 1 hour. After addition of compound 44 (800 mg) and sodium iodide (441 mg), the reaction mixture was stirred at 80° C. to 90° C. for 22 hours. After cooling, water was added and the reaction mixture was extracted with ethyl acetate. The organic layer was washed with brine and dried over magnesium sulfate. After filtration and evaporation of the solvent under reduced pressure, the resulting residue was purified by silica gel column chromatography (developing solvent: ethyl acetate:n-hexane=1:1) to give the desired compound (compound 45) (813 mg, yield 75%).

[0230] ¹H-NMR (300 MHz, CDCl₃) δ: 1.27(3H, t, J=7.1 Hz), 1.28(6H, s), 1.54(9H, s), 1.87-1.92(2H, m), 2.59(2H, t, J=6.5 Hz), 3.19(3H, s), 3.54-3.59(2H, m), 4.16(2H, q, J=7.1 Hz).

[0231] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=1:1): 0.32.

(Third Step)

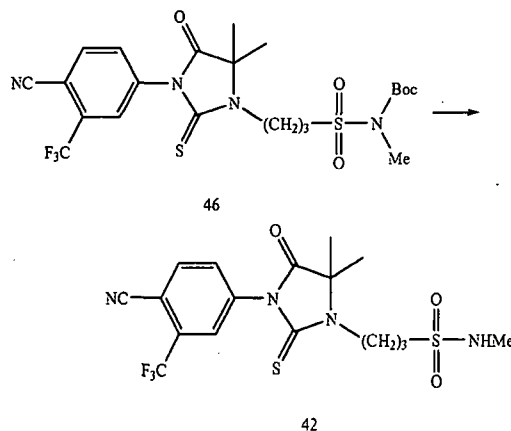


[0232] 4-Cyano-3-trifluoromethylphenyl isothiocyanate (274 mg) was dissolved in tetrahydrofuran (5.5 ml). To this solution, compound 45 (400 mg) and triethylamine (0.034 ml) were added and stirred at room temperature for 2 hours. The reaction solution was concentrated under reduced pressure and then purified by silica gel column chromatography (developing solvent: ethyl acetate:n-hexane=1:3) to give the desired compound (compound 46) (624 mg).

[0233] ¹H-NMR (300 MHz, CDCl₃) δ: 1.54(9H, s), 1.62(6H, s), 2.35-2.40(2H, m), 3.23(3H, s), 3.60(2H, t, J=7.1 Hz), 3.88-3.93(2H, m), 7.77(1H, dd, J=1.8, 8.1 Hz), 7.89(1H, d, J=1.8 Hz), 7.96(1H, d, J=8.1 Hz).

[0234] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=1:1): 0.47.

(Fourth Step)



[0235] Compound 46 (300 mg) was dissolved in dichloromethane (2.7 ml) and cooled to 0° C. To this solution, trifluoroacetic acid (0.421 ml) was added dropwise and stirred at room temperature for 5.5 hours. The reaction solution was purified by silica gel column chromatography (developing solvent: ethyl acetate:n-hexane=1:1 to ethyl acetate:n-hexane:dichloromethane=1:1:1) to give the desired compound (compound 42) (235 mg, yield 96%).

[0236] ¹H-NMR (300 MHz, CDCl₃) δ: 1.62(6H, s), 2.33-2.39(2H, m), 2.84(3H, d, J=5.2 Hz), 3.16(2H, t, J=7.1 Hz), 3.89-3.94(2H, m), 4.35(1H, q, J=5.2 Hz), 7.77(1H, dd, J=1.7, 8.4 Hz), 7.90(1H, d, J=1.7 Hz), 7.96(1H, d, J=8.4 Hz).

[0237] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=1:1): 0.18.

[0238] MS(ESI⁻): 447.1([M-H]⁻).

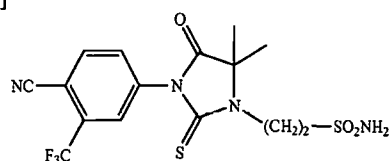
[0239] The following compounds were synthesized by the same method as in Example 15.

TABLE 4

Example No.	n	Data
16	4	Rf: 0.32 (dichloromethane:methanol = 30:1) MS(ESI): 461.0([M-H] ⁻)
17	5	Rf: 0.12 (n-hexane:ethyl acetate = 1:2) MS(ESI): 477.1([M+H] ⁺)

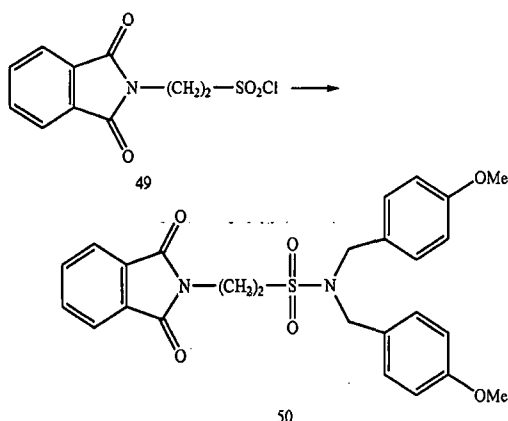
Example 18

[0240]



48

(First Step)



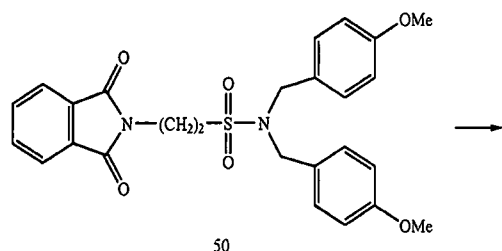
50

[0241] Bis(4-methoxybenzyl)amine (900 mg) was dissolved in dichloromethane (20 ml) and cooled to 0° C. To this solution, triethylamine (1.02 ml) was added and compound 49 (1.05 g) was added in small portions, followed by stirring at room temperature for 3 hours. After addition of water, the reaction mixture was extracted with dichloromethane. The organic layer was washed with brine and dried over magnesium sulfate. After filtration and evaporation of the solvent under reduced pressure, the resulting residue was purified by silica gel column chromatography (developing solvent: ethyl acetate:n-hexane=1:1 to ethyl acetate) to give the desired compound (compound 50) (1.4 g, yield 81%).

[0242] ¹H-NMR (300 MHz, CDCl₃) δ: 3.24(2H, t, J=6.8 Hz), 3.81(6H, s), 4.10-4.14(2H, m), 4.29(4H, s), 6.88(4H, d, J=8.7 Hz), 7.23(4H, d, J=8.7 Hz), 7.73(2H, dd, J=3.1, 5.3 Hz), 7.87(2H, dd, J=3.1, 5.3 Hz).

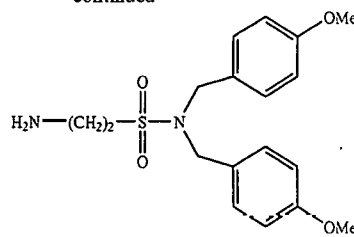
[0243] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=1:1): 0.24.

(Second Step)



50

-continued



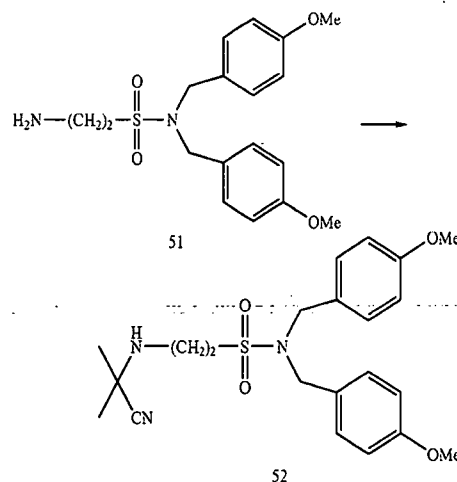
51

[0244] Compound 50 (1.4 g) was suspended in ethanol (15 ml). To this suspension, hydrazine monohydrate (0.151 ml) was added and stirred overnight at room temperature. The reaction solution was filtered and the filtrate was concentrated under reduced pressure. The resulting residue was purified by silica gel column chromatography (developing solvent: dichloromethane:methanol=100:1 to 50:1 to 20:1) to give the desired compound (compound 51) (460 mg, yield 45%).

[0245] ¹H-NMR (300 MHz, CDCl₃) δ: 2.96(2H, t, J=6.5 Hz), 3.16(2H, t, J=6.5 Hz), 3.82(6H, s), 4.27(4H, s), 6.89(4H, d, J=8.5 Hz), 7.22(4H, d, J=8.5 Hz).

[0246] Rf value (silica gel plate, developing solvent: dichloromethane:methanol=10:1): 0.41.

(Third Step)



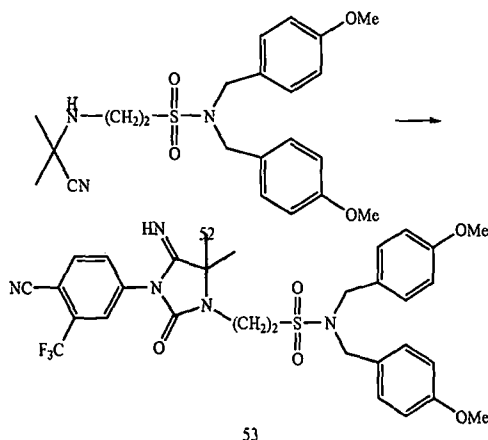
52

[0247] Compound 51 (450 mg) was dissolved in methanol (5 ml). To this solution, acetone cyanohydrin (0.136 ml) was added and stirred overnight at room temperature. After further addition of acetone cyanohydrin (0.226 ml), stirring was continued at 40° C. to 50° C. for an additional 3 hours. The reaction solution was concentrated under reduced pressure and purified by silica gel chromatography (developing solvent: dichloromethane:methanol=50:1) to give the desired compound (compound 52) (330 mg, yield 62%).

[0248] ¹H-NMR (300 MHz, CDCl₃) δ: 1.44(6H, s), 1.95(1H, brs), 3.00-3.16(4H, m), 3.82(6H, s), 4.30(4H, s), 6.89(4H, d, J=8.7 Hz), 7.23(4H, d, J=8.7 Hz).

[0249] Rf value (silica gel plate, developing solvent: dichloromethane:methanol=20:1): 0.67.

(Fourth Step)

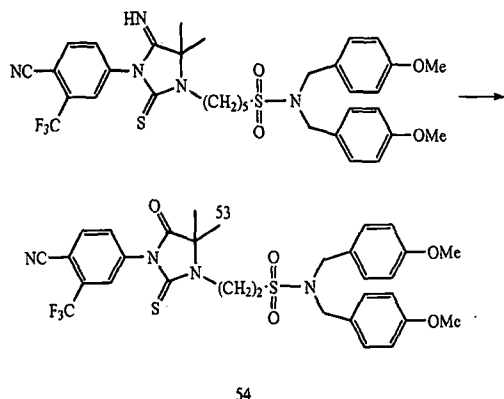


[0250] Compound 52 (220 mg) was dissolved in tetrahydrofuran (4.5 ml). To this solution, triethylamine (0.014 ml) and 4-cyano-3-trifluoromethylphenyl isothiocyanate (116 mg) were added and stirred at room temperature for 3 hours. The reaction solution was concentrated under reduced pressure and purified by silica gel column chromatography (developing solvent: dichloromethane:methanol=40:1) to give the desired compound (compound 53) (259 mg, yield 77%).

[0251] $^1\text{H-NMR}$ (300 MHz, CDCl_3) δ : 1.55(6H, s), 3.37-3.42(2H, m), 3.81(6H, s), 4.01-4.06(2H, m), 4.29(4H, s), 6.88(4H, d, $J=8.8$ Hz), 7.25(4H, d, $J=8.8$ Hz), 7.53-7.93(4H, m).

[0252] Rf value (silica gel plate, developing solvent: dichloromethane:methanol=20:1): 0.24.

(Fifth Step)



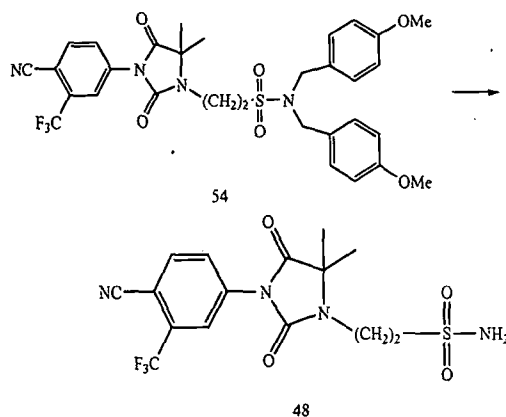
[0253] Compound 53 (259 mg) was dissolved in 1,4-dioxane (2.5 ml), followed by addition of 6N-HCl (2.5 ml). The resulting mixture was heated under reflux for 1 hour. After cooling, water was added and the reaction mixture was extracted with dichloromethane. The organic layer was washed with brine and dried over magnesium sulfate. After filtration and concentration under reduced pressure, the

resulting residue was purified by silica gel column chromatography (developing solvent: ethyl acetate:n-hexane=1:2 to 1:1) to give the desired compound (compound 54) (144 mg, yield 56%).

[0254] $^1\text{H-NMR}$ (300 MHz, CDCl_3) δ : 1.54(6H, s), 3.37-3.39(2H, m), 3.81(6H, s), 4.04-4.07(2H, m), 4.30(4H, s), 6.89(4H, d, $J=8.9$ Hz), 7.25(4H, d, $J=8.9$ Hz), 7.75(1H, dd, $J=8.5, 2.0$ Hz), 7.88(1H, d, $J=2.0$ Hz), 7.96(1H, d, $J=8.5$ Hz).

[0255] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=1:1): 0.21.

(Sixth Step)



[0256] Compound 54 (140 mg), trifluoroacetic acid (1 ml) and anisole (0.02 ml) were mixed and stirred at room temperature for 2 hours, followed by heating under reflux for 1 hour. After cooling, water was added and the reaction mixture was extracted with ethyl acetate. The organic layer was washed with brine and dried over magnesium sulfate. After filtration and concentration under reduced pressure, the resulting residue was purified by silica gel column chromatography (developing solvent: ethyl acetate:n-hexane=1:1 to 2:1 to 4:1) to give the desired compound (compound 48) (64 mg, yield 72%).

[0257] $^1\text{H-NMR}$ (300 MHz, CDCl_3) δ : 1.64(6H, s), 3.67-3.72(2H, m), 4.17-4.22(2H, m), 4.88(2H, brs), 7.76(1H, dd, $J=1.8, 8.5$ Hz), 7.88(1H, d, $J=1.8$ Hz), 7.97(1H, d, $J=8.5$ Hz).

[0258] Rf value (silica gel plate, developing solvent: ethyl acetate:n-hexane=3:1): 0.21.

[0259] MS(ESI^-): 419.1([M-H] $^-$).

Preparation of Cells Used in Test Examples

Preparation of H1A11B2 Cells

[0260] HeLa cells (purchased from Dai-Nippon Seiyaku K. K.) were cultured overnight in Dulbecco's Modified Eagle Medium containing no phenol red, but containing 3% charcoal-treated fetal bovine serum (hereafter referred to as DCC-FBS) (this medium is hereafter referred to as phenol-red-free DMEM). An MMTV-Luc-Hyg vector (reporter plasmid with Mouse tumor Long terminal repeat, containing an androgen response element and a hygromycin resistance gene: a vector obtained by substituting the chloramphenicol acetyl transferase gene of a GM-CAT vector (A.T.C.C. No. 67282) purchased from the A.T.C.C. for the firefly luciferase

gene, and inserting a hygromycin resistance gene), and pSG5-hAR-neo (human androgen receptor expression vector: a vector having an androgen receptor gene under the control of the SV40 promoter, and having a neomycin resistance gene inserted as a drug resistance gene) were transfected into the HeLa cells using an FuGENE™ 6 Transfection Reagent (obtained from Roche).

[0261] A clone in which transcription activity was elevated in a dose-dependent manner by dihydrotestosterone (DHT) was obtained by culturing the transfected cells in DMEM containing 500 µg/mL neomycin, 300 µg/mL hygromycin and 10% FBS. The clone cells thus obtained (11A11B2 cells) were maintained and propagated using DMEM containing 400 µg/mL neomycin, 200 µg/mL hygromycin and 10% FBS, and were propagated using phenol-red-free DMEM containing 10% DCC-FBS three to four days prior to the performance of an androgen receptor reporter gene assay.

Test Example 1

Investigation of Agonist Effects of Compounds of the Examples and Compounds of the Comparative Examples

[0262] The 11A11B2 cells were inoculated in a white clear-bottomed 96-well microplate (COSTAR) so that the cell concentration was 1.0×10^4 /well, and were cultured overnight using phenol-red-free DMEM containing 3% DCC-FBS (hereafter referred to as the assay medium). Samples of the assay medium containing the compounds of the examples and compounds of the comparative examples were added so that the final concentrations of the compounds of the examples and compounds of the comparative examples were 1, 10, 100, 1,000 and 10,000 nmol/L (however, in the case of the compounds of Examples 1 and 2, the compounds were added so that the final concentrations were 1, 10, 100, 1,000, 10,000 and 100,000 nmol/L), and the cells were cultured for 48 hours, after which the transcription activity value was measured. The transcription activity was measured using a Bright-Glo™ Luciferase Assay System (Promega).

[0263] The transcription activity rates of the compounds of the examples were calculated from the transcription activity measured by the abovementioned method, with the transcription activity value obtained at 0.1 nmol/L DHT taken as 100%, and the transcription activity value in the case of the assay medium alone taken as 0%. The compound concentration showing a transcription activity of 5% (EC5 value) was calculated from a linear equation for two points on either side of 5%.

Test Example 2

Investigation of Antagonist Effects of Compounds of the Examples and Compounds of the Comparative Examples

[0264] The 11A11B2 cells were inoculated in a white clear-bottomed 96-well microplate (COSTAR) so that the cell concentration was 1.0×10^4 /well, and were cultured overnight using phenol-red-free DMEM containing 3% DCC-FBS (hereafter referred to as the assay medium). The assay medium containing DHT was added so that the final concentration of DHT was 0.1 nmol/L, and samples of the assay medium containing the compounds of the examples or compounds of the comparative examples were added so that

the final concentrations of the compounds of the examples or compounds of the comparative examples were 1, 10, 100, 1,000 and 10,000 nmol/L, respectively. After culturing for 48 hours, the transcription activity values were measured. The transcription activity was measured using a Bright-Glo™ Luciferase Assay System (Promega).

[0265] The transcription activity rates of the compounds of the examples were calculated from the transcription activity measured by the abovementioned method, with the transcription activity value obtained at 0.1 nmol/L DHT taken as 100%, and the transcription activity value in the case of the assay medium alone taken as 0%.

[0266] In the present test system (Test Example 2), there were cases in which the transcription activity dropped to 50% in compounds showing both antagonist activity and agonist activity. Accordingly, the value obtained by subtracting the transcription activity rate of Test Example 1 (Investigation of Agonist Activity) from the transcription activity rate of Test Example 2 (Investigation of Antagonist Activity) was used to calculate the compound concentration at which a transcription activity of 50% was shown (IC50 value). The IC50 value was calculated from a linear equation for two points on either side of 50%.

[0267] The results of Test Examples 1 and 2 are shown in Table 1.

TABLE 1

Compound	EC5 (nM)	IC50 (nM)	EC5/IC50
Compound of Example 1	20000	200	100
Compound of Example 2	>100000	600	>170
Compound of Example 3	3000	200	15
Compound of Example 7	>10000	900	>11
Compound of Example 8	2000	100	20
Compound of Example 10	2000	200	10
Compound of Example 16	>10000	700	>14
Compound of Example 18	7000	600	12
Comparative Example 1	0.08	1	0.080
Comparative Example 2 (BP-139)	3000	800	3.8
Comparative Example 3 (bicalutamide)	20	300	0.067
Comparative Example 4 (hydroxyflutamide)	10	100	0.1

Comparative Example 1

[0268] Compound of Example 12 in Japanese Patent Application Laid-Open No. 4-308579 (4-(5-oxo-2-thioxo-3,4,4-trimethyl-1-imidazolidinyl)-2-trifluoromethylbenzotriazole)

Comparative Example 2

[0269] Compound of Example 15 in Japanese Patent Publication No. 10-510845 ((4-[3'-(2"-N-acetylaminoethyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzotriazole)

[0270] Comparative Examples 3 and 4 are universally known compounds, and can be manufactured by universally known methods.

[0271] The effect as an anti-androgen agent with reduced agonist activity can be judged by comparing the EC5/IC50 values. Specifically, compounds that have a high EC5/IC50

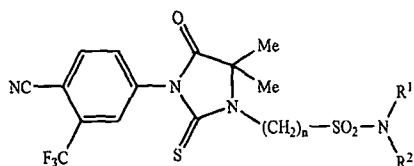
value are compounds that have a more desirable effect. In concrete terms, it is desirable that the EC50/IC50 value be 5 or greater, preferably 10 or greater, and even more preferably 20 or greater.

[0272] In Test Examples 1 and 2, it was confirmed that the compounds expressed by formula (I) of the present invention have EC50/IC50 values that are clearly higher than those of the compounds of the comparative examples.

INDUSTRIAL APPLICABILITY

[0273] It is expected that the compounds of the present invention expressed by formula (I) will act as anti-androgen agents that show no manifestation of androgen resistance due to long-term administration, and/or side effects such as liver toxicity or the like. Furthermore, it is expected that these compounds will be useful in drug compositions, e.g., therapeutic agents for diseases such as prostate cancer, benign prostatic hypertrophy, male pattern baldness, sexual precociousness, common acne, seborrhea, hypertrichosis and the like. Furthermore, it is expected that the compounds of the present invention expressed by general formula (I) will prevent or delay the onset of diseases such as prostate cancer, benign prostatic hypertrophy, male pattern baldness, sexual precociousness, common acne, seborrhea, hypertrichosis and the like, if these compounds are administered in advance. Accordingly, it is expected that these compounds will act as prophylactic agents for such diseases.

1. A compound represented by formula (I):



wherein n is an integer selected from 1 to 20, and R¹ and R², which may be the same or different, each represent a hydrogen atom or a linear or branched C₁-C₆ alkyl group, or a salt, a prodrug or a solvate thereof.

2. A compound according to claim 1 or a salt, a prodrug or a solvate thereof, wherein n is an integer selected from 1 to 10.

3. A compound according to claim 1 or a salt, a prodrug or a solvate thereof, wherein R¹ and R² are each a hydrogen atom.

4. A compound according to claim 1 to 3 or a salt, a prodrug or a solvate thereof, wherein at least one of R¹ and R² is a methyl group.

5. A compound according to claim 1, which is selected from the group consisting of:

- 4-[3'-(3"-aminosulfonylpropyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;
- 4-[3'-(4"-aminosulfonylbutyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(6"-aminosulfonylhexyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(7"-aminosulfonylheptyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(8"-aminosulfonyloctyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(9"-aminosulfonylnonyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(5"-aminosulfonylpentyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(4"-N,N-dimethylaminosulfonylbutyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(3"-N,N-dimethylaminosulfonylpropyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(5"-N,N-dimethylaminosulfonylpentyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(6"-N,N-dimethylaminosulfonylhexyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(7"-N,N-dimethylaminosulfonylheptyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(8"-N,N-dimethylaminosulfonyloctyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(9"-N,N-dimethylaminosulfonylnonyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(3"-N-methylaminosulfonylpropyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(4"-N-methylaminosulfonylbutyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile;

4-[3'-(5"-N-methylaminosulfonylpentyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile; and

4-[3'-(2"-aminosulfonylethyl)-4',4'-dimethyl-5'-oxo-2'-thioxo-1'-imidazolidinyl]-2-trifluoromethylbenzonitrile or a salt, a prodrug or a solvate thereof.

6. A drug which comprises the compound according to claim 1 or a salt, a prodrug or a solvate thereof as an active ingredient.

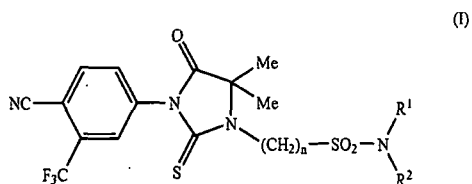
7. A pharmaceutical composition which comprises the compound according to claim 1 or a salt, a prodrug or a solvate thereof as an active ingredient.

8. An anti-androgen agent which comprises the compound according to claim 1 or a salt, a prodrug or a solvate thereof as an active ingredient.

9. A prophylactic or therapeutic agent for a disease selected from prostate cancer, benign prostatic hypertrophy, male pattern baldness, sexual precociousness, common acne, seborrhea and hypertrichosis, which comprises the compound according to claim 1 or a salt, a prodrug or a solvate thereof as an active ingredient.

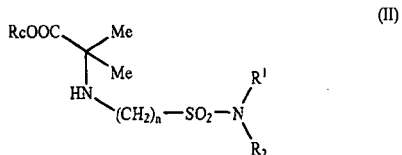
10. The use of the compound according to claim 1 or a salt, a prodrug or a solvate thereof in manufacturing a medicament used as an androgen receptor antagonist.

11. A process for preparing a compound represented by formula (I):



wherein n is an integer selected from 1 to 20, and R^1 and R^2 , which may be the same or different, each represent a hydrogen atom or a linear or branched C_1 - C_6 alkyl group, which comprises the steps of:

reacting a compound represented by formula (II):



wherein

n is an integer selected from 1 to 20;

R_a and R_b , which may be the same or different, are each selected from the group consisting of a linear or branched C_1 - C_6 alkyl group substituted with one or more W^1 , a linear or branched C_1 - C_6 alkylcarbonyl group which may be substituted with one or more W^1 , an arylcarbonyl group which may be substituted with one or more W^2 , a linear or branched C_1 - C_6 alkoxy-carbonyl group which may be substituted with one or more W^1 , an aryloxy-carbonyl group which may be substituted with one or more W^2 , a linear or branched C_1 - C_6 alkylaminocarbonyl group which may be substituted with one or more W^1 , a linear or branched C_1 - C_6 dialkylaminocarbonyl group which may be substituted with one or more W^1 , a linear or branched C_1 - C_6 alkylsulfonyl group which may be substituted with one or more W^1 , an arylsulfonyl group which may be substituted with one or more W^2 , and R^1 and R^2 ; or

R_a and R_b may be joined together to form a group $=CH-W^3$;

W^1 is a linear or branched C_1 - C_6 alkoxy group, a linear or branched C_1 - C_6 alkylthio group, a linear or branched C_1 - C_6 alkylsulfanyl group, a linear or branched C_1 - C_6 alkylsulfonyl group, an aryl group which may be sub-

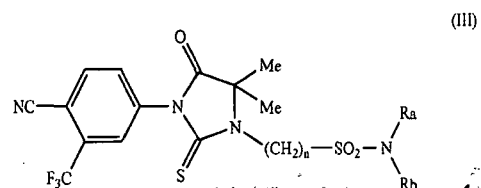
stituted with one or more W^2 , an aryloxy group which may be substituted with one or more W^2 , or a C_1 - C_3 aralkyloxy group which may be substituted with one or more W^2 ;

W^2 is a linear or branched C_1 - C_6 alkyl group, a linear or branched C_1 - C_6 alkoxy group, a linear or branched C_1 - C_6 haloalkyl group, a halogen atom, a cyano group, or a nitro group;

W^3 is a linear or branched C_1 - C_6 alkyl group, a linear or branched C_1 - C_6 alkoxy group, a linear or branched C_1 - C_6 alkylamino group, or a linear or branched C_1 - C_6 dialkylamino group;

R^1 and R^2 are as defined in claim 1; and

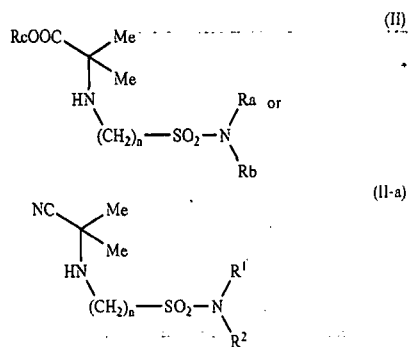
R_c is a linear or branched C_1 - C_6 alkyl group with 4-cyano-3-trifluoromethylphenyl isothiocyanate to obtain a compound represented by formula (III):



wherein n , R_a and R_b are as defined above; and

a deprotection in cases where at least one of the groups R_a and R_b is other than R^1 and R^2 .

12. A compound represented by formula (II) or (II-a):



wherein n is an integer selected from 1 to 20;

R_a and R_b , which may be the same or different, are each selected from the group consisting of a linear or branched C_1 - C_6 alkyl group substituted with one or more W^1 , a linear or branched C_1 - C_6 alkylcarbonyl group which may be substituted with one or more W^1 , an arylcarbonyl group which may be substituted with one or more W^2 , a linear or branched C_1 - C_6 alkoxy-carbonyl group which may be substituted with one or more W^1 , an aryloxy-carbonyl group which may be substituted with one or more W^2 , a linear or branched C_1 - C_6 alkylaminocarbonyl group which may be substituted with one or more W^1 , a linear or branched

C₁-C₆ dialkylaminocarbonyl group which may be substituted with one or more W¹, a linear or branched C₁-C₆ alkylsulfonyl group which may be substituted with one or more W¹, an arylsulfonyl group which may be substituted with one or more W², and R¹ and R²; or

Ra and Rb may be joined together to form a group =CH—W³;

W¹ is a linear or branched C₁-C₆ alkoxy group, a linear or branched C₁-C₆ alkylthio group, a linear or branched C₁-C₆ alkylsulfinyl group, a linear or branched C₁-C₆ alkylsulfonyl group, an aryl group which may be substituted with one or more W², an aryloxy group which may be substituted with one or more W², or a C₁-C₃ aralkyloxy group which may be substituted with one or more W²;

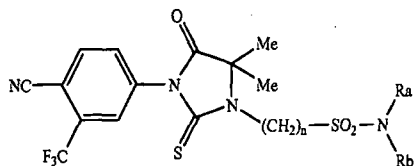
W² is a linear or branched C₁-C₆ alkyl group, a linear or branched C₁-C₆ alkoxy group, a linear or branched C₁-C₆ haloalkyl group, a halogen atom, a cyano group, or a nitro group;

W³ is a linear or branched C₁-C₆ alkyl group, a linear or branched C₁-C₆ alkoxy group, a linear or branched C₁-C₆ alkylamino group, or a linear or branched C₁-C₆ dialkylamino group;

Rc is a linear or branched C₁-C₆ alkyl group; and

R¹ and R², which may be the same or different, each represent a hydrogen atom or a linear or branched C₁-C₆ alkyl group.

13. A compound represented by formula (III):



wherein n is an integer selected from 1 to 20;

Ra and Rb, which may be the same or different, are each selected from the group consisting of a linear or branched C₁-C₆ alkyl group substituted with one or more W¹, a linear or branched C₁-C₆ alkylcarbonyl group which may be substituted with one or more W¹, an arylcarbonyl group which may be substituted with one or more W², a linear or branched C₁-C₆ alkoxy-carbonyl group which may be substituted with one or more W¹, an aryloxy-carbonyl group which may be substituted with one or more W², a linear or branched C₁-C₆ alkylaminocarbonyl group which may be substituted with one or more W¹, a linear or branched C₁-C₆ dialkylaminocarbonyl group which may be substituted with one or more W¹, a linear or branched C₁-C₆ alkylsulfonyl group which may be substituted with one or more W¹, an arylsulfonyl group which may be substituted with one or more W², and R¹ and R²; or

Ra and Rb may be joined together to form a group =CH—W³;

W¹ is a linear or branched C₁-C₆ alkoxy group, a linear or branched C₁-C₆ alkylthio group, a linear or branched C₁-C₆ alkylsulfinyl group, a linear or branched C₁-C₆ alkylsulfonyl group, an aryl group which may be substituted with one or more W², an aryloxy group which may be substituted with one or more W², or a C₁-C₃ aralkyloxy group which may be substituted with one or more W²;

W² is a linear or branched C₁-C₆ alkyl group, a linear or branched C₁-C₆ alkoxy group, a linear or branched C₁-C₆ haloalkyl group, a halogen atom, a cyano group, or a nitro group; and

W³ is a linear or branched C₁-C₆ alkyl group, a linear or branched C₁-C₆ alkoxy group, a linear or branched C₁-C₆ alkylamino group, or a linear or branched C₁-C₆ dialkylamino group.

* * * * *

DIARYLHYDANTOIN COMPOUNDS

FIELD OF THE INVENTION

[0001] The present invention relates to diarylhydantoin compounds including diarylthiohydantoins, and methods for synthesizing them and using them in the treatment of hormone refractory prostate cancer. This application claims priority from U.S. provisional applications bearing serial numbers 60/756,552, 60/750,351, and 60/680,835, the specifications of which are hereby incorporated by reference.

BACKGROUND OF THE INVENTION

[0002] Prostate cancer is the most common incidence of cancer and the second leading cause of cancer death in Western men. When the cancer is confined locally, the disease can be cured by surgery or radiation. However, 30% of such cancer relapses with distant metastatic disease and others have advanced disease at diagnoses. Advanced disease is treated by castration and/or administration of antiandrogens, the so-called androgen deprivation therapy. Castration lowers the circulating levels of androgens and reduces the activity of androgen receptor (AR). Administration of antiandrogens blocks AR function by competing away androgen binding, therefore, reducing the AR activity. Although initially effective, these treatments quickly fail and the cancer becomes hormone refractory.

[0003] Recently, overexpression of AR has been identified and validated as a cause of hormone refractory prostate cancer. See Chen, C.D., Welsbie, D.S., Tran, C., Baek, S.H., Chen, R., Vessella, R., Rosenfeld, M.G., and Sawyers, C.L., Molecular determinants of resistance to antiandrogen therapy, *Nat. Med.*, 10: 33-39, 2004, which is hereby incorporated by reference. Overexpression of AR is sufficient to cause progression from hormone sensitive to hormone refractory prostate cancer, suggesting that better AR inhibitors than the current drugs can slow the progression of prostate cancer. It was demonstrated that AR and its ligand binding are necessary for growth of hormone refractory prostate cancer, indicating that AR is still a target for this disease. It was also demonstrated that overexpression of AR converts anti-androgens from antagonists to agonists in hormone refractory prostate cancer (an AR antagonist inhibits AR activity and an AR agonist stimulates AR activity). Data from this work explains why castration and anti-androgens fail to prevent prostate cancer progression and reveals unrecognized properties of hormone refractory prostate cancer.

[0004] Bicalutamide (brand name: Casodex) is the most commonly used anti-androgen. While it has an inhibitory effect on AR in hormone sensitive prostate cancer, it fails to suppress AR when

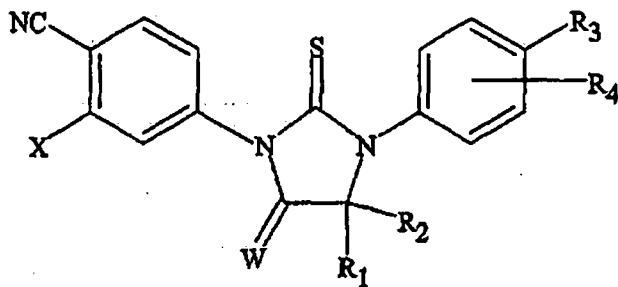
ineffective, even if it differs from the first compound only slightly, say by the replacement of a single substituent.

[0009] Identification of compounds which have high potency to antagonize the androgen activity, and which have minimal agonistic activity should overcome hormone refractory prostate cancer (HRPC) and avoid or slow down the progression of hormone sensitive prostate cancer (HSPC). Therefore, there is a need in the art for the identification of selective modulators of the androgen receptor, such as modulators which are non-steroidal, non-toxic, and tissue selective.

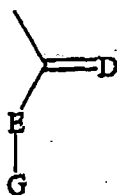
SUMMARY OF THE INVENTION

[0010] The invention provides a series of compounds having strong antagonistic activities with minimal agonistic activities against AR. These compounds inhibit the growth of hormone refractory prostate cancer.

[0011] The invention includes a compound having the formula



wherein X is selected from the group consisting of trifluoromethyl and iodo, wherein W is selected from the group consisting of O and NR₅, wherein R₅ is selected from the group consisting of H, methyl, and



wherein D is S or O and E is N or O and G is alkyl, aryl, substituted alkyl or substituted aryl; or D is S or O and E-G together are C1-C4 lower alkyl,

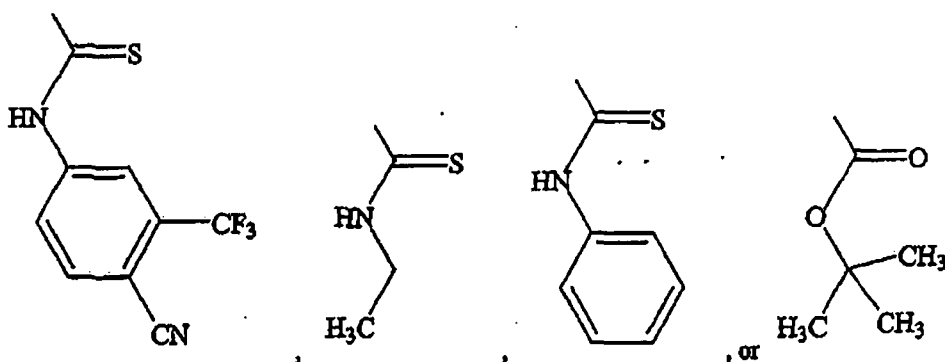
[0012] wherein R1 and R2 together comprise eight or fewer carbon atoms and are selected from the group consisting of alkyl, substituted alkyl including haloalkyl, and, together with the carbon to which they are linked, a cycloalkyl or substituted cycloalkyl group,

[0013] wherein R3 is selected from the group consisting of hydrogen, halogen, methyl, C1-C4 alkoxy, formyl, haloacetoxy, trifluoromethyl, cyano, nitro, hydroxyl, phenyl, amino, methylcarbamoyl, methoxycarbonyl, acetamido, methanesulfonamino, methanesulfonyl, 4-methanesulfonyl-1-piperazinyl, piperazinyl, and C1-C6 alkyl or alkenyl optionally substituted with hydroxyl, methoxycarbonyl, cyano, amino, amido, nitro, carbamoyl, or substituted carbamoyl including methylcarbamoyl, dimethylcarbamoyl, and hydroxyethylcarbamoyl,

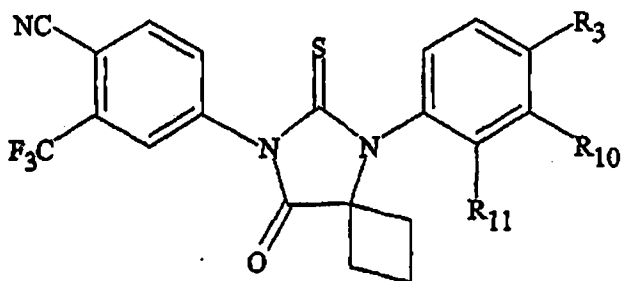
[0014] wherein R4 is selected from the group consisting of hydrogen, halogen, alkyl, and haloalkyl, and

[0015] wherein R3 is not methylaminomethyl or dimethylaminomethyl.

[0016] R5 may be



[0017] The compound may have the formula



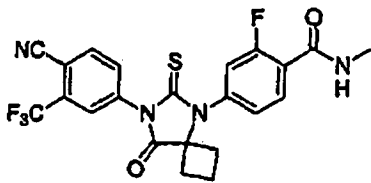
wherein R3 is selected from the group consisting of hydroxy, methylcarbonyl, methylcarbonylpropyl, methylcarbamoylethyl, methylcarbamoymethyl, methylsulfoncarbamoylethyl, methylaminomethyl, dimethylaminomethyl, methylsulfonyloxymethyl, carbamoymethyl, carbamoylethyl, carboxymethyl, methoxycarbonylmethyl, methanesulfonyl, 4-cyano-3-trifluoromethylphenylcarbamoylethyl, carboxypropyl, 4-methanesulfonyl-1-piperazinyl, piperazinyl, methoxycarbonyl, 3-cyano-4-trifluoromethylphenylcarbamoylethyl, hydroxyethylcarbamoylethyl, and hydroxyethoxycarbamoylethyl, and

[0018] wherein R10 and R11 are both H or, respectively, F and H, or H and F. In certain embodiments, R10 and R11 may both be H or, respectively, F and H. R3 may be methylcarbonyl.

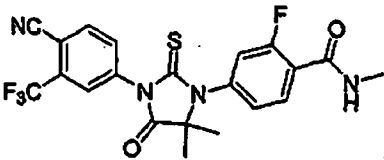
[0019] In some embodiments, R1 and R2 are independently methyl or, together with the carbon to which they are linked, a cycloalkyl group of 4 to 5 carbon atoms, and R3 is selected from the group consisting of carbonyl, alkylcarbonyl, carbonylalkyl, and alkylcarbonylalkyl, and R4 is H or F or R4 is 3-fluoro.

[0020] In other embodiments, R1 and R2 are independently methyl or, together with the carbon to which they are linked, a cycloalkyl group of 4 to 5 carbon atoms, R3 is selected from the group consisting of cyano, hydroxy, methylcarbonyl, methylcarbonyl-substituted alkyl, methylsulfoncarbonyl-substituted alkyl, methylaminomethyl, dimethylaminomethyl, methylsulfonyloxymethyl, methoxycarbonyl, acetamido, methanesulfonamido, carbonyl-substituted alkyl, carboxymethyl, methoxycarbonylmethyl, methanesulfonyl, 4-cyano-3-trifluoromethylphenylcarbonyl-substituted alkyl, carboxy-substituted alkyl, 4-(1,1-dimethylethoxy)carbonyl-1-piperazinyl, 4-methanesulfonyl-1-piperazinyl, piperazinyl, hydroxyethylcarbonyl-substituted alkyl, hydroxyethoxycarbonyl-substituted alkyl, and 3-cyano-4-trifluoromethylphenylcarbonyl, and R4 is F.

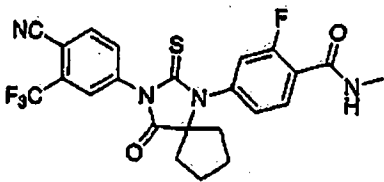
[0021] Compounds of the invention may have the formula



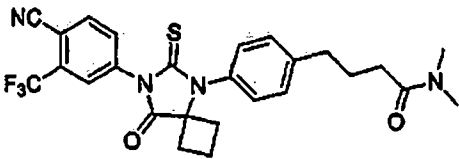
[RD162]



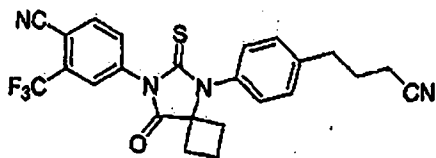
[RD162']



[RD162'']



[RD169]



[RD170]

[0025] The invention also provides a pharmaceutical composition comprising a therapeutically effective amount of a compound according to any of the preceding compounds or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier or diluent.

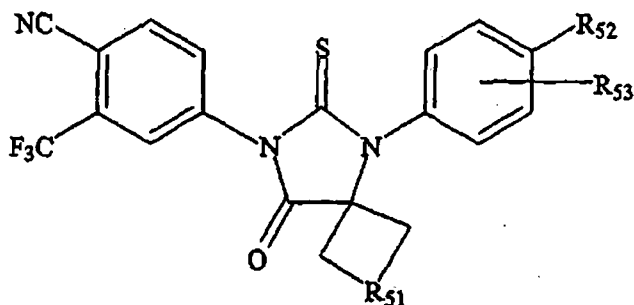
[0026] The invention encompasses a method for treating a hyperproliferative disorder

comprising administering such a pharmaceutical composition to a subject in need of such treatment, thereby treating the hyperproliferative disorder. The hyperproliferative disorder may be hormone refractory prostate cancer. The dosage may be in the range of from about 0.001 mg per kg body weight per day to about 100 mg per kg body weight per day, about 0.01 mg per kg body weight per day to about 100 mg per kg body weight per day, about 0.1 mg per kg body weight per day to about 10 mg per kg body weight per day, or about 1 mg per kg body weight per day.

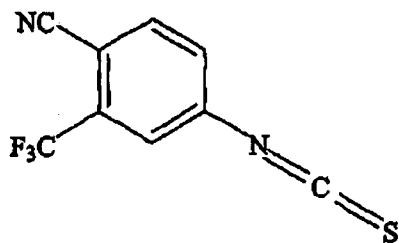
[0027] The compound may be administered by intravenous injection, by injection into tissue, intraperitoneally, orally, or nasally. The composition may have a form selected from the group consisting of a solution, dispersion, suspension, powder, capsule, tablet, pill, time release capsule, time release tablet, and time release pill.

[0028] The administered compound may be selected from the group consisting of RD162', RD162'', RD 169, or RD170, or a pharmaceutically acceptable salt thereof. The administered compound may be RD162 or a pharmaceutically acceptable salt thereof.

[0029] The invention provides a method of synthesizing a diaryl compound of formula:

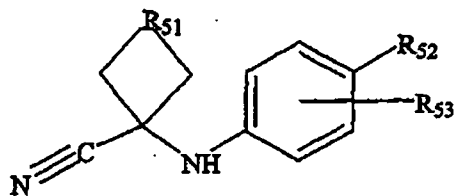


comprising mixing Compound I



Compound I

with Compound II

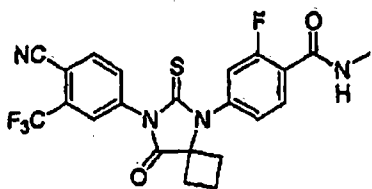


Compound II

in a first polar solvent to form a mixture, heating the mixture, adding a second polar solvent, the same as or different from the first polar solvent, and an aqueous acid to the mixture, refluxing the mixture, cooling the mixture and combining with water, and separating the diaryl compound from the mixture, wherein R51 comprises an alkyl chain of from 1 to 4 carbon atoms, R52 is selected from the group consisting of cyano, hydroxy, methylcarbamoyl, methylcarbamoyl-substituted alkyl, methylsulfonocarbamoyl-substituted alkyl, methylaminomethyl, dimethylaminomethyl, methylsulfonyloxymethyl, methoxycarbonyl, 3-cyano-4-trifluoromethylphenylcarbamoyl, carbamoyl-substituted alkyl, carboxymethyl, methoxycarbonylmethyl, methanesulfonyl, 4-cyano-3-trifluoromethylphenylcarbamoyl-substituted alkyl, carboxy-substituted alkyl, 4-methanesulfonyl-1-piperazinyl, piperazinyl, hydroxyethylcarbamoyl-substituted alkyl, and hydroxyethoxycarbonyl-substituted alkyl, and R53 is selected from the group consisting of F and H.

[0030] R51 may comprise an alkyl chain of from 1 to 2 carbon atoms, R52 may be selected from the group consisting of carbamoyl and methylcarbamoyl, and R53 may be F.

[0031] The invention provides methods of synthesizing a compound of formula:



[RD162]

comprising mixing 4-isothiocyanato-2-trifluoromethylbenzonitrile and *N*-methyl-4-(1-cyanocyclobutylamino)-2-fluorobenzamide in dimethylformamide to form a first mixture, heating the

first mixture to form a second mixture, adding alcohol and acid to the second mixture to form a third mixture, refluxing the third mixture to form a fourth mixture, cooling the fourth mixture, combining the fourth mixture with water and extracting an organic layer; isolating the compound from the organic layer.

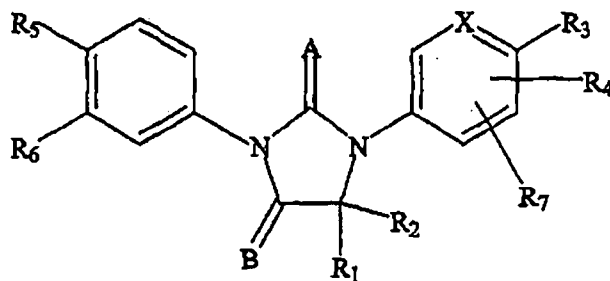
[0032] Likewise, the invention provides a method of synthesizing RD162' comprising mixing *N*-Methyl-2-fluoro-4-(1,1-dimethyl-cyanomethyl)-aminobenzamide, and 4-Isothiocyanato-2-trifluoromethylbenzonitrile in DMF and heating to form a first mixture, and processing as above.

[0033] The invention also provides a method of synthesizing RD162", comprising mixing *N*-Methyl-2-fluoro-4-(1-cyanocyclopentyl)aminobenzamide, 4-isothiocyanato-2-trifluoromethyl benzonitrile, and DMF and heating under reflux to form a first mixture, and processing as above.

[0034] The invention further provides a method of synthesizing RD169, comprising mixing *N,N*-Dimethyl 4-[4-(1-cyanocyclobutylamino)phenyl]butanamide, 4-isothiocyanato-2-trifluoromethyl benzonitrile, and DMF and heating under reflux to form a first mixture; and processing as above.

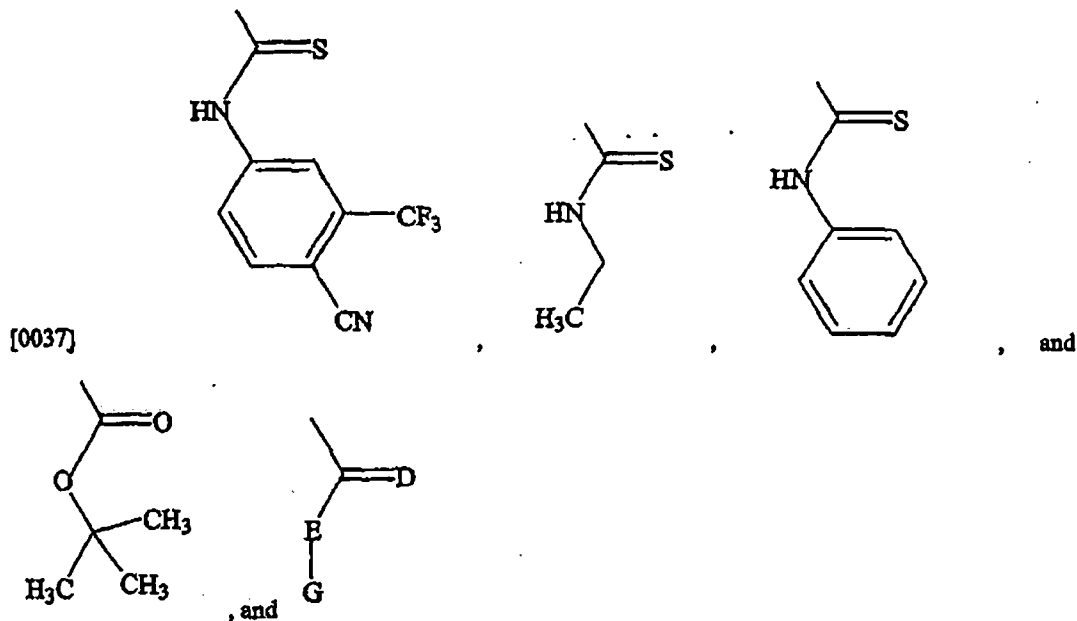
[0035] The invention provides a method of synthesizing RD170, comprising mixing DMSO, dichloromethane, and oxalyl chloride to form a first mixture, adding 4-(4-(7-(4-Cyano-3-(trifluoromethyl)phenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]octan-5-yl)phenyl)butanamide to the first mixture to form a second mixture; adding triethylamine to the second mixture to form a third mixture; warming the third mixture and quenching with aqueous NH_4Cl to form a fourth mixture; extracting an organic layer from the fourth mixture; and isolating the compound from the organic layer.

[0036] Further compounds according to the invention have the formula



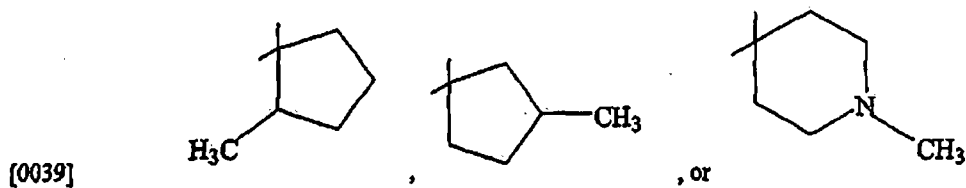
wherein R5 is CN or NO₂ or SO₂R₁₁, wherein R6 is CF₃, alkyl, substituted alkyl, alkenyl, substituted alkenyl, alkynyl, substituted alkynyl, halogenated alkyl, halogenated alkenyl, halogenated alkynyl, halogen, wherein A is sulfur (S) or oxygen (O), wherein B is O or S or NR₈, wherein R₈ is selected from the group consisting of H, methyl, aryl, substituted aryl, alkyl, substituted alkyl, alkenyl, substituted alkenyl, alkynyl, substituted alkynyl, arylalkyl, arylalkenyl, arylalkynyl, heterocyclic aromatic or non-aromatic, substituted heterocyclic aromatic or non-aromatic, cycloalkyl, substituted cycloalkyl, SO₂R₁₁,

NR₁R₂, (CO)OR₁, (CO)NR₁R₂, (CO)R₁, (CS)R₁, (CS)NR₁R₂, (CS)OR₁,



wherein D is S or O and E is N or O and G is alkyl, aryl, substituted alkyl or substituted aryl; or D is S or O and E-G together are C₁-C₄ lower alkyl,

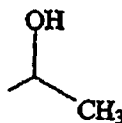
[0038] wherein R₁ and R₂ are independently alkyl, haloalkyl, hydrogen, aryl, substituted alkyl, alkenyl, substituted alkenyl, alkynyl, substituted alkynyl, halogenated alkenyl, halogenated alkynyl, arylalkyl, arylalkenyl, arylalkynyl, heterocyclic aromatic or non-aromatic, substituted heterocyclic aromatic or non-aromatic, cycloalkyl, substituted cycloalkyl, or R₁ and R₂ are connected to form a cycle which can be heterocyclic, substituted heterocyclic, cycloalkyl, substituted cycloalkyl,



[0040] wherein X is carbon or nitrogen and can be at any position in the ring, and

[0041] wherein R₃, R₄, and R₇ are independently selected from the group consisting of hydrogen, halogen, methyl, methoxy, formyl, haloacetoxy, trifluoromethyl, cyano, nitro, hydroxyl, phenyl, amino, methylcarbamoyl, methylcarbamoyl-substituted alkyl, dimethylcarbamoyl-substituted

alkyl, methoxycarbonyl, acetamido, methanesulfonamino, carbamoyl-substituted alkyl, methanesulfonyl, 4-methanesulfonyl-1-piperazinyl, piperazinyl, hydroxyethylcarbamoyl-substituted alkyl, hydroxyl-substituted alkyl, hydroxyl-substituted alkenyl, carbamoyl-substituted alkenyl, methoxycarbonyl-



substituted alkyl, cyano-substituted alkyl, , aryl, substituted aryl, alkyl, substituted alkyl, alkenyl, substituted alkenyl, alkynyl, substituted alkynyl, halogenated alkenyl, halogenated alkynyl, SO₂R₁₁, NR₁₁R₁₂, NR₁₂(CO)OR₁₁, NH(CO)NR₁₁R₁₂, NR₁₂(CO)R₁₁, O(CO)R₁₁, O(CO)OR₁₁, O(CS)R₁₁, NR₁₂(CS)R₁₁, NH(CS)NR₁₁R₁₂, NR₁₂(CS)OR₁₁, arylalkyl, arylalkenyl, arylalkynyl, heterocyclic aromatic or non-aromatic, substituted heterocyclic aromatic or non-aromatic, cycloalkyl, substituted cycloalkyl, haloalkyl, methylsulfonocarbamoyl-substituted alkyl, methylaminomethyl, dimethylaminomethyl, methylsulfonyloxymethyl, methoxycarbonyl, acetamido, methanesulfonamido, carbamoyl-substituted alkyl, carboxymethyl, methoxycarbonylmethyl, methanesulfonyl, 4-cyano-3-trifluoromethylphenylcarbamoyl-substituted alkyl, carboxy-substituted alkyl, 4-(1,1-dimethylethoxy)carbonyl)-1-piperazinyl, hydroxyethylcarbamoyl-substituted alkyl, hydroxyethoxycarbonyl-substituted alkyl, 3-cyano-4-trifluoromethylphenylcarbamoyl,

[0042] wherein R₁₁ and R₁₂ are independently hydrogen, aryl, aralkyl, substituted aralkyl, alkyl, substituted alkyl, alkenyl, substituted alkenyl, alkynyl, substituted alkynyl, halogenated alkyl, halogenated alkenyl, halogenated alkynyl, arylalkyl, arylalkenyl, arylalkynyl, heterocyclic aromatic or non-aromatic, substituted heterocyclic aromatic or non-aromatic, cycloalkyl, or substituted cycloalkyl, or R₁₁ and R₁₂ can be connected to form a cycle which can be heterocyclic aromatic or non-aromatic, substituted heterocyclic aromatic, cycloalkyl, or substituted cycloalkyl.

[0043] Such compounds have substantial androgen receptor antagonist activity and no substantial agonist activity on hormone refractory prostate cancer cells.

[0044] The invention encompasses a method comprising providing at least one such compound, measuring inhibition of androgen receptor activity for the compound and determining if the inhibition is above a first predetermined level, measuring stimulation of androgen receptor activity in hormone refractory cancer cells for the compound and determining if the stimulation is below a second predetermined level, and selecting the compound if the inhibition is above the first predetermined level and the stimulation is below the second predetermined level. The predetermined levels may be those of bicalutamide. The step of measuring inhibition may comprise measuring inhibitory concentration (IC₅₀) in an AR response reporter system or a prostate specific antigen secreting system. The step of measuring

stimulation may comprise measuring fold induction by increasing concentrations in an AR response reporter system or a prostate specific antigen secreting system. The method of measuring inhibition and/or stimulation may comprise measuring an effect of the compound on tumor growth in an animal.

BRIEF DESCRIPTION OF THE DRAWINGS

[0045] The following Figures present the results of pharmacological examination of certain compounds.

[0046] Figure 1 is a graph depicting that bicalutamide displays an agonistic effect on LNCaP-AR. Agonistic activities of bicalutamide in AR-overexpressed hormone refractory prostate cancer. LNCaP cells with overexpressed AR were treated with increasing concentrations of DMSO as vehicle or bicalutamide in the absence of R1881. Activities of AR response reporter were measured.

[0047] Figure 2 is a graph depicting an antagonistic assay of bicalutamide on LNCaP-AR. Agonistic activities of bicalutamide in hormone sensitive prostate cancer. LNCaP cells were treated with increasing concentrations of DMSO as vehicle or bicalutamide in the absence of R1881. Activities of AR response reporter were measured.

[0048] Figure 3 is a graph depicting the effect of compounds on LNCaP-AR.

[0049] Figure 4 is a graph depicting the effect of compounds on LNCaP-AR.

[0050] Figure 5 is a graph depicting the inhibition effect on LNCaP-AR.

[0051] In Figures 6-10, example 5-3b is RD7 and example 7-3b is RD37.

[0052] Figure 6. Inhibition on growth of AR-overexpressed LNCaP cells. Androgen starved LNCaP cells with overexpressed AR were treated with increasing concentrations of DMSO as vehicle or test substances in the presence of 100 pM of R1881. After 4 days of incubation, cell growth was measured by MTS assay.

[0053] Figure 7. Inhibitory effect on growth of AR-overexpressed LNCaP xenograft model. Mice with established LN-AR xenograft tumors were randomized and treated with indicated compounds orally once daily. Tumor size was measured by caliber. (A), mice were treated with 1 mg per kg of bicalutamide, example 7-3b, or vehicle for 44 days. (B), mice were treated with vehicle, 0.1, 1, or 10 mg per kg of example 7-3b for 44 days.

[0054] Figure 8. Inhibitory effect on PSA expression of AR-overexpressed LNCaP xenograft model. Mice were treated with vehicle, 0.1, 1, or 10 mg per kg of example 7-3b for 44 days orally once daily. The tumors were taken out from the mice after 44 days of treatment, tumor lysate was extracted, and PSA level in tissue lysate was determined by ELISA.

[0055] Figure 9. Inhibitory effect on growth and PSA of hormone refractory LAPC4 xenograft model. Mice with established tumors were randomized and treated with 1 mg per kg of bicalutamide, example 7-3b, or vehicle for 17 days orally once daily. (A), tumor size was measured by caliper. (B), the tumors were taken out from the mice after 17 days of treatment, tumor lysate was extracted, and PSA level in tissue lysate was determined by ELISA.

[0056] Figure 10. Inhibitory effect on growth of hormone sensitive prostate cancer cells. Androgen starved LNCaP cells were treated with increasing concentrations of DMSO as vehicle or test substances in the presence of 1 pM of R1881. After 4 days of incubation, cell growth was measured by MTS assay.

[0057] Figure 11 is a graph of tumor size. AR overexpressing LNCaP cells were injected in the flanks of castrated SCID mice, subcutaneously. When tumors reached about 100 cubic mm, they were randomized into five groups. Each group had nine animals. After they reached this tumor volume, they were given orally with either vehicle, bicalutamide or RD162 at 10 or 50 mg/kg everyday. The tumors were measured three-dimensionally, width, length and depth, using a caliper.

[0058] Figure 12 depicts experimental results of tumor size. At day 18, the animals were imaged via an optical CCD camera, 3 hours after last dose of treatment. A ROI was drawn over the tumor for luciferase activity measurement in photon/second. The right panels is a representation of the ROIs measurements.

[0059] Figure 13 is a graph depicting the pharmacokinetic curves of RD162 from intravenous (upper curve) and oral administration (lower curve).

[0060] Figure 14 is a graph depicting PSA absorbance measured for LN-AR cells after treatment with various doses of several compounds.

[0061] Figure 15 presents a table providing several characteristics of compounds. Figure 15 also presents a graph providing the pharmacokinetic characteristics of several compounds in terms of compound serum concentration as a function of time.

[0062] Figure 16 is a chart depicting prostate weight after treatment with various compounds.

10, 25, or 50 mg of compound per kilogram body weight were administered per day, as indicated by the label of a bar. The compounds were administered to healthy FVB mice. After treatment with compound for 14 days, the urogenital tract weight was determined by removing and weighing the semi-vesicles, prostate, and bladder. Three mice were administered a given compound to obtain the data presented by a bar in the chart. A set of mice was not treated with a compound: data are presented in the bar labeled "untreated". Another set of mice was treated only with vehicle solution: data are presented in the bar labeled "vehicle".

[0063] Figure 17 is a graph presenting a PSA assay performed along with the experimental protocol presented in Fig. 6.

[0064] Figure 18 is a graph presenting the effect of various dose regimens of RD162 on tumor volume.

[0065] Figure 19 is a graph presenting the rate of photon emission associated with luciferase activity at day 17 relative to the rate at day 0 after treatment with RD162 at doses of 0.1, 1, and 10 mg per kilogram body weight per day and without treatment with RD162.

[0066] Figure 20 presents the results of an experiment in which SCID mice were injected with the LN-AR (HR) cell line to induce tumor growth. One set of mice were treated with the compound RD162 at a dose of 10 mg per kilogram body weight per day; the other set of mice were treated only with vehicle solution. (A) The relative tumor volume as a function of time shown for each set of mice. (B) Images of each set of mice with photon emission associated with luciferase activity at day 31 shown as color contours. (C) Rate of photon emission associated with luciferase activity shown at several times for each set of mice.

[0067] Figure 21 is a graph presenting PSA absorbance associated with LN-AR cells treated with various concentrations of RD162, RD162', RD162", and RD170 and vehicle solution.

[0068] Figure 22 is a graph presenting PSA absorbance associated with LN-CaP cells treated with various concentrations of RD37, RD131, RD162, bicalutamide, and DMSO.

[0069] Figure 23 presents results of an experiment conducted with wild type nontransgenic mice (WT), castrated luciferase transgenic mice (Cast), and non-castrated luciferase transgenic mice (Intact). Data are shown for castrated luciferase transgenic mice treated with an implanted testosterone pellet yielding 12.5 mg per kilogram body weight with a 90 day release period (T/Cast), and data are shown for non-castrated luciferase transgenic mice treated with an implanted testosterone pellet yielding 12.5 mg per kilogram body weight with a 90 day release period (Intact+T). Data are shown for castrated

Luciferase transgenic mice treated with the implanted testosterone pellet and with bicalutamide (BIC+T/Cast) or with RD162 (RD162+T/Cast) at 10 mg per kilogram body weight per day. (A) Urogenital tract weight at 14 days. (B) Photon emission rate at 14 days. In all cases, a hormone refractory disease state was not induced.

[0070] Figure 24 is a graph of luciferase activity of the L1AR cell line dosed with various compounds administered at concentrations ranging from 125 nmol to 1000 nmol.

[0071] Figure 25 is a graph of luciferase activity for the LN/AR cell line for various compounds administered at concentrations ranging from 1.25 to 10 μ mol.

[0072] Figure 26 is a graph of luciferase activity for the 4AR cell line for various compounds administered at concentrations ranging from 1.25 to 10 μ mol.

[0073] Figure 27 is a graph of PSA levels for the 1AR cell line for various compounds administered at concentrations ranging from 1.25 to 10 μ mol.

[0074] Figure 28 is a graph of PSA levels for the LN/AR cell line for various compounds administered at concentrations ranging from 125 nmol to 1000 nmol.

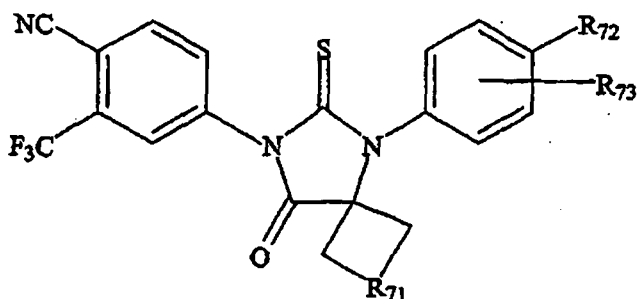
[0075] Figure 29 is a graph of luciferase activity for various compounds administered at concentrations ranging from 125 nmol to 1000 nmol.

DETAILED DESCRIPTION

[0076] Embodiments of the invention are discussed in detail below. In describing embodiments, specific terminology is employed for the sake of clarity. However, the invention is not intended to be limited to the specific terminology so selected. A person skilled in the relevant art will recognize that other equivalent parts can be employed and other methods developed without parting from the spirit and scope of the invention. All references cited herein are incorporated by reference as if each had been individually incorporated.

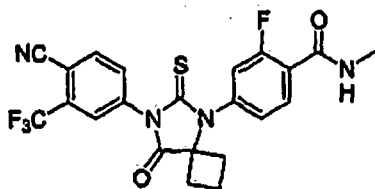
Synthesis of Diarylhydantoin Compounds

[0077] The invention provides for synthesis of diarylthiohydantoin compound having the formula



with R71 including an alkyl chain of from 1 to 4 carbon atoms. For example, R72 can be carbamoyl, e.g., $-(CO)NH_2$, or methylcarbamoyl, e.g., $-(CO)NHCH_3$. An amide group bonded at the carbon atom of the carbonyl to another structure is termed a carbamoyl substituent. For example, R73 can be a fluorine or a hydrogen atom. That is, a fluorine atom can be attached to any one of the carbons of the right-hand aryl ring which are not bonded to the R72 substituent or the nitrogen atom. Alternatively, no fluorine atom can be attached to the carbons of the right-hand aryl ring which are not bonded to the R72 substituent or the nitrogen atom. For example, a hydrogen atom can be attached to each of the carbons of the right-hand aryl ring which are not bonded to the R72 substituent or the nitrogen atom.

[0078] For example, as further presented below (see, for example, Figs. 3, 5, 11-13), the compound having the formula



[RD162]

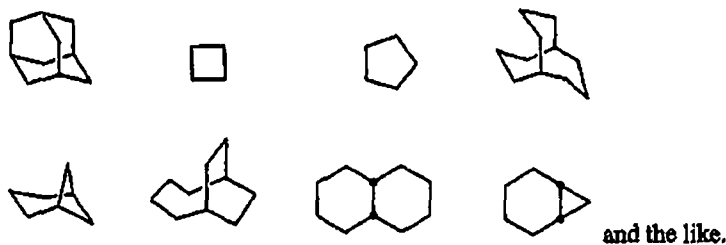
exhibited surprisingly potent antagonistic activities with minimal agonistic activities for overexpressed AR in hormone refractory prostate cancer.

[0079] A list of several compounds according to this invention is presented in Tables 5 - 11. The compounds are grouped into tiers, with Tier 1 to Tier 3 compounds being expected to be superior to bicalutamide for the treatment of prostate cancer, Tier 4 compounds being comparable to bicalutamide in effectiveness, and Tier 5 and Tier 6 compounds being worse than bicalutamide for the treatment of prostate cancer. A more detailed description of the protocol used to rank the compounds into tiers is presented below.

Definitions

[0080] As used herein, the term "alkyl" denotes branched or unbranched hydrocarbon chains, preferably having about 1 to about 8 carbons, such as, methyl, ethyl, n-propyl, iso-propyl, n-butyl, sec-butyl, iso-butyl, tert-butyl, 2-methylpentyl, pentyl, hexyl, isohexyl, heptyl, 4,4-dimethyl pentyl, octyl, 2,2,4-trimethylpentyl and the like. "Substituted alkyl" includes an alkyl group optionally substituted with one or more functional groups which may be attached to such chains, such as, hydroxyl, bromo, fluoro, chloro, iodo, mercapto or thio, cyano, alkylthio, heterocyclyl, aryl, heteroaryl, carboxyl, carbalkoyl, alkyl, alkenyl, nitro, amino, alkoxy, amido, and the like to form alkyl groups such as trifluoro methyl, 3-hydroxyhexyl, 2-carboxypropyl, 2-fluoroethyl, carboxymethyl, cyanobutyl and the like.

[0081] Unless otherwise indicated, the term "cycloalkyl" as employed herein alone or as part of another group includes saturated or partially unsaturated (containing 1 or more double bonds) cyclic hydrocarbon groups containing 1 to 3 rings, including monocycloalkyl, bicyclicalkyl and tricyclicalkyl, containing a total of 3 to 20 carbons forming the rings, preferably 3 to 10 carbons, forming the ring and which may be fused to 1 or 2 aromatic rings as described for aryl, which include cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, cyclooctyl, cyclodecyl and cyclododecyl, cyclohexenyl. "Substituted cycloalkyl" includes a cycloalkyl group optionally substituted with 1 or more substituents such as halogen, alkyl, alkoxy, hydroxy, aryl, aryloxy, arylalkyl, cycloalkyl, alkylamido, alkanoylamino, oxo, acyl, arylcarbonylamino, amino, nitro, cyano, thiol and/or alkylthio and/or any of the substituents included in the definition of "substituted alkyl." For example,



[0082] Unless otherwise indicated, the term "alkenyl" as used herein by itself or as part of another group refers to straight or branched chain radicals of 2 to 20 carbons, preferably 2 to 12 carbons, and more preferably 2 to 8 carbons in the normal chain, which include one or more double bonds in the normal chain, such as vinyl, 2-propenyl, 3-butenyl, 2-butenyl, 4-pentenyl, 3-pentenyl, 2-hexenyl, 3-

hexenyl, 2-heptenyl, 3-heptenyl, 4-heptenyl, 3-octenyl, 3-nonenyl, 4-decenyl, 3-undecenyl, 4-dodecenyl, 4,8,12-tetradecatrienyl, and the like. "Substituted alkenyl" includes an alkenyl group optionally substituted with one or more substituents, such as the substituents included above in the definition of "substituted alkyl" and "substituted cycloalkyl."

[0083] Unless otherwise indicated, the term "alkynyl" as used herein by itself or as part of another group refers to straight or branched chain radicals of 2 to 20 carbons, preferably 2 to 12 carbons and more preferably 2 to 8 carbons in the normal chain, which include one or more triple bonds in the normal chain, such as 2-propynyl, 3-butynyl, 2-butynyl, 4-pentynyl, 3-pentynyl, 2-hexynyl, 3-hexynyl, 2-heptynyl, 3-heptynyl, 4-heptynyl, 3-octynyl, 3-nonynyl, 4-decynyl, 3-undecynyl, 4-dodecynyl and the like. "Substituted alkynyl" includes an alkynyl group optionally substituted with one or more substituents, such as the substituents included above in the definition of "substituted alkyl" and "substituted cycloalkyl."

[0084] The terms "arylalkyl", "arylalkenyl" and "arylalkynyl" as used alone or as part of another group refer to alkyl, alkenyl and alkynyl groups as described above having an aryl substituent. Representative examples of arylalkyl include, but are not limited to, benzyl, 2-phenylethyl, 3-phenylpropyl, phenethyl, benzhydryl and naphthylmethyl and the like. "Substituted arylalkyl" includes arylalkyl groups wherein the aryl portion is optionally substituted with one or more substituents, such as the substituents included above in the definition of "substituted alkyl" and "substituted cycloalkyl."

[0085] The terms "arylalkyl", "arylalkenyl" and "arylalkynyl" as used alone or as part of another group refer to alkyl, alkenyl and alkynyl groups as described above having an aryl substituent. Representative examples of arylalkyl include, but are not limited to, benzyl, 2-phenylethyl, 3-phenylpropyl, phenethyl, benzhydryl and naphthylmethyl and the like. "Substituted arylalkyl" includes arylalkyl groups wherein the aryl portion is optionally substituted with one or more substituents, such as the substituents included above in the definition of "substituted alkyl" and "substituted cycloalkyl."

[0086] The term "halogen" or "halo" as used herein alone or as part of another group refers to chlorine, bromine, fluorine, and iodine.

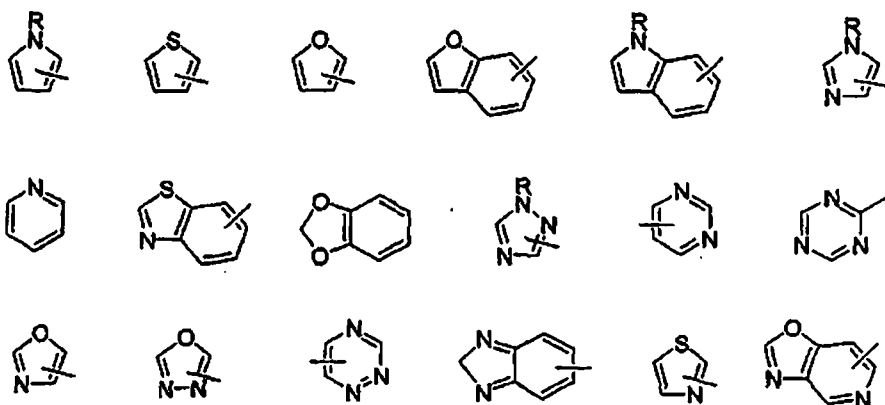
[0087] The terms "halogenated alkyl", "halogenated alkenyl" and "alkynyl" as used herein alone or as part of another group refers to "alkyl", "alkenyl" and "alkynyl" which are substituted by one or more atoms selected from fluorine, chlorine, bromine, fluorine, and iodine.

[0088] Unless otherwise indicated, the term "aryl" or "Ar" as employed herein alone or as part of another group refers to monocyclic and polycyclic aromatic groups containing 6 to 10 carbons in the ring portion (such as phenyl or naphthyl including 1-naphthyl and 2-naphthyl) and may optionally include

one to three additional rings fused to a carbocyclic ring or a heterocyclic ring (such as aryl, cycloalkyl, heteroaryl or cycloheteroalkyl rings).

[0089] "Substituted aryl" includes an aryl group optionally substituted with one or more functional groups, such as halo, haloalkyl, alkyl, haloalkyl, alkoxy, haloalkoxy, alkenyl, trifluoromethyl, trifluoromethoxy, alkynyl, cycloalkyl-alkyl, cycloheteroalkyl, cycloheteroalkylalkyl, aryl, heteroaryl, arylalkyl, aryloxy, aryloxyalkyl, arylalkoxy, alkoxycarbonyl, arylcarbonyl, arylalkenyl, aminocarbonylaryl, arylthio, arylsulfinyl, arylazo, heteroarylalkyl, heteroarylalkenyl, heteroarylheteroaryl, heteroaryloxy, hydroxy, nitro, cyano, amino, substituted amino wherein the amino includes 1 or 2 substituents (which are alkyl, aryl or any of the other aryl compounds mentioned in the definitions), thiol, alkylthio, arylthio, heteroarylthio, arylthioalkyl, alkoxyarylthio, alkylcarbonyl, arylcarbonyl, alkylaminocarbonyl, arylaminocarbonyl, alkoxycarbonyl, aminocarbonyl, alkylcarbonyloxy, arylcarbonyloxy, alkylcarbonylamino, arylcarbonylamino, arylsulfinyl, arylsulfinylalkyl, arylsulfonylamino or arylsulfonaminocarbonyl and/or any of the alkyl substituents set out herein.

[0090] Unless otherwise indicated, the term "heterocyclic" or "heterocycle", as used herein, represents an unsubstituted or substituted stable 5- to 10-membered monocyclic ring system which may be saturated or unsaturated, and which consists of carbon atoms and from one to four heteroatoms selected from N, O or S, and wherein the nitrogen and sulfur heteroatoms may optionally be oxidized, and the nitrogen heteroatom may optionally be quaternized. The heterocyclic ring may be attached at any heteroatom or carbon atom which results in the creation of a stable structure. Examples of such heterocyclic groups include, but is not limited to, piperidinyl, piperazinyl, oxopiperazinyl, oxopiperidinyl, oxopyrrolidinyl, oxoazepinyl, azepinyl, pyrrolyl, pyrrolidinyl, furanyl, thienyl, pyrazolyl, pyrazolidinyl, imidazolyl, imidazoliny, imidazolidinyl, pyridyl, pyrazinyl, pyrimidinyl, pyridazinyl, oxazolyl, oxazolidinyl, isooxazolyl, isoxazolidinyl, morpholinyl, thiazolyl, thiazolidinyl, isothiazolyl, thiadiazolyl, tetrahydropyranyl, thiamorpholinyl, thiamorpholinyl sulfoxide, thiamorpholinyl sulfone, and oxadiazolyl. The term "heterocyclic aromatic" as used here in alone or as part of another group refers to a 5- or 7-membered aromatic ring which includes 1, 2, 3 or 4 hetero atoms such as nitrogen, oxygen or sulfur and such rings fused to an aryl, cycloalkyl, heteroaryl or heterocycloalkyl ring (e.g. benzothiophenyl, indolyl), and includes possible N-oxides. "Substituted heteroaryl" includes a heteroaryl group optionally substituted with 1 to 4 substituents, such as the substituents included above in the definition of "substituted alkyl" and "substituted cycloalkyl." Examples of heteroaryl groups include the following:



and the like.

Example 1

4-isothiocyanato-2-trifluoromethylbenzonitrile, (1a)

[0091] 4-Amino-2-trifluoromethylbenzonitrile, (2.23 g, 12 mmol) was added portionwise over 15 minutes into the well-stirred heterogeneous mixture of thiophosgene (1 ml, 13 mmol) in water (22 ml) at room temperature. Stirring was continued for an additional 1 h. The reaction medium was extracted with chloroform (3 × 15 ml). The combined organic phase was dried over MgSO₄ and evaporated to dryness under reduced pressure to yield desired product, 4-isothiocyanato-2-trifluoromethylbenzonitrile, (1a), as brownish solid and was used as such for the next step (2.72 g, 11.9 mmol, 99%).

Example 2

2-1). (4-aminophenyl)carbamic acid *tert*-butyl ester, (2a)

[0092] An aqueous solution of potassium carbonate (1.52 g, 11 mmol in 5 ml of water) was added to a solution of 1,4-diaminobenzene (3.24 g, 30 mmol) in THF (30 ml) and DMF (10 ml). To this mixture was added di-*tert*-butyl pyrocarbonate, Boc₂O (2.18 g, 10 mmol), dropwise over 0.5 h. The reaction mixture was stirred for an additional 4 h at room temperature. The mixture was then poured into cold water (40 ml) and extracted with chloroform (3 × 50 ml). The combined organic phase was dried over MgSO₄ and concentrated to yield a brown residue which was subjected to flash chromatography (dichloromethane/acetone, 4:1) to afford (4-aminophenyl)carbamic acid *tert*-butyl ester, (2a) as a yellow solid (1.98 g, 9.5 mmol, 95%) (yield based on Boc₂O).

2-2). 4-[(1-cyano-1-methylethyl)amino]phenyl carbamic acid *tert*-butyl ester, 2b

[0093] The mixture of 2a (0.83 g, 4 mmol), acetone cyanohydrin (4 ml) and MgSO₄ (2 g) was

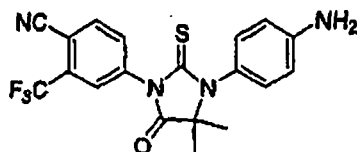
heated to 80 °C and stirred over 2.5 h. After cooling down to room temperature, compound 2b was crystallized into water (30 ml). The solid was filtered and dried to yield {4-[(1-cyano-1-methylethyl)amino]phenyl}carbamic acid *tert*-butyl ester, 2b (1.08 g, 3.9 mmol, 98%).

2-3). **{4-[3-(4-cyano-3-trifluoromethylphenyl)-4-imino-5,5-dimethyl-2-thioxoimidazolidin-1-yl]phenyl}carbamic acid *tert*-butyl ester, (2c)**

[0094] Triethylamine (0.202 g, 2 mmol) was added to a solution of 1a (0.456 g, 2 mmol) and 2b (0.57 g, 2 mmol) in dry THF (5 ml). The reaction mixture was stirred at room temperature for 15 h and then concentrated to yield a dark residue which was subjected to flash chromatography (ethyl ether/acetone, 97:3) to afford {4-[3-(4-cyano-3-trifluoromethylphenyl)-4-imino-5,5-dimethyl-2-thioxoimidazolidin-1-yl]phenyl}carbamic acid *tert*-butyl ester, (2c) (0.15 g, 0.3 mmol, 15%).

2-4). **4-[3-(4-aminophenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 2d, [RD9]**

[0095] The mixture of 2c (0.15 g, 0.3 mmol) in HCl aq, 3N. (1 ml) and methanol (4 ml) was heated to reflux for 2 h. After being cooled to room temperature, the reaction mixture was poured into cold water (5 ml) and extracted with dichloromethane (8 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane/acetone, 9:1) to yield 4-[3-(4-aminophenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 2d, [RD9] (0.118 g, 0.29 mmol, 97%) as a yellow solid.

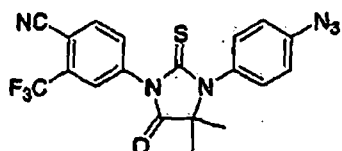


¹H NMR (400 MHz, CDCl₃) δ 1.54 (s, 6H), 6.73-6.75 (m, 2H), 7.00-7.03 (m, 2H), 8.02 (dd, *J*₁ = 8.2 Hz, *J*₂ = 1.8 Hz, 1H), 8.16 (d, *J* = 1.8 Hz, 1H), 8.20 (d, *J* = 8.2 Hz, 1H); ¹³C NMR (100 MHz, CDCl₃) δ 22.7, 66.2, 109.1, 114.3, 114.9, 120.4, 122.0 (q, *J* = 272.5 Hz), 127.0 (q, *J* = 4.9 Hz), 130.4, 132.5 (q, *J* = 33.0 Hz), 133.4, 135.6, 138.5, 149.2, 175.3, 180.4.

2-5). **4-[3-(4-azidophenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 2e, [RD10]**

[0096] An aqueous solution of sulfuric acid (25% wt, 1 ml) was added to a solution of 2d (0.10

g, 0.25 mmol) in acetone (1 ml) at -5 °C. An aqueous solution of NaNO₂ (0.024 g, 0.35 mmol, in 0.5 ml of water) was added slowly to the above mixture over 0.1 h. The reaction mixture was allowed to stir at -5 °C for an additional 1 h and then an aqueous solution of NaN₃ (0.02 g, 0.3 mmol in 0.3 ml of water) was added dropwise. Upon completion of the addition, the reaction medium was warmed to room temperature and stirred for an additional 3 h. The product was extracted with dichloromethane (3 × 5 ml). The combined organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 4-[3-(4-azidophenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, **2e**, [RD10] (0.08 g, 0.18 mmol, 72%) as a yellowish solid.



¹H NMR (400 MHz, CDCl₃) δ 1.54 (s, 6H), 7.17-7.20 (m, 2H), 7.27-7.30 (m, 2H), 7.84 (dd, *J*₁ = 8.3 Hz, *J*₂ = 1.8 Hz, 1H), 7.96 (d, *J* = 1.8 Hz, 1H), 7.97 (d, *J* = 8.3 Hz, 1H); ¹³C NMR (100 MHz, CDCl₃) δ 23.7, 66.4, 110.1, 114.8, 120.4, 122.1 (q, *J* = 272.5 Hz), 127.0 (q, *J* = 4.7 Hz), 131.1, 131.5, 132.3, 133.3 (q, *J* = 33.0 Hz), 135.3, 137.1, 141.7, 174.8, 180.1. MS for C₁₉H₁₃F₃N₆OS, calculated 430.4, found 430.1.

Example 3

3-1. 2-(4-hydroxyphenylamino)-2-methylpropanenitrile, **3a**

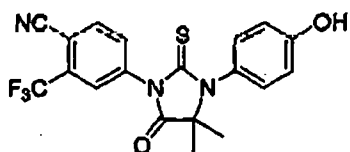
[0097] A mixture of 4-aminophenol (1.09 g, 10 mmol), acetone cyanohydrin (10 ml) and MgSO₄ (2 g) was heated to 80 °C and stirred for 4 h. After concentration of the medium under vacuum, compound **3a** was crystallized from water (20 ml). The solid was filtered and dried to yield 2-(4-hydroxyphenylamino)-2-methylpropanenitrile, **3a** (1.69 g, 9.6 mmol, 96%).

3-2. 4-[3-(4-hydroxyphenyl)-5-imino-4,4-dimethyl-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, **3b**

[0098] Triethylamine (0.101 g, 1 mmol) was added to a solution of **1a** (0.456 g, 2 mmol) and **3a** (0.352 g, 2 mmol) in dry THF (5 ml). The reaction mixture was stirred at 0 °C for 48 h and then concentrated to yield a dark residue which was subjected to flash chromatography (dichloromethane/acetone, 85:15) to afford 4-[3-(4-hydroxyphenyl)-5-imino-4,4-dimethyl-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, **3b** (0.274 g, 0.68 mmol, 34%).

3-3). **4-[3-(4-hydroxyphenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 3c, [RD8]**

A mixture of 3b (0.202 g, 0.5 mmol) in HCl aq., 2N (2 ml) and methanol (5 ml) was heated to reflux for 2 h. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (10 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane/acetone, 9:1) to yield 4-[3-(4-hydroxyphenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 3c, [RD8] (0.198 g, 0.49 mmol, 98%) as a white powder.

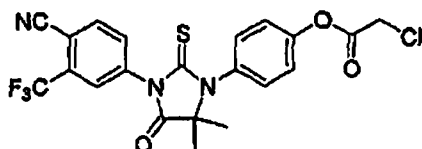


¹H NMR (CDCl₃, 400 MHz) δ 1.57 (s, 6H), 6.26 (s, OH), 6.90-6.93 (m, 2H), 7.11-7.14 (m, 2H), 7.84 (dd, J₁ = 8.3 Hz, J₂ = 1.8 Hz, 1H), 7.95-7.98 (m, 2H); ¹³C NMR (CDCl₃, 100 MHz) δ 23.6, 66.5, 109.9, 114.9, 115.7, 116.8, 121.9 (q, J = 272.7 Hz), 127.2 (q, J = 4.7 Hz), 130.6, 132.3, 133.5 (q, J = 33.2 Hz), 135.3, 137.2, 157.0, 175.3, 180.2.

Example 4

Chloroacetic acid 4-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]phenyl ester, 4a, [RD13]

Chloroacetyl chloride (0.045 g, 0.4 mmol) was added to a mixture of 3c (0.101g, 0.25 mmol) and triethylamine (0.041g, 0.41 mmol) in dry THF (1.5 ml). The mixture was stirred at room temperature for 4 h. Triethylamine hydrochloride was filtered off. The filtrate was concentrated and chromatographed (dichloromethane/acetone, 95:5) to yield 84% of Chloroacetic acid 4-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]phenyl ester, 4a, [RD13] (0.101 g, 0.21 mmol) as white powder.



¹H NMR (CDCl₃, 400 MHz) δ 1.58 (s, 6H), 4.32 (s, 2H), 7.33 (s, 4H), 7.83 (dd, J₁ = 8.3 Hz, J₂ = 1.9 Hz, 1H), 7.95-7.97 (m, 2H); ¹³C NMR (CDCl₃, 100 MHz) δ 23.7, 40.8, 66.5, 110.1, 114.8, 121.9 (q, J = 272.5 Hz), 122.7, 127.1 (q, J = 4.7 Hz), 130.9, 132.3, 132.9, 133.5 (q, J = 33.2 Hz), 135.3, 137.1, 150.9, 165.5, 174.8, 180.0.

Example 5

5-1a). 2-methyl-2-(4-methylphenyl)aminopropanenitrile, 5a

A mixture of *p*-toluidine (1.07 g, 10 mmol) and acetone cyanohydrin (10 ml) was heated to 80 °C and stirred for 4 h. The medium was concentrated and dried under vacuum to yield 2-methyl-2-(4-methylphenyl)aminopropanenitrile, 5a (1.72g, 9.9 mmol, 99%) as brown solid.

5-1b). 2-methyl-2-(4-methylphenyl)aminopropanenitrile, 5a

Sodium cyanide (0.735g, 15 mmol) was added to a mixture of *p*-toluidine (1.07 g, 10 mmol) and acetone (1.16 g, 20 mmol) in 90% acetic acid (10 ml). The reaction mixture was stirred at room temperature for 12 h and then ethyl acetate (50 ml) was added. The organic layer was washed with water (4 × 30 ml), dried over magnesium sulfate and concentrated under vacuum to dryness to yield 2-methyl-2-(4-methylphenyl)aminopropanenitrile, 5a (1.65g, 9.5 mmol, 95%) as a brown solid.

5-2). 4-[3-(4-methylphenyl)-5-imino-4,4-dimethyl-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 5b

Triethylamine (0.101 g, 1 mmol) was added to a solution of 1a (0.456 g, 2 mmol) and 5a (0.348 g, 2 mmol) in dry THF (3 ml). The reaction mixture was stirred at 0 °C for 2 days and then concentrated to yield a dark residue which was subjected to flash chromatography (dichloromethane/acetone, 95:5) to afford 4-[3-(4-methylphenyl)-5-imino-4,4-dimethyl-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 5b (0.136 g, 0.34 mmol, 17%).

5-3a). 4-[3-(4-methylphenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 5c

A mixture of 5b (0.121 g, 0.3 mmol) in HCl aq., 2N (2 ml) and methanol (5 ml) was heated to reflux for 2 h. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (10 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 4-[3-(4-methylphenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 5c (0.118 g, 0.294 mmol, 98%) as a white powder.

5-3b). 4-[3-(4-methylphenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 5c, [RD7]

A mixture of 1a (0.547 g, 2.4 mmol) and 5a (0.348 g, 2 mmol) in dry DMF (0.6 ml) was stirred for 36 h. To this mixture were added methanol (20 ml) and 2N HCl (5 ml). The second mixture was refluxed for 6

¹H NMR (CDCl₃, 400 MHz) δ 1.60 (s, 6H), 7.28-7.31 (m, 2H), 7.50-7.58 (m, 3H), 7.85 (dd, *J*₁ = 8.3 Hz, *J*₂ = 1.8 Hz, 1H), 7.96-7.99 (m, 2H); ¹³C NMR (CDCl₃, 100 MHz) δ 23.7, 66.4, 110.2, 114.8, 121.9 (q, *J* = 272.6 Hz), 127.1 (q, *J* = 4.7 Hz), 129.5, 129.8, 129.9, 132.2, 133.4 (q, *J* = 33.2 Hz), 135.1, 135.2, 137.2, 175.0, 179.9.

Example 7

7-1a). 1-(4-methylphenyl)aminocyclobutanenitrile, 7a

Sodium cyanide (0.147g, 3 mmol) was added to a mixture of *p*-toluidine (0.214 g, 2 mmol) and cyclobutanone (0.21 g, 3 mmol) in 90% acetic acid (3 ml). The reaction mixture was stirred at room temperature for 12 h and then 20 ml of ethyl acetate was added. The organic layer was washed with water (3 × 10 ml), dried over magnesium sulfate and concentrated under vacuum to dryness to yield 1-(4-methylphenyl)aminocyclobutanenitrile, 7a (0.343 g, 1.84 mmol, 92%) as a brown solid.

7-1b). 1-(4-methylphenyl)aminocyclobutanenitrile, 7a

Trimethylsilyl cyanide (0.93 ml, 7 mmol) was added dropwise to a mixture of *p*-toluidine (0.535 g, 5 mmol) and cyclobutanone (0.42 g, 6 mmol). The reaction mixture was stirred at room temperature for 6 h and then concentrated under vacuum to obtain a brown liquid which was subjected to chromatography (dichloromethane) to yield 1-(4-methylphenyl)aminocyclobutanenitrile, 7a (0.912 g, 4.9 mmol, 98%) as a yellowish solid.

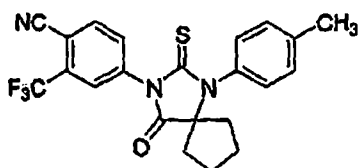
7-2). 4-(8-imino-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile, 7b

To a solution of 1a (2.28 g, 10 mmol) in dry DMF (3 ml) was added progressively, over 20 hours, a solution of 7a (1.764 g, 9 mmol) in dry DMF (3 ml) at room temperature. The medium was stirred for an additional 4 h. After DMF being evaporated, the residue was chromatographed (dichloromethane/acetone, 95:5) to afford 4-(8-imino-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile, 7b (1.937 g, 4.68 mmol, 52%).

7-3a). 4-(8-oxo-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile, 7c [RD37]

A mixture of 7b (0.041 g, 0.1 mmol) in HCl aq., 2N (3 ml) and methanol (1 ml) was heated to reflux for 2 h. After being cooled to room temperature, the reaction mixture was poured into cold water (5 ml) and extracted with ethyl acetate (6 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 4-(8-oxo-6-thioxo-5-(4-methylphenyl)-5,7-

h. After being cooled to room temperature, the reaction mixture was poured into cold water (20 ml) and extracted with ethyl acetate (30 ml). The organic layer was dried over MgSO_4 , concentrated and chromatographed (dichloromethane) to yield 4-(4-Oxo-2-thioxo-1-(4-methylphenyl)-1,3-diazaspiro[4.4]non-3-yl)-2-trifluoromethylbenzonitrile, **8b**, [RD35] (0.3 g, 0.7 mmol, 70%) as a white powder.



^1H NMR (CDCl_3 , 400 MHz) δ 1.47-1.57 (m, 2H), 1.81-1.92 (m, 2H), 2.20-2.24 (m, 2H), 2.27-2.34 (m, 2H), 2.43 (s, 3H), 7.18-7.22 (m, 2H), 7.33-7.36 (m, 2H), 7.86 (dd, $J_1 = 8.2$ Hz, $J_2 = 1.8$ Hz, 1H), 7.96 (d, $J = 8.2$ Hz, 1H), 7.98 (d, $J = 1.8$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 21.3, 25.2, 36.3, 75.1, 110.0, 114.9, 121.9 (q, $J = 272.5$ Hz), 127.1 (q, $J = 4.7$ Hz), 129.5, 130.7, 123.2, 133.0, 133.4 (q, $J = 33.2$ Hz), 135.1, 137.4, 140.0, 176.3, 180.2.

Example 9

9-1. 1-(4-methylphenyl)aminocyclohexanenitrile, **9a**

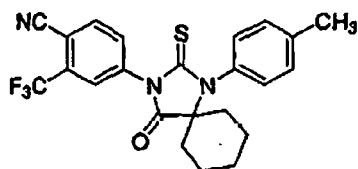
Sodium cyanide (0.147g, 3 mmol) was added to a mixture of *p*-toluidine (0.214 g, 2 mmol) and cyclohexanone (0.294 g, 3 mmol) in acetic acid 90% (3 ml). The reaction mixture was stirred at room temperature for 12 h and then 20 ml of ethyl acetate was added. The organic layer was washed with water (3 \times 10 ml), dried over magnesium sulfate and concentrated under vacuum to dryness to yield 1-(4-methylphenyl)aminocyclohexanenitrile, **9a** (0.398 g, 1.86 mmol, 93%) as a brown solid.

9-2. 4-(4-imino-2-thioxo-1-(4-methylphenyl)-1,3-diazaspiro[4.5]dec-3-yl)-2-trifluoromethylbenzonitrile, **9b**

Triethylamine (0.05 g, 0.5 mmol) was added to a solution of **1a** (0.228 g, 1 mmol) and **9a** (0.214 g, 1 mmol) in dry THF (2 ml). The reaction mixture was stirred at room temperature for 2 days and then concentrated to yield a dark residue which was subjected to flash chromatography (dichloromethane/acetone, 95:5) to afford 4-(4-imino-2-thioxo-1-(4-methylphenyl)-1,3-diazaspiro[4.5]dec-3-yl)-2-trifluoromethylbenzonitrile, **9b** (0.035 g, 0.08 mmol, 8%).

9-3. 4-(4-Oxo-2-thioxo-1-(4-methylphenyl)-1,3-diazaspiro[4.5]dec-3-yl)-2-trifluoromethylbenzonitrile, **9c**, [RD48]

A mixture of 9b (0.035 g, 0.08 mmol) in HCl aq., 2N (1 ml) and methanol (3 ml) was heated to reflux for 2 h. After being cooled to room temperature, the reaction mixture was poured into cold water (5 ml) and extracted with ethyl acetate (6 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 4-(4-Oxo-2-thioxo-1-(4-methylphenyl)-1,3-diazaspiro[4.5]dec-3-yl)-2-trifluoromethylbenzonitrile, 9c, [RD48] (0.034 g, 0.076 mmol, 95%) as a white powder.



¹H NMR (CDCl₃, 400 MHz) δ 1.02-1.05 (m, 1H), 1.64-1.76 (m, 4H), 2.03-2.12 (m, 5H), 2.44 (s, 3H), 7.12-7.15 (m, 2H), 7.33-7.36 (m, 2H), 7.85 (dd, *J*₁ = 8.2 Hz, *J*₂ = 1.8 Hz, 1H), 7.96 (d, *J* = 8.3 Hz, 1H), 7.97 (d, *J* = 1.8 Hz, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 20.7, 21.3, 24.0, 32.6, 67.4, 109.9, 114.9, 122.0 (q, *J* = 272.5 Hz), 127.3 (q, *J* = 4.6 Hz), 130.0, 130.5, 132.0, 132.5, 133.3 (q, *J* = 33.2 Hz), 135.2, 137.3, 140.1, 174.1, 180.1.

Example 10

10-1). 1-(4-methylphenyl)aminocyclohexanenitrile, 10a

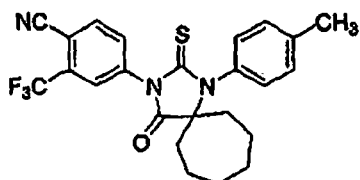
Sodium cyanide (0.147g, 3 mmol) was added to a mixture of *p*-toluidine (0.214 g, 2 mmol) and cycloheptanone (0.337 g, 3 mmol) in acetic acid 90% (3 ml). The reaction mixture was stirred at room temperature for 12 h and then 20 ml of ethyl acetate was added. The organic layer was washed with water (3 × 10 ml), dried over magnesium sulfate and concentrated under vacuum to dryness to yield 1-(4-methylphenyl)aminocyclohexanenitrile, 10a (0.438 g, 1.92 mmol, 96%) as a brown solid.

10-2). 4-(4-imino-2-thioxo-1-(4-methylphenyl)-1,3-diazaspiro[4.5]undec-3-yl)-2-trifluoromethylbenzonitrile, 10b

Triethylamine (0.05 g, 0.5 mmol) was added to a solution of 1a (0.228 g, 1 mmol) and 9a (0.228 g, 1 mmol) in dry THF (2 ml). The reaction mixture was stirred at room temperature for 2 days and then concentrated to yield a dark residue which was subjected to flash chromatography (dichloromethane/acetone, 95:5) to afford 4-(4-imino-2-thioxo-1-(4-methylphenyl)-1,3-diazaspiro[4.5]undec-3-yl)-2-trifluoromethylbenzonitrile, 10b (0.036 g, 0.08 mmol, 8%).

10-3). 4-(4-oxo-2-thioxo-1-(4-methylphenyl)-1,3-diazaspiro[4.5]undec-3-yl)-2-trifluoromethylbenzonitrile, 10c, [RD49]

A mixture of 9b (0.036 g, 0.08 mmol) in HCl aq., 2N (1 ml) and methanol (3 ml) was heated to reflux for 2 h. After being cooled to room temperature, the reaction mixture was poured into cold water (5 ml) and extracted with ethyl acetate (6 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 10c (0.034 g, 0.075 mmol, 94%) as a white powder.



¹H NMR (CDCl₃, 400 MHz) δ 1.24-1.34 (m, 2H), 1.37-1.43 (m, 2H), 1.53-1.60 (m, 2H), 1.74-1.82 (m, 2H), 2.19-2.25 (m, 4H), 2.44 (s, 3H), 7.16-7.19 (m, 2H), 7.32-7.35 (m, 2H), 7.83 (dd, *J*₁ = 8.2 Hz, *J*₂ = 1.8 Hz, 1H), 7.95-7.97 (m, 2H); ¹³C NMR (CDCl₃, 100 MHz) δ 21.4, 22.2, 30.9, 36.3, 71.1, 110.0, 114.9, 121.9 (q, *J* = 272.5 Hz), 127.2 (q, *J* = 4.6 Hz), 129.6, 130.5, 132.3, 133.0, 133.2 (q, *J* = 33.2 Hz), 135.1, 137.4, 140.0, 175.9, 179.7.

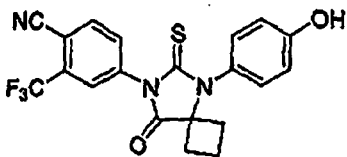
Example 11

11-1). 1-(4-hydroxyphenyl)aminocyclobutanenitrile, 11a

Trimethylsilyl cyanide (0.93 ml, 7 mmol) was added dropwise to a mixture of 4-hydroxyaniline (0.545 g, 5 mmol) and cyclobutanone (0.42 g, 6 mmol). The reaction mixture was stirred at room temperature for 6 h and then concentrated under vacuum to obtain a brown liquid which was subjected to chromatography (dichloromethane:acetone, 98:2) to yield 11a (0.903 g, 4.8 mmol, 96%) as a yellowish solid.

11-2). 4-(8-oxo-6-thioxo-5-(4-hydroxyphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile, 11b, [RD58]

A mixture of 1a (0.57 g, 2.5 mmol) and 7a (0.376 g, 2 mmol) in dry DMF (0.5 ml) was stirred at room temperature for 40 h. To this mixture were added methanol (30 ml) and HCl aq. (5 ml). The second mixture was refluxed for 6 h. After being cooled to room temperature, the reaction mixture was poured into cold water (40 ml) and extracted with ethyl acetate (50 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane:acetone, 98:2) to yield 11b (0.659 g, 1.58 mmol, 79%) as a white powder.



^1H NMR (CDCl_3 , 400 MHz) δ 1.55-1.63 (m, 1H), 2.01-2.09 (m, 1H), 2.50-2.65 (m, 4H), 6.97-7.01 (m, 2H), 7.20-7.24 (m, 2H), 8.02 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 8.14 (d, $J = 1.8$ Hz, 1H), 8.21 (d, $J = 8.3$ Hz, 1H); ^{13}C NMR (Acetone- d_6 , 100 MHz) δ 13.4, 31.3, 67.5, 108.9, 114.8, 116.1, 123.5 (q, $J = 271.5$ Hz), 127.4 (q, $J = 4.9$ Hz), 131.3, 131.8 (q, $J = 32.7$ Hz), 133.3, 135.5, 136.2, 138.5, 158.1, 175.1, 180.7.

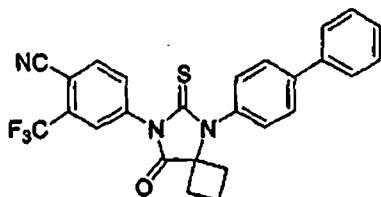
Example 12

12-1). 1-(4-biphenylamino)cyclobutanecarbonitrile, 12a

Trimethylsilyl cyanide (0.2 ml, 1.5 mmol) was added dropwise to a mixture of 4-biphenylamine (0.169 g, 1 mmol) and cyclobutanone (0.098 g, 1.4 mmol). The reaction mixture was stirred at room temperature for 6 h and then concentrated under vacuum to obtain a brown liquid which was subjected to chromatography (dichloromethane) to yield 12a (0.24 g, 0.97 mmol, 97%) as a white solid.

12-2). 4-(8-oxo-6-thioxo-5-(4-biphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile, 12b [RD57]

A mixture of 1a (0.137 g, 0.6 mmol) and 12a (0.124 g, 0.5 mmol) in dry DMF (0.2 ml) was stirred at room temperature for 3 days. To this mixture were added methanol (5 ml) and HCl aq. 2N (1 ml). The second mixture was refluxed for 6 h. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (15 ml). The organic layer was dried over MgSO_4 , concentrated and chromatographed (dichloromethane) to yield 12b (0.162 g, 0.34 mmol, 68%) as a white powder.



^1H NMR (CDCl_3 , 400 MHz) δ 1.67-1.76 (m, 1H), 2.19-2.31 (m, 1H), 2.59-2.74 (m, 4H), 7.40-7.44 (m, 3H), 7.47-7.53 (m, 2H), 7.64-7.67 (m, 2H), 7.79-7.82 (m, 2H), 7.88 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 7.97 (d, $J = 8.2$ Hz, 1H), 8.02 (d, $J = 1.8$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.7, 31.5, 67.5, 110.0, 114.9, 122.0 (q, $J = 272.6$ Hz), 127.1 (q, $J = 4.7$ Hz), 127.3, 128.1, 128.7, 129.0, 130.2, 132.3, 133.5 (q, $J = 33.2$ Hz), 134.2, 135.2, 137.2, 139.6, 142.8, 174.9, 179.9.

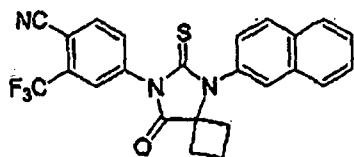
Example 13

13-1). 1-(2-naphthylamino)cyclobutanecarbonitrile, 13a

Trimethylsilyl cyanide (0.27 ml, 2 mmol) was added dropwise to a mixture of 2-aminonaphthalene (0.143 g, 1 mmol) and cyclobutanone (0.098 g, 1.4 mmol). The reaction mixture was stirred at room temperature for 12 h and then concentrated under vacuum to obtain a brown liquid which was subjected to chromatography (dichloromethane) to yield 13a (0.209 g, 0.94 mmol, 94%) as a yellow solid.

13-2). 4-(8-oxo-6-thioxo-5-(4-biphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile, 12b, [RD85]

A mixture of 1a (0.137 g, 0.6 mmol) and 13a (0.111 g, 0.5 mmol) in dry DMF (0.2 ml) was stirred at room temperature for 3 days. To this mixture were added methanol (5 ml) and HCl aq. (1 ml). The second mixture was refluxed for 6 h. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (15 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 12b (0.146 g, 0.325 mmol, 65%) as a white powder.



¹H NMR (CDCl₃, 400 MHz) δ 1.58-1.68 (m, 1H), 2.17-2.29 (m, 1H), 2.61-2.75 (m, 4H), 7.40 (dd, *J*₁ = 8.6 Hz, *J*₂ = 2.0 Hz, 1H), 7.58-7.65 (m, 2H), 7.86-8.00 (m, 5H), 8.04 (*J* = 1.8 Hz, 1H), 8.06 (d, *J* = 8.6 Hz, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 13.7, 31.6, 67.7, 110.0, 114.9, 122.0 (q, *J* = 272.6 Hz), 126.8, 127.1 (q, *J* = 4.8 Hz), 127.2, 127.7, 128.0, 128.3, 129.1, 130.2, 132.2, 132.5, 133.4, 133.5 (q, *J* = 33.1 Hz), 133.6, 135.2, 137.2, 175.0, 180.1.

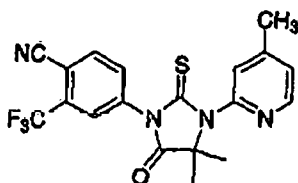
Example 14

14-1). 2-(4-methyl-2-pyridinamino)-2-methylpropanenitrile, 14a

Trimethylsilyl cyanide (0.27 ml, 2 mmol) was added dropwise to a mixture of 2-amino-4-methylpyridine (0.108 g, 1 mmol) and acetone (0.58 g, 10 mmol). The reaction mixture was stirred at room temperature for 6 days and then concentrated under vacuum to obtain a brown liquid which was subjected to chromatography (dichloromethane: acetone, 60:40) to yield 14a (0.133 g, 0.76 mmol, 76%) as a white solid.

14-2). 4-[4,4-dimethyl-3-(4-methylpyridin-2-yl)-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 14b, [RD83]

A mixture of 1a (0.91 g, 0.4 mmol) and 14a (0.053 g, 0.3 mmol) in dry DMF (0.2 ml) was stirred at room temperature for 6 days. To this mixture were added methanol (5 ml) and HCl aq. (1ml). The second mixture was refluxed for 5 h. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (15 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 14b (0.07 g, 0.174 mmol, 58%) as a white powder.



¹H NMR (CDCl₃, 400 MHz) δ 1.70 (s, 6H), 2.44 (s, 3H), 7.19 (d, *J* = 4.4 Hz, 1H), 7.45 (t, *J* = 0.6 Hz, 1H), 7.82 (dd, *J*₁ = 8.2 Hz, *J*₂ = 1.8 Hz, 1H), 7.95 (d, *J* = 1.8 Hz, 1H), 7.97 (d, *J* = 8.2 Hz, 1H), 8.47 (d, *J* = 5.0 Hz, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 21.1, 24.1, 67.1, 110.2, 114.8, 121.9 (q, *J* = 272.6 Hz), 124.4, 125.1, 127.3 (q, *J* = 4.8 Hz), 132.4, 133.5 (q, *J* = 33.2 Hz), 135.3, 137.1, 149.2, 149.5, 150.0, 175.2, 179.0.

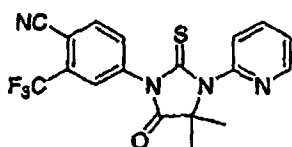
Example 15

15-1). 2-(2-pyridinamino)-2-methylpropanenitrile, 15a

Trimethylsilyl cyanide (0.27 ml, 2 mmol) was added dropwise to a mixture of 2-aminopyridine (0.094 g, 1 mmol) and acetone (0.58 g, 10 mmol). The reaction mixture was stirred at room temperature for 6 days and then concentrated under vacuum to obtain a brown liquid which was subjected to chromatography (dichloromethane: acetone, 60:40) to yield 15a (0.131 g, 0.81 mmol, 81%) as a white solid.

15-2). 4-[4,4-dimethyl-3-(4-pyridin-2-yl)-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 15b, [RD82]

A mixture of 1a (0.91 g, 0.4 mmol) and 15a (0.048 g, 0.3 mmol) in dry DMF (0.3 ml) was stirred at room temperature for 10 days. To this mixture were added methanol (5 ml) and of HCl aq. (1 ml). The second mixture was refluxed for 5 h. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (15 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 15b (0.059 g, 0.153 mmol, 51%) as a white powder.



^1H NMR (CDCl_3 , 400 MHz) δ 1.73 (s, 6H), 7.38 (dd, $J_1 = 7.3$ Hz, $J_2 = 5.4$ Hz, 1H), 7.71 (d, $J = 8.0$ Hz, 1H), 7.87 (dd, $J_1 = 8.2$ Hz, $J_2 = 1.8$ Hz, 1H), 7.95 (td, $J_1 = 7.8$ Hz, $J_2 = 1.8$ Hz, 1H), 7.95 (d, $J = 1.3$ Hz, 1H), 7.98 (d, $J = 8.2$ Hz, 1H), 8.62 (dd, $J_1 = 4.7$ Hz, $J_2 = 1.3$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 24.2, 67.1, 110.3, 114.8, 121.9 (q, $J = 272.6$ Hz), 123.7, 123.8, 127.3 (q, $J = 4.8$ Hz), 132.4, 133.6 (q, $J = 33.2$ Hz), 135.3, 137.1, 138.2, 149.5, 149.6, 175.1, 179.0.

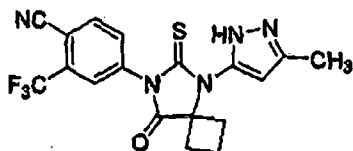
Example 16

16-1). 1-(5-methyl-2H-pyrazol-3-ylamino)-cyclobutanecarbonitrile, 16a

Trimethylsilyl cyanide (0.532 ml, 4.0 mmol) was added dropwise to the mixture of 3-amino-5-methylpyrazole (0.194 g, 2.0 mmol) and cyclobutanone (0.154 g, 2.2 mmol). The reaction mixture was stirred at room temperature for 40 h and then concentrated under vacuum to obtain a dark liquid which was subjected to chromatography (dichloromethane) to yield 16a (0.267 g, 1.52 mmol, 76%) as a off-white powder.

16-2). 4-[5-(5-methyl-2H-pyrazol-3-yl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-7-yl]-2-trifluoromethyl-benzonitrile, 16b, [RD84]

A mixture of 1a (0.0684 g, 0.3 mmol) and 16a (0.053 g, 0.3 mmol) in dry DMF (0.2 ml) was stirred at room temperature for 4 days. To this mixture were added methanol (10 ml) and HCl aq. 2N (2 ml). The second mixture was refluxed for 5 h. After being cooled to room temperature, the reaction mixture was poured into cold water (30 ml) and extracted with ethyl acetate (30 ml). The organic layer was dried over MgSO_4 , concentrated and chromatographed (dichloromethane:acetone, 97:3) to yield 16b (0.0826 g, 0.2 mmol, 67%) as a white powder.

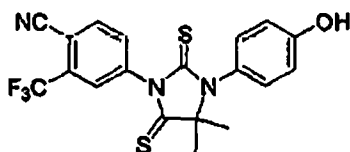


^1H NMR (acetone d_6 , 400 MHz) δ 1.66-1.76 (m, 1H), 2.00-2.07 (m, 1H), 3.35 (s, 3H), 2.56-2.63 (m, 2H), 2.85-2.93 (m, 2H), 8.04 (dd, $J_1 = 8.2$ Hz, $J_2 = 1.6$ Hz, 1H), 8.18 (d, $J = 1.6$ Hz, 1H), 8.22 (d, $J = 8.2$ Hz, 1H), 11.99 (s, 1H); ^{13}C NMR (acetone d_6 , 100 MHz) δ 10.2, 13.1, 31.1, 67.4, 102.5, 109.1, 114.8, 122.5 (q, $J = 271.4$ Hz), 127.8 (q, $J = 4.8$ Hz), 131.9 (q, $J = 33.6$ Hz), 133.6, 135.6, 138.4, 139.9, 145.0, 175.0, 179.6.

Example 17

4-[3-(4-hydroxyphenyl)-4,4-dimethyl-2,5-dithioximidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 17a, [RD59]

A mixture of 3c (0.081 g, 0.2 mmol) and Lawesson reagent (0.097 g, 0.24 mmol) in toluene (3 ml) was heated to reflux for 15 h. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (10 ml). The organic layer was dried over $MgSO_4$, concentrated and chromatographed (dichloromethane:pentane, 9:1) to yield 17a (0.0185 g, 0.044 mmol, 22%) as a white powder.

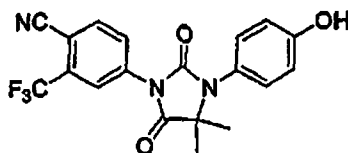


1H NMR ($CDCl_3$, 400 MHz) δ 1.65 (s, 6H), 6.95-6.97 (m, 2H), 7.15-7.18 (m, 2H), 7.75 (d, $J = 8.2$ Hz, 1H), 7.86 (d, $J = 1.8$ Hz, 1H), 7.98 (dd, $J_1 = 8.2$ Hz, $J_2 = 1.8$ Hz, 1H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 27.9, 77.8, 110.9, 114.7, 116.7, 121.9 (q, $J = 272.6$ Hz), 128.1 (q, $J = 4.8$ Hz), 129.1, 130.7, 133.3, 133.5 (q, $J = 33.2$ Hz), 135.5, 140.3, 156.8, 179.9, 207.9.

Example 18

4-[3-(4-hydroxyphenyl)-4,4-dimethyl-2,5-dioximidazolidin-1-yl]-2-trifluoromethylbenzonitrile, 18a, [RD60]

Hydrogen peroxide, 30% (3 ml, 26 mmol) was added dropwise to a solution of 3c (0.121 g, 0.4 mmol) in glacial acetic acid (3 ml). The mixture was stirred at room temperature for 12 h and then 20 ml of ethyl acetate was added. The organic layer was washed with water (3×15 ml), dried over magnesium sulfate, concentrated and chromatographed (dichloromethane) to yield 18a (0.102 g, 0.261 mmol, 87%) as a white powder.



1H NMR ($CDCl_3$, 400 MHz) δ 1.52 (s, 6H), 6.70-6.73 (m, 2H), 7.01-7.04 (m, 2H), 7.92 (d, $J = 8.4$ Hz, 1H), 8.00 (dd, $J_1 = 8.4$ Hz, $J_2 = 1.8$ Hz, 1H), 8.15 (d, $J = 1.8$ Hz, 1H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 23.7, 63.7, 108.4, 115.0, 116.7, 121.9 (q, $J = 272.6$ Hz), 123.5 (q, $J = 4.8$ Hz), 124.0, 128.5, 130.5, 133.6 (q, $J = 33.2$ Hz), 135.5, 136.2, 153.4, 157.2, 174.5.

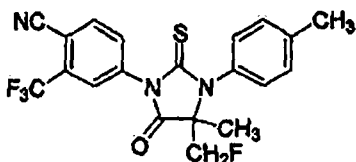
Example 19

19-1). 3-fluoro-2-methyl-2-(4-methylphenyl)aminopropanitrile, 19a

Trimethylsilyl cyanide (0.146 ml, 1.1 mmol) was added dropwise to the mixture of *p*-toluidine (0.107 g, 1 mmol) and fluoroacetone (0.082 g, 1.1 mmol). The reaction mixture was stirred at room temperature for 12 h and then concentrated under vacuum to obtain a brown liquid which was subjected to chromatography (dichloromethane) to yield 19a (0.179 g, 0.93 mmol, 93%) as a yellowish solid.

19-2). 4-(4-fluoromethyl-4-methyl-5-oxo-2-thioxo-3-(4-methylphenyl)imidazolidin-1-yl)-2-trifluoromethylbenzonitrile, 19b, [RD68]

A mixture of 1a (0.16 g, 0.7 mmol) and 19a (0.096 g, 0.5 mmol) in dry DMF (0.3 ml) was stirred at room temperature for 48 h. To this mixture were added methanol (10 ml) and HCl aq. 2N (2 ml). The second mixture was refluxed for 6 h. After being cooled to room temperature, the reaction mixture was poured into cold water (30 ml) and extracted with ethyl acetate (30 ml). The organic layer was dried over $MgSO_4$, concentrated and chromatographed (dichloromethane) to yield 19b (0.168 g, 0.4 mmol, 80%) as a white powder.



1H NMR ($CDCl_3$, 400 MHz) δ 1.49 (s, 3H), 2.44 (s, 3H), 4.35 (dd, $J_1 = 47.2$ Hz, $J_2 = 10.0$ Hz, 1H), 4.71 (dd, $J_1 = 45.2$ Hz, $J_2 = 10$ Hz, 1H), 7.22-7.26 (m, 2H), 7.35-7.39 (m, 2H), 7.82 (dd, $J_1 = 8.2$ Hz, $J_2 = 1.8$ Hz, 1H), 7.93 (d, $J = 1.8$ Hz, 1H), 7.98 (d, $J = 8.2$ Hz, 1H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 17.0 (d, $J = 4.6$ Hz), 21.3, 69.3 (d, $J = 18.3$ Hz), 81.9 (d, $J = 179.5$ Hz), 109.9, 114.8, 121.8 (q, $J = 272.6$ Hz), 127.2 (q, $J = 4.7$ Hz), 129.3, 130.9, 131.6, 132.3, 133.3 (q, $J = 33.2$ Hz), 135.3, 137.0, 140.5, 174.1, 181.4; ^{19}F NMR ($CDCl_3$, 376 MHz) δ -62.5, 110.9.

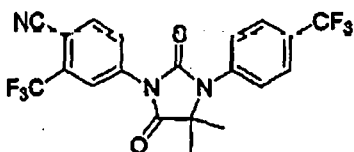
Example 20

20-1). 2-methyl-2-(4-trifluoromethylphenyl)aminopropanenitrile, 20a

A mixture of 4-trifluoromethylaniline (1.61 g, 10 mmol), acetone cyanohydrin (5 ml) and magnesium sulfate (2 g) was heated to 80 °C and stirred for 12 h. To the medium was added ethyl acetate (50 ml) and then washed with water (3 \times 30 ml). The organic layer was dried over $MgSO_4$ and concentrated under vacuum to dryness to yield 20a (2.166 g, 9.5 mmol, 95%) as brown solid.

20-2). **4-(4,4-dimethyl-5-oxo-2-thioxo-3-(4-trifluoromethylphenyl)imidazolidin-1-yl)-2-trifluoromethylbenzonitrile, 20b, [RD66]**

A mixture of 1a (0.114 g, 0.5 mmol) and 20a (0.092 g, 0.4 mmol) in dry DMF (0.3 ml) was stirred at room temperature for 48 h. To this mixture were added methanol (10 ml) and HCl aq. (3 ml). The second mixture was refluxed for 6 h. After being cooled to room temperature, the reaction mixture was poured into cold water (20 ml) and extracted with ethyl acetate (20 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 20b (0.117 g, 0.256 mmol, 64%) as a white powder.



¹H NMR (CDCl₃, 400 MHz) δ 1.61 (s, 6H), 7.45-7.49 (m, 2H), 7.80-7.83 (m, 2H), 7.85 (dd, *J*₁ = 8.3 Hz, *J*₂ = 1.8 Hz, 1H), 7.97 (d, *J* = 1.8 Hz, 1H), 7.99 (d, *J* = 8.2 Hz, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 23.8, 66.6, 110.3, 114.8, 121.8 (q, *J* = 272.6 Hz), 123.5 (q, *J* = 271.1 Hz), 127.0 (q, *J* = 4.6 Hz), 127.1 (q, *J* = 4.7 Hz), 130.3, 131.9 (q, *J* = 32.9 Hz), 132.2, 133.5 (q, *J* = 33.3 Hz), 135.3, 136.9, 138.4, 174.6, 179.9.

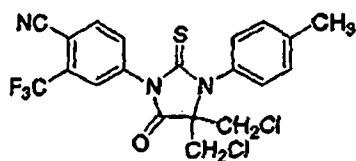
Example 21

21-1). 3-chloro-2-chloromethyl-2-(4-methylphenyl)aminopropanenitrile, 21a

Trimethylsilyl cyanide (0.27 ml, 2 mmol) was added dropwise to a mixture of *p*-toluidine (0.107 g, 1 mmol) and 1,3-dichloroacetone (0.254 g, 2 mmol). The reaction mixture was heat to 80 °C and stirred for 6 h. To the mixture was added 20 ml of ethyl acetate and then wash with water (2 × 20 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 21a (0.192 g, 0.79 mmol, 79%) as a brown powder.

21-2). **4-(4,4-bis(chloromethyl)-5-oxo-2-thioxo-3-(4-methylphenyl)imidazolidin-1-yl)-2-trifluoromethylbenzonitrile, 21b, [RD67]**

A mixture of 1a (0.16 g, 0.7 mmol) and 21a (0.122 g, 0.5 mmol) in dry DMF (0.5 ml) was stirred at room temperature for 10 days. To this mixture were added methanol (10 ml) and of HCl aq. 2N (2 ml). The second mixture was refluxed for 6 h. After being cooled to room temperature, the reaction mixture was poured into cold water (20 ml) and extracted with ethyl acetate (30 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 21b (0.09 g, 0.19 mmol, 38%) as a white powder.



$^1\text{H NMR}$ (CDCl_3 , 400 MHz) δ 2.44 (s, 3H), 3.54 (d, $J = 11.8$ Hz, 2H), 3.93 (d, $J = 11.8$ Hz, 2H), 7.37-7.40 (m, 2H), 7.48-7.51 (m, 2H), 7.79 (dd, $J_1 = 8.2$ Hz, $J_2 = 1.8$ Hz, 1H), 7.88 (d, $J = 1.8$ Hz, 1H), 7.98 (d, $J = 8.2$ Hz, 1H); $^{13}\text{C NMR}$ (CDCl_3 , 100 MHz) δ 21.4, 42.8, 74.3, 110.7, 114.7, 121.7 (q, $J = 272.6$ Hz), 127.2 (q, $J = 4.7$ Hz), 128.8, 131.0, 131.1, 132.4, 133.8 (q, $J = 33.2$ Hz), 135.5, 136.9, 140.9, 169.5, 182.5.

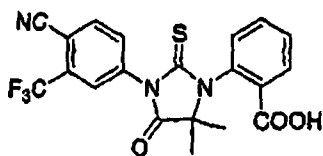
Example 22

22-1). 1-(4-methylphenyl)aminocyclohexanenitrile, 22a

Sodium cyanide (0.245 g, 5 mmol) was added to a mixture of anthranilic acid (0.411 g, 3 mmol) and acetone (1 ml, 13.6 mmol) in acetic acid 90% (3 ml). The reaction mixture was stirred at room temperature for 12 h and then 50 ml of ethyl acetate was added. The organic layer was washed with brine (3 \times 30 ml). The organic layer was dried over magnesium sulfate, concentrated and chromatographed (dichloromethane:acetone, 90:10) to yield 22a (0.551 g, 2.7 mmol, 90%) as a brown solid.

22-2). 2-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]benzoic acid, 22b, [RD65]

A mixture of 1a (0.114 g, 0.5 mmol) and 22a (0.103 g, 0.5 mmol) in dry DMF (0.5 ml) was stirred at room temperature for 3 days. To this mixture were added methanol (10 ml) and HCl aq. 2N, (3 ml). The second mixture was refluxed for 6 h. After being cooled to room temperature, the reaction mixture was poured into cold water (20 ml) and extracted with ethyl acetate (30 ml). The organic layer was dried over MgSO_4 , concentrated and chromatographed (ethyl acetate:pentane, 2:1) to yield 22b (0.143 g, 0.33 mmol, 66%) as a white powder.



$^1\text{H NMR}$ (CDCl_3 , 400 MHz) δ 1.47 (s, 3H), 1.78 (s, 3H), 7.39 (d, $J = 7.7$ Hz, 1H), 7.63 (t, $J = 7.7$ Hz, 1H), 7.76-7.82 (m, 2H), 7.90-7.98 (m, 2H), 8.22 (d, $J = 6.8$ Hz, 1H), 8.96 (bs, 1H); $^{13}\text{C NMR}$ (CDCl_3 , 100 MHz) δ 20.6, 26.2, 67.6, 110.1, 114.8, 121.9 (q, $J = 272.6$ Hz), 127.2 (q, $J = 4.7$ Hz), 128.9, 131.0, 130.2, 132.5, 133.2 (q, $J = 33.3$ Hz), 133.7, 134.7, 135.4, 135.8, 137.3, 169.8, 175.3, 180.7.

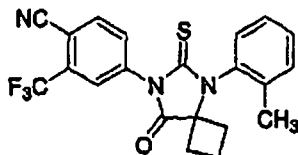
Example 23

23-1). 1-(2-methylphenyl)aminocyclobutanenitrile, 23a

Trimethylsilyl cyanide (0.66 ml, 5 mmol) was added dropwise to the mixture of *p*-toluidine (0.321 g, 3 mmol) and cyclobutanone (0.28 g, 4 mmol). The reaction mixture was stirred at room temperature for 6 h and then concentrated under vacuum to obtain a brown liquid which was subjected to chromatography (dichloromethane) to yield 23a (0.541 g, 2.91 mmol, 97%) as a yellowish solid.

23-2). 4-(8-oxo-6-thioxo-5-(2-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile, 23b, [RD71]

A mixture of 1a (0.114 g, 0.5 mmol) and 23a (0.093 g, 0.5 mmol) in dry DMF (0.3 ml) was stirred at room temperature for 3 days. To this mixture were added methanol (10 ml) and HCl aq. 2N, (3 ml). The second mixture was refluxed for 6 h. After being cooled to room temperature, the reaction mixture was poured into cold water (20 ml) and extracted with ethyl acetate (30 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 23b (0.116 g, 0.28 mmol, 56%) as a white powder.



¹H NMR (CDCl₃, 400 MHz) δ 1.63-1.69 (m, 1H), 2.26 (s, 3H), 2.28-2.41 (m, 2H), 2.58-2.76 (m, 3H), 7.21 (d, *J* = 7.6 Hz, 1H), 7.39-7.49 (m, 3H), 7.89 (dd, *J*₁ = 8.2 Hz, *J*₂ = 1.8 Hz, 1H), 7.97 (d, *J* = 8.2 Hz, 1H), 8.00 (d, *J* = 1.8 Hz, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 14.2, 18.0, 30.7, 32.2, 67.6, 109.9, 114.9, 121.9 (q, *J* = 272.6 Hz), 127.0 (q, *J* = 4.7 Hz), 127.5, 129.8, 130.2, 131.9, 132.3, 133.4, 133.5 (q, *J* = 34.3 Hz), 135.2, 135.8, 137.1, 138.0, 175.3, 178.7.

Example 24

24-1). 1-aminocyclopentanecarbonitrile, 24a

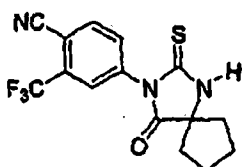
Ammonia anhydrous was bubbled into a mixture of cyclopentanone (0.452 g) and trimethylsilyl cyanide (0.66 ml, 5 mmol). The excess of ammonia was refluxed by a dry ice-acetone condenser. After 1 h of reflux, the ammonia was allowed to degas from the medium and then the remaining mixture was concentrated under vacuum to yield 24a (0.522 g, 4.75 mmol, 95%) as a colorless liquid.

24-2). 4-(4-imino-2-thioxo-1,3-diazaspiro[4.4]non-3-yl)-2-trifluoromethylbenzonitrile, 24b

Triethylamine (0.101 g, 0.1 mmol) was added to a solution of 1a (0.684 g, 3 mmol) and 24a (0.33 g, 3 mmol) in dry THF (5 ml). The reaction mixture was stirred at room temperature for 5 h and then concentrated to yield a brown residue which was subjected to flash chromatography (dichloromethane/acetone, 93:7) to afford 24b (0.741 g, 2.19 mmol, 73%).

24-3). 4-(4-oxo-2-thioxo-1,3-diazaspiro[4.4]non-3-yl)-2-trifluoromethylbenzonitrile, 24c, [RD77]

A mixture of 24b (0.741 g, 2.19 mmol) in HCl aq., 2N (4 ml) and methanol (20 ml) was heated to reflux for 1 h. After being cooled to room temperature, the reaction mixture was poured into cold water (20 ml) and extracted with ethyl acetate (40 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 24c (0.72 g, 2.12 mmol, 97%) as a white powder.

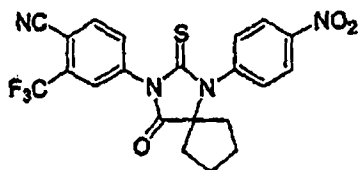


¹H NMR (CDCl₃, 400 MHz) δ 1.86-1.90 (m, 2H), 1.96-2.05 (m, 4H), 2.26-2.30 (m, 2H), 7.80 (dd, $J_1 = 8.2$ Hz, $J_2 = 1.8$ Hz, 1H), 7.92 (d, $J = 1.8$ Hz, 1H), 7.97 (d, $J = 8.2$ Hz, 1H) 8.20 (bs, NH); ¹³C NMR (CDCl₃, 100 MHz) δ 25.3, 38.1, 71.0, 110.1, 114.8, 121.8 (q, $J = 272.7$ Hz), 126.8 (q, $J = 4.7$ Hz), 131.9, 133.6 (q, $J = 34.3$ Hz), 135.3, 136.7, 176.1, 179.8.

Example 25

25). 4-[1-(4-nitrophenyl)-4-oxo-2-thioxo-1,3-diazaspiro[4.4]non-3-yl]-2-trifluoromethylbenzonitrile, 25a, [RD55]

A mixture of 25c (0.0678 g, 0.2 mmol), 1,8-Diazabicyclo[5.4.0]undec-7-ene (0.05 g, 0.33 mmol) and 4-fluoronitrobenzene (0.056 g, 0.4 mmol) in dimethylformamide (0.5 ml) was placed under argon in a sealed-tube and heated to 130 °C for 40 h. The reaction mixture was poured into ethyl acetate (5 ml) and washed with water (2 \times 10 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 25a (0.038 g, 0.084 mmol, 42%) as a white powder.

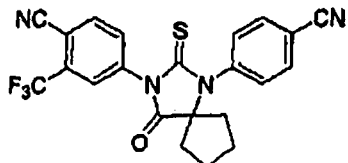


¹H NMR (CDCl₃, 400 MHz) δ 1.53-1.56 (m, 2H), 1.90-1.93 (m, 2H), 2.14-2.18 (m, 2H), 2.37-2.40 (m, 2H), 7.54-7.57 (m, 2H), 7.85 (dd, *J*₁ = 8.2 Hz, *J*₂ = 1.8 Hz, 1H), 7.97 (d, *J* = 1.8 Hz, 1H), 7.98 (d, *J* = 8.2 Hz, 1H), 8.39-8.43 (m, 2H); ¹³C NMR (CDCl₃, 100 MHz) δ 25.2, 36.5, 75.3, 110.3, 114.8, 121.8 (q, *J* = 272.6 Hz), 125.2, 127.0 (q, *J* = 4.7 Hz), 131.4, 132.1, 133.6 (q, *J* = 34.3 Hz), 135.3, 136.9, 141.7, 148.1, 175.6, 180.2.

Example 26

26). 4-[1-(4-cyanophenyl)-4-oxo-2-thioxo-1,3-diazaspiro[4.4]non-3-yl]-2-trifluoromethylbenzonitrile, 26a, [RD54]

A mixture of 24c (0.0678 g, 0.2 mmol), 1,8-diazabicyclo[5.4.0]undec-7-ene (0.061 g, 0.4 mmol) and 4-fluorocyanobenzene (0.048 g, 0.4 mmol) in dimethylformamide (0.5 ml) was placed under argon in a sealed-tube and heated to 140 °C for 5 days. The reaction mixture was poured into ethyl acetate (5 ml) and washed with water (2 × 10 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 26a (0.023 g, 0.052 mmol, 26%) as a white powder.



¹H NMR (CDCl₃, 400 MHz) δ 1.51-1.55 (m, 2H), 1.90-1.93 (m, 2H), 2.12-2.16 (m, 2H), 2.33-2.38 (m, 2H), 7.47-7.50 (m, 2H), 7.81-7.87 (m, 3H), 7.95-7.99 (m, 2H); ¹³C NMR (CDCl₃, 100 MHz) δ 25.2, 36.5, 75.3, 110.3, 113.9, 114.7, 117.5, 121.8 (q, *J* = 272.6 Hz), 127.0 (q, *J* = 4.8 Hz), 131.2, 132.1, 133.6 (q, *J* = 34.3 Hz), 133.8, 135.3, 136.9, 140.0, 175.6, 180.1.

Example 27

27-1). 1-methyl-4-(4-methylphenylamino)piperidine-4-carbonitrile, 27a

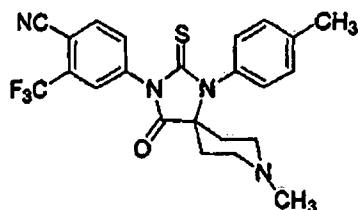
Sodium cyanide (0.318 g, 6.5 mmol) was added to a mixture of *p*-toluidine (0.535 g, 5 mmol) and 1-methyl-4-piperidinone (0.678 g, 6 mmol) in acetic acid 90% (5 ml). The reaction mixture was stirred at room temperature for 6 h and then 100 ml of dichloromethane was added. The organic layer was washed with a solution NaOH, 2N (2 × 50 ml), dried over magnesium sulfate, concentrated and chromatographed (DCM and then acetone) to obtain 27a (0.722 g, 3.15 mmol, 63%).

27.2). 4-(4-amino-8-methyl-2-thioxo-1-(4-methylphenyl)-1,3,8-triazaspiro[4.5]dec-3-yl)-2-trifluoromethylbenzonitrile, 27b

Triethylamine (0.02, 0.2 mmol) was added to a solution of 1a (0.228 g, 1 mmol) and 27a (0.114 g, 0.5 mmol) in dry THF (2 ml). The reaction mixture was stirred at room temperature for 20 h and then concentrated to yield a dark residue which was subjected to flash chromatography (dichloromethane/acetone, 90:10, and then acetone) to afford 27b (0.059 g, 0.13 mmol, 26%).

27-3). 4-(8-methyl-4-oxo-2-thioxo-1-(4-methylphenyl)-1,3,8-triazaspiro[4.5]dec-3-yl)-2-trifluoromethylbenzonitrile, 27c, [RD53]

A mixture of 27b (0.059 g, 0.13 mmol) in HCl aq., 2N (1 ml) and methanol (3 ml) was heated to reflux for 2 h. After being cooled to room temperature, the reaction mixture was poured into cold water (5 ml) and extracted with ethyl acetate (10 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane:acetone, 60:40) to yield 27c (0.055 g, 0.012 mmol, 92%) as a white powder.

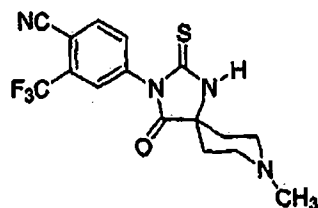


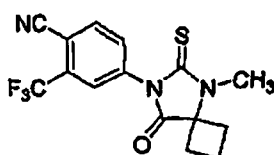
¹H NMR (Acetone-*d*₆, 400 MHz) δ 1.93-1.99 (m, 1H), 2.00-2.04 (m, 1H), 2.18 (s, 3H), 2.24-2.28 (m, 2H), 2.38 (s, 3H), 2.61-2.72 (m, 4H), 7.18-7.20 (m, 2H), 7.32-7.35 (m, 2H), 8.03 (dd, *J*₁ = 8.2 Hz, *J*₂ = 1.8 Hz, 1H), 8.16 (d, *J* = 1.8 Hz, 1H), 8.22 (d, *J* = 8.2 Hz, 1H); ¹³C NMR (Acetone-*d*₆, 100 MHz) δ 20.3, 31.4, 45.1, 49.8, 65.1, 109.1, 114.8, 122.4 (q, *J* = 275.1 Hz), 127.7 (q, *J* = 4.8 Hz), 130.0, 130.5, 131.9 (q, *J* = 32.6 Hz), 132.6, 133.5, 135.6, 138.3, 139.4, 174.0, 180.6.

Example 28

4-(8-methyl-4-oxo-2-thioxo-1,3,8-triazaspiro[4.5]dec-3-yl)-2-(trifluoromethylbenzonitrile, 28a, [RD52]

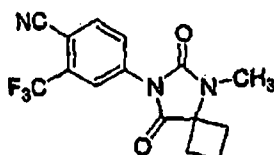
Compound 28a was synthesized according to the procedure described in patent US 5958936.





^1H NMR (CDCl_3 , 400 MHz) δ 1.95-2.06 (m, 1H), 2.21-2.32 (m, 1H), 2.58-2.71 (m, 4H), 3.44 (s, 3H), 7.77 (dd, $J_1 = 8.2$ Hz, $J_2 = 2.0$ Hz, 1H), 7.89 (d, $J = 2.0$ Hz, 1H), 7.93 (d, $J = 8.2$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.7, 30.3, 30.4, 66.1, 109.7, 114.9, 121.9 (q, $J = 272.6$ Hz), 126.9 (q, $J = 4.8$ Hz), 132.1, 133.2 (q, $J = 34.3$ Hz), 135.2, 137.3, 175.1, 178.7.

30-3). 4-(5-methyl-6,8-dioxo-5,7-diazaspiro[3.4]oct-7-yl)-2-(trifluoromethyl)benzonitrile, 30c, [RD74]
Hydrogen peroxide (2 ml, 30%) was added to the mixture of 30b (0.068 g, 0.2 mmol) in glacial acetic acid (3 ml). After being stirred at room temperature for 10 h, the reaction mixture was poured into ethyl acetate (20 ml) and then washed with water (2×20 ml). The organic layer was dried over MgSO_4 , concentrated and chromatographed (dichloromethane:acetone) to yield 30c (0.057 g, 0.176 mmol, 88%) as a white powder.



^1H NMR (CDCl_3 , 400 MHz) δ 1.91-2.35 (m, 1H), 2.21-2.31 (m, 1H), 2.50-2.61 (m, 4H), 3.12 (s, 3H), 7.89 (d, $J = 8.2$ Hz, 1H), 7.97 (dd, $J_1 = 8.2$ Hz, $J_2 = 2.0$ Hz, 1H), 8.12 (d, $J = 2.0$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.9, 25.4, 29.3, 63.4, 108.1, 115.1, 121.6 (q, $J = 272.6$ Hz), 122.9 (q, $J = 4.8$ Hz), 127.9, 133.5 (q, $J = 34.3$ Hz), 135.3, 136.5, 152.7, 174.4.

Example 31

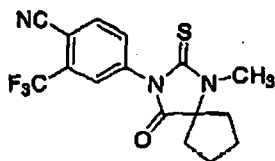
31-1). 1-methylaminocyclopentanecarbonitrile, 31a

Methylamine was bubbled into a refrigerated mixture of cyclopentanone (0.252 g, 3 mmol) and trimethylsilyl cyanide (0.396 g, 4 mmol) until the volume doubled. The mixture was stirred 3 h and then concentrated to dryness to obtain 31a (0.372 g, quantitative).

31-2). 4-(1-methyl-4-oxo-2-thioxo-1,3-diazaspiro[4.4]non-3-yl)-2-(trifluoromethyl)benzonitrile, 31b, [RD75]

A mixture of 1a (0.114 g, 0.5 mmol) and 31a (0.062 g, 0.5 mmol) in dry DMF (0.2 ml) was stirred at room temperature for 0.5 h. To this mixture were added 10 ml of methanol and 2 ml of 2N HCl. The

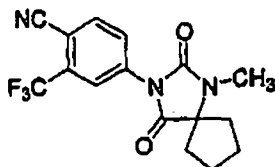
second mixture was refluxed for 2 h. After being cooled to room temperature, the reaction mixture was poured into cold water (20 ml) and extracted with ethyl acetate (30 ml). The organic layer was dried over MgSO_4 , concentrated and chromatographed (dichloromethane) to yield 31b (0.159 g, 0.45 mmol, 90%) as a white powder.



^1H NMR (CDCl_3 , 400 MHz) δ 1.91-2.05 (m, 6H), 2.16-2.21 (m, 2H), 3.27 (s, 3H), 7.77 (dd, $J_1 = 8.2$ Hz, $J_2 = 1.8$ Hz, 1H), 7.89 (d, $J = 1.8$ Hz, 1H), 7.91 (d, $J = 8.2$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 26.4, 30.3, 35.4, 73.2, 109.5, 114.9, 121.9 (q, $J = 272.6$ Hz), 126.9 (q, $J = 4.8$ Hz), 132.2, 133.2 (q, $J = 34.3$ Hz), 135.2, 137.5, 176.8, 178.5.

31-3). 4-(1-methyl-2,4-dioxo-1,3-diazaspiro[4.4]non-3-yl)-2-(trifluoromethyl)benzonitrile, 31c, [RD76]

Hydrogen peroxide (2 ml, 30%) was added to the mixture of 31b (0.07 g, 0.2 mmol) in glacial acetic acid (3 ml). After being stirred at room temperature for 10 h, the reaction mixture was poured into ethyl acetate (20 ml) and then washed with water (2×20 ml). The organic layer was dried over MgSO_4 , concentrated and chromatographed (dichloromethane:acetone) to yield 31c (0.057 g, 0.168 mmol, 84%) as a white powder.

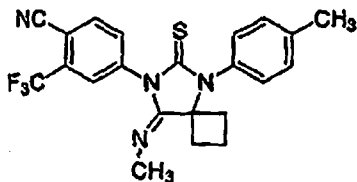


^1H NMR (CDCl_3 , 400 MHz) δ 1.88-1.99 (m, 6H), 2.12-2.17 (m, 2H), 2.98 (s, 3H), 7.88 (d, $J = 8.2$ Hz, 1H), 7.97 (dd, $J_1 = 8.2$ Hz, $J_2 = 1.8$ Hz, 1H), 8.12 (d, $J = 1.8$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 25.2, 26.5, 34.8, 70.1, 108.0, 115.1, 122.0 (q, $J = 272.5$ Hz), 122.9 (q, $J = 4.9$ Hz), 127.9, 133.5 (q, $J = 32.9$ Hz), 135.3, 136.6, 152.7, 176.1.

Example 32

4-(8-methylimino-6-thioxo-5-p-tolyl-5,7-diazaspiro[3.4]oct-7-yl)-2-(trifluoromethyl)benzonitrile, 32a, [RD90]

A mixture of 7b (0.042 g, 0.1 mmol), DBU (0.023 g, 0.15 mmol) and iodomethane (0.073 g, 0.5 mmol) in DMF (0.3 ml) was stirred for 15 h at room temperature. After DMF being evaporated, the medium was chromatographed (dichloromethane) to yield 32a (0.011g, 0.026 mmol, 26%) as white powder.

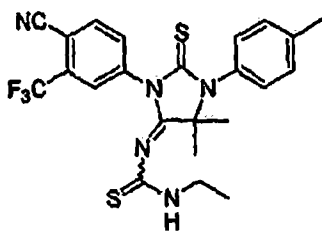


$^1\text{H NMR}$ (CDCl_3 , 400 MHz) δ 1.58-1.65 (m, 1H), 2.04-2.13 (m, 1H), 2.45 (s, 3H), 2.70-2.77 (m, 2H), 3.06-3.10 (m, 2H), 3.58 (s, $\text{CH}_3\text{-N}$, major isomer) [2.70 (s, $\text{CH}_3\text{-N}$, minor isomer)], 7.20-7.34 (m, 4H), 7.75-7.91 (m, 3H); (CDCl_3 , 100 MHz) δ 12.6, 21.4, 30.2, 33.7 (35.3 for the other isomer), 66.9, 109.1, 115.2, 122.1 (q, $J = 272.5$ Hz), 128.5 (q, $J = 4.9$ Hz), 129.8, 130.4, 130.6, 132.8, 133.2 (q, $J = 32.9$ Hz), 133.5, 134.9, 139.8, 157.0, 180.2.

Example 33

1-[3-(4-cyano-3-trifluoromethyl-phenyl)-5,5-dimethyl-2-thioxo-1-p-tolyl-imidazolidin-4-ylidene]-3-ethylthiourea, 33a, [RD91]

A mixture of 5b (0.06 g, 0.149 mmol), ethylthioisocyanate (0.087 g, 1 mmol) and CuI (0.01 g, 0.05 mmol) in DMF (0.1 ml) was heated under microwave for 45 minutes. Then the medium was washed with brine and extracted with ethyl acetate. The organic layer was dried over MgSO_4 , concentrated and chromatographed (HPLC, alumina column) to yield 33a (0.054 g, 0.108 mmol, 72%) as white powder.

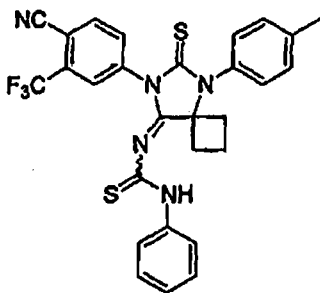


$^1\text{H NMR}$ (CDCl_3 , 400 MHz) δ 1.15 (t, $J = 7.23$ Hz, 3H), 1.70 [1.75 minor isomer] (s, 6H), 2.42 (s, 3H), 3.28-3.39 (m, 2H) [3.15-3.22 (m, 2H), minor isomer], 6.50 (bs, 1H) [6.93 (bs, 1H), minor isomer], 7.14-7.18 (m, 2H), 7.32-7.35 (m, 2H), 7.77-7.94 (m, 3H); $^{13}\text{C NMR}$ (CDCl_3 , 100 MHz) δ 13.31 (13.83 minor), 21.3, 25.22 (24.89 minor), 40.31 (40.67 minor), 68.1, 109.9, 114.9, 122.3 (q, $J = 272.5$ Hz), 127.6 (q, $J = 4.9$ Hz), 129.1, 129.59 (129.55 minor), 130.52 (130.57 minor), 132.27 (132.15 minor), 132.9 (q, $J = 32.9$ Hz), 134.27 (134.15 minor), 134.9, 135.2, 156.33 (156.06 minor), 180.28 (180.06 minor), 187.24 (186.63 minor).

Example 34

1-[7-(4-cyano-3-trifluoromethyl-phenyl)-6-thioxo-5-p-tolyl-5,7-diaza-spiro[3.4]oct-8-ylidene]-3-phenyl-thiourea, 34a, [RD92]

A mixture of 7b (0.021 g, 0.05 mmol) and phenylthioisocyanate (0.027 g, 0.2 mmol) in DMF (0.3 ml) was stirred for 2 days at 60°C. After DMF being evaporated, the medium was chromatographed (dichloromethane) to yield 34a (0.015 g, 0.028 mmol, 57%) as white powder.

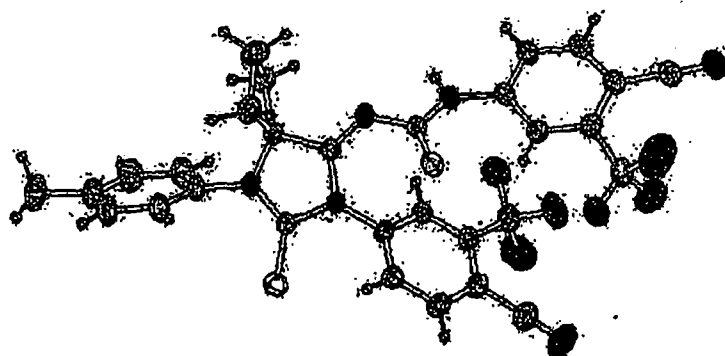
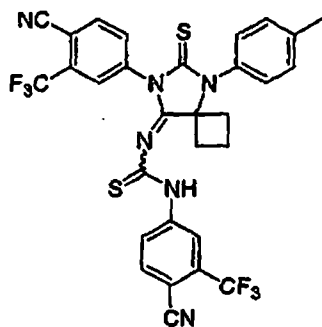


¹H NMR (CDCl₃, 400 MHz) δ 1.59-1.67 (m, 1H), 2.12-2.22 (m, 1H), 2.45 (s, 3H), 2.61-2.71 (m, 2H), 2.81-2.87 (m, 2H), 7.18-7.27 (m, 6H), 7.33-7.41 (m, 5H), 7.60-7.62 (m, 1H), 8.40 (bs, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 13.6, 21.4, 32.3, 69.6, 110.7, 114.8, 121.6, 122.0 (q, *J* = 272.5 Hz), 126.3, 128.0 (q, *J* = 4.9 Hz), 128.9, 129.4, 130.7, 132.5, 133.2 (q, *J* = 32.9 Hz), 134.1, 134.9, 137.7, 139.2, 140.2, 154.8, 180.3, 185.5.

Example 35

1-(4-Cyano-3-trifluoromethyl-phenyl)-3-[7-(4-cyano-3-trifluoromethyl-phenyl)-6-thioxo-5-p-tolyl-5,7-diaza-spiro[3.4]oct-8-ylidene]-thiourea, 35a, [RD93]

A mixture of 1a (0.502 g, 2.2 mmol) and 7a (0.186 g, 1 mmol) in DMF (1 ml) was stirred at room temperature. After 20 hours of stirring, the mixture was concentrated under reduced pressure to yield an orange viscous liquid, which was chromatographed (dichloromethane:acetone, 99:1) to yield 35a (0.269 g, 0.42 mmol, 42%) as a yellow powder.



X-ray structure of 35a

Example 36

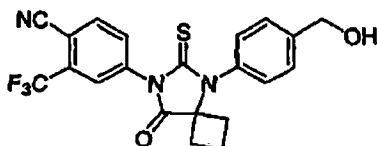
36-1). 1-(4-hydroxymethylphenylamino)-cyclobutanecarbonitrile, 36a

Trimethylsilyl cyanide (0.66 ml, 5 mmol) was added dropwise to a mixture of 4-aminobenzoic acid (0.492 g, 4 mmol) and cyclobutanone (0.35 g, 5 mmol) in dichloromethane (10 ml). The reaction mixture was stirred at room temperature for 6 h and then concentrated under vacuum to obtain a brown liquid which was subjected to chromatography (dichloromethane) to yield 36a (0.677 g, 3.36 mmol, 84%) as a brown solid.

36-2). 4-[8-(4-hydroxymethylphenyl)-5-oxo-7-thioxo-6-azaspiro[3.4]oct-6-yl]-2-trifluoromethylbenzonitrile, 36b, [RD110]

A mixture of 1a (0.342 g, 1.5 mmol) and 36a (0.21 g, 1 mmol) in dry DMF (0.5 ml) was stirred at room temperature for 24 h. To this mixture were added methanol (20-ml) and HCl aq. 2N (5 ml). The second mixture was refluxed for 6 h. After being cooled to room temperature, the reaction mixture was poured into cold water (40 ml) and extracted with ethyl acetate (60 ml). The organic layer was dried over

MgSO₄, concentrated and chromatographed (dichloromethane:acetone, 90:10) to yield 36b (0.296 g, 0.69 mmol, 69%) as a white powder.

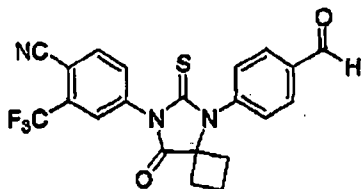


¹H NMR (CDCl₃, 400 MHz) δ 1.63-1.68 (m, 1H), 2.17-2.26 (m, 1H), 2.52-2.68 (m, 4H), 4.75 (s, 2H), 7.30 (d, $J = 8.1$ Hz, 2H), 7.58 (d, $J = 8.1$ Hz, 2H), 7.88 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 7.95-7.98 (m, 2H); ¹³C NMR (CDCl₃, 100 MHz) δ 13.7, 31.5, 64.4, 67.5, 109.9, 114.9, 121.9 (q, $J = 272.6$ Hz), 127.1 (q, $J = 4.7$ Hz), 128.3, 130.0, 132.2, 133.3, 133.4 (q, $J = 33.2$ Hz), 134.2, 137.2, 142.9, 174.9, 179.9.

Example 37

4-[5-(4-formylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl]-2-trifluoromethyl-benzonitrile, 37a, [RD114]

To a mixture of 36b (0.303 g, 0.7 mmol) and Dess-Martin periodinane (0.417g, 1 mmol) in dichloromethane (5 ml) was added pyridine (1.01g, 1 mmol). The mixture was stirred for 2 hours at room temperature and then ethyl ether (10 ml) was added to precipitate the by-product of the reaction. After filtration and concentration under reduced pressure, the mixture was chromatographed (dichloromethane:acetone, 95:5) to yield 37a (0.24 g, 0.56 mmol, 80%) as white powder.

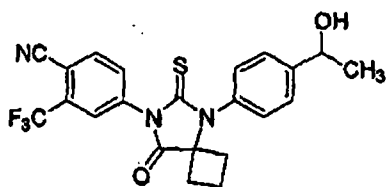


¹H NMR (CDCl₃, 400 MHz) δ 1.62-1.73 (m, 1H), 2.24-2.30 (m, 1H), 2.50-2.58 (m, 2H), 2.69-2.75 (m, 2H), 7.53 (d, $J = 8.1$ Hz, 2H), 7.85 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 7.97-7.99 (m, 2H), 8.11 (d, $J = 8.1$ Hz, 2H), 10.12 (s, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 13.7, 31.7, 67.5, 110.2, 114.8, 121.9 (q, $J = 272.6$ Hz), 127.0 (q, $J = 4.7$ Hz), 129.1, 131.0, 131.2, 132.2, 133.3 (q, $J = 33.2$ Hz), 135.3, 136.9, 140.5, 174.5, 179.8, 190.8.

Example 38

4-[5-[4-(1-hydroxyethyl)phenyl]-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl]-2-trifluoromethyl-benzonitrile, 38a [RD116]

The mixture of 37a (0.043 g, 0.1 mmol) and dry THF (1 ml) in a flamed-dried flash was placed under argon and cooled to -78°C . Then, methylmagnesium iodide (1.1 ml, 0.1 M) was added. The mixture was stirred at -78°C for 30 minutes and warmed slowly to room temperature. The medium was washed with water (3 ml) and extracted with ethyl acetate (10 ml). The organic layer was dried over MgSO_4 , concentrated and chromatographed (dichloromethane:acetone, 95:5) to yield 38a (0.037 g, 0.082 mmol, 82%) as a white powder.

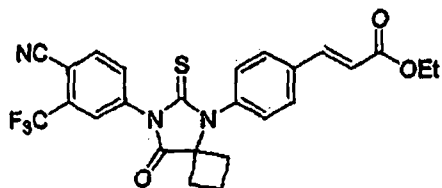


^1H NMR (CDCl_3 , 400 MHz) δ 1.57 (d, $J = 6.5$ Hz, 3H), 1.61-1.71 (m, 1H), 2.09 (d, $J = 3.2$ Hz, OH), 2.16-2.28 (m, 1H), 2.52-2.60 (m, 2H), 2.63-2.69 (m, 2H), 5.00 (dd, $J_1 = 6.5$ Hz, $J_2 = 3.1$ Hz, 1H), 7.29 (d, $J = 8.3$ Hz, 2H), 7.60 (d, $J = 8.2$ Hz, 2H), 7.85 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 7.95-7.98 (m, 2H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.7, 25.3, 31.5, 67.4, 69.8, 110.0, 114.9, 121.9 (q, $J = 272.6$ Hz), 127.0 (q, $J = 4.7$ Hz), 127.1, 129.9, 132.2, 133.4 (q, $J = 33.2$ Hz), 134.1, 135.2, 137.1, 147.6, 174.9, 179.9.

Example 39

3-(4-(7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl)-phenyl)-acrylic acid ethyl ester, 39a [RD117]

A mixture of 37a (0.043 g, 0.1 mmol) and (carboethoxyethylidene)triphenylphosphorane (0.039 g, 0.12 mmol) in dichloromethane (2 ml) was stirred at room temperature for 10 hours. The medium was concentrated and chromatographed (dichloromethane) to yield 39a (0.048 g, 0.096 mmol, 96%) as white powder.

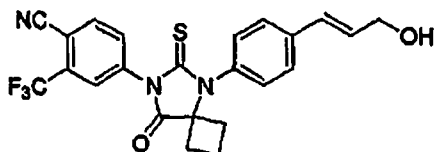


^1H NMR (CDCl_3 , 400 MHz) δ 1.35 (t, $J = 7.1$ Hz, 3H), 1.66-1.70 (m, 1H), 2.19-2.65 (m, 1H), 2.51-2.69 (m, 2H), 2.66-2.72 (m, 2H), 4.28 (q, $J = 7.1$ Hz, 2H), 6.51 (d, $J = 16.1$ Hz, 1H), 7.35 (d, $J = 8.3$ Hz, 2H), 7.72 (d, $J = 8.3$ Hz, 2H), 7.73 (d, $J = 16.1$ Hz, 1H), 7.85 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 7.96-7.98 (m, 2H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.7, 14.3, 31.6, 60.8, 67.5, 110.0, 114.9, 120.5, 121.8 (q, $J = 272.6$ Hz), 127.0 (q, $J = 4.7$ Hz), 129.5, 130.5, 132.2, 133.4 (q, $J = 33.2$ Hz), 135.2, 136.0, 136.5, 137.0, 142.7, 166.5, 174.7, 179.8.

Example 40

4-{5-[4-(3-hydroxypropenyl)-phenyl]-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl}-2-trifluoromethylbenzonitrile, 40a [RD120]

To a mixture of 39a (0.05 g, 0.1 mmol) in dichloromethane (2 ml) at -78°C was added a solution of diisobutylaluminum hydride in THF (0.11 ml, 1M, 0.11 mmol). The mixture was stirred at -78°C for 3 hours. After being warmed to room temperature, the mixture was washed with an aqueous solution of sodium thiosulfate and extracted with ethyl acetate. The organic layer was dried over MgSO_4 , concentrated and chromatographed (dichloromethane:acetone, 95:5) to yield 40a (0.040 g, 0.089 mmol, 89%) as a white powder.



^1H NMR (CDCl_3 , 400 MHz) δ 1.57-1.68 (m, 1H), 2.17-2.39 (m, 1H), 2.55-2.61 (m, 2H), 2.61-2.67 (m, 2H), 4.39 (d, $J = 4.7$ Hz, 2H), 6.47 (dt, $J_1 = 16.0$ Hz, $J_2 = 5.3$ Hz, 1H), 6.70 (d, $J = 16.0$ Hz, 1H), 7.29 (d, $J = 8.3$ Hz, 2H), 7.59 (d, $J = 8.3$ Hz, 2H), 7.85 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 7.96-7.98 (m, 2H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.7, 31.5, 63.4, 67.4, 110.0, 114.8, 120.5, 121.8 (q, $J = 272.6$ Hz), 127.0 (q, $J = 4.7$ Hz), 127.9, 129.2, 130.1, 131.1, 132.1, 133.4 (q, $J = 33.2$ Hz), 135.2, 137.1, 138.4, 174.8, 179.9.

Example 41

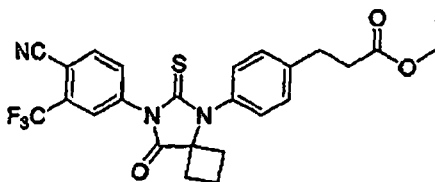
41-1) 3-[4-(1-cyanocyclobutylamino)-phenyl]-propionic acid, 41a (41-1)

Trimethylsilyl cyanide (0.4 g, 4 mmol) was added dropwise to a mixture of 3-(4-aminophenyl)-propionic acid (0.33 g, 2 mmol), cyclobutanone (0.35 g, 5 mmol) and sodium sulfate (1 g) in 1,4-dioxane (5 ml). The mixture was stirred for 15 hours. After filtration to eliminate sodium sulfate, the medium was concentrated under vacuum to obtain a brown liquid which was subjected to chromatography (dichloromethane:acetone, 50:50) to yield 41a (0.472 g, 1.93 mmol, 97%) as a yellowish solid.

41-2) 3-{4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]-phenyl}-propionic acid methyl ester, 41b (41-2) [RD128]

A mixture of 1a (0.661 g, 2.9 mmol) and 41a (0.472 g, 1.93 mmol) in dry DMF (2 ml) was stirred at room temperature for 15 hours. To this mixture were added methanol (10 ml) and HCl aq. (5 ml, 2M). The second mixture was refluxed for 3 h. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (3×30 ml). The organic layer was

dried over $MgSO_4$, concentrated and chromatographed (dichloromethane) to yield 41b (0.582 g, 1.19 mmol, 62%) as a white powder.



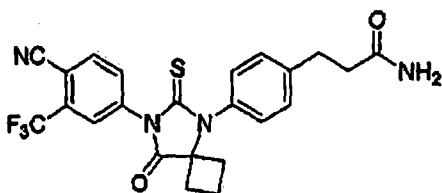
1H NMR ($CDCl_3$, 400 MHz) δ 1.60-1.70 (m, 1H), 2.14-2.26 (m, 1H), 2.51-2.56 (m, 2H), 2.58-2.67 (m, 2H), 2.71 (t, $J = 7.8$ Hz, 2H), 3.05 (t, $J = 7.8$ Hz, 2H), 3.69 (s, 3H), 7.23 (d, $J = 8.2$ Hz, 2H), 7.41 (d, $J = 8.2$ Hz, 2H), 7.85 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 7.95 (d, $J = 8.3$ Hz, 1H), 7.98 (d, $J = 1.8$ Hz, 1H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 13.7, 30.5, 31.4, 35.1, 51.8, 67.5, 109.9, 114.9, 121.9 (q, $J = 272.7$ Hz), 127.1 (q, $J = 4.7$ Hz), 129.9, 130.0, 133.2, 132.3, 133.3 (q, $J = 33.2$ Hz), 135.7, 137.2, 142.5, 173.1, 174.9, 179.9.

41-3) 3-(4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl)-propionic acid, 41c (41-3) [RD132]

A mixture of 41b (0.487 g, 1 mmol) in methanol (10 ml) and solution of sodium hydroxide (10 ml, 2M) was stirred at room temperature for 5 hours. Methanol was evaporated. The residue was adjusted to pH = 5 by HCl aq. (2M) and then extracted with ethyl acetate (3 \times 50 ml). The organic layer was dried over $MgSO_4$ and concentrated to dryness to obtain 41c (0.472 g, 0.99 mmol, 99%).

41-4) 3-(4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl)-propanamide, 41d (41-4) [RD133]

To a suspension of 41c (0.094 g, 0.2 mmol) in THF (10 ml) at $-5^\circ C$ was added thionyl chloride (0.019 ml, 0.26 mmol). The medium was stirred at $-5^\circ C$ for one hour. Then ammonia was bubbled into the mixture. The excess of ammonia was condensed by reflux condenser at $-78^\circ C$ for 30 minutes and then was allowed to evaporate. The medium was filtered. The filtrate was concentrated and chromatographed (dichloromethane:acetone, 70:30) to yield 41d (0.09 g, 0.19 mmol, 95%) as an off-white powder.

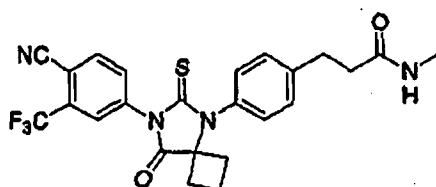


1H NMR (acetone- d_6 , 400 MHz) δ 1.52-1.60 (m, 1H), 2.01-2.09 (m, 1H), 2.49-2.58 (m, 4H), 2.61-2.67 (m, 2H), 2.98 (t, $J = 7.5$ Hz, 2H), 6.20 (bs, 1H), 6.78 (bs, 1H), 7.31 (d, $J = 8.2$ Hz, 2H), 7.44 (d, $J = 8.2$ Hz, 2H), 8.03 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 8.15 (d, $J = 1.8$ Hz, 1H), 8.22 (d, $J = 8.3$ Hz, 1H); ^{13}C

NMR (acetone- d_6 , 100 MHz) δ 13.4, 30.7, 31.2, 36.4, 67.5, 109.0, 114.8, 122.5 (q, $J = 271.5$ Hz), 127.5 (q, $J = 4.7$ Hz), 129.5, 130.0, 131.8 (q, $J = 32.5$ Hz), 133.3, 133.8, 135.6, 138.4, 143.2, 171.6, 174.9, 178.0.

41-5) 3-[4-[7-(4-Cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl]-N-methyl-propionamide, 41e (41-5) [RD134]

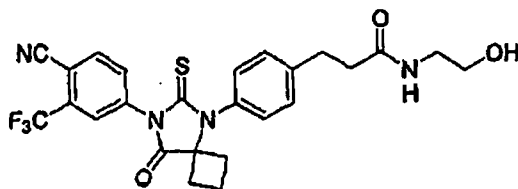
To a suspension of 41c (0.094 g, 0.2 mmol) in THF (10 ml) at -5°C was added thionyl chloride (0.019 ml, 0.26 mmol). The medium was stirred at -5°C for one hour. Then methylamine was bubbled into the mixture at -5°C for 30 minutes. The medium was filtered. The filtrate was concentrated and chromatographed (dichloromethane:acetone, 75:25) to yield 41e (0.092 g, 0.19 mmol, 95%) as an off-white powder.



^1H NMR (acetone- d_6 , 400 MHz) δ 1.51-1.60 (m, 1H), 2.01-2.11 (m, 1H), 2.48-2.58 (m, 4H), 2.61-2.67 (m, 2H), 2.77 (d, $J = 4.6$ Hz, 3H), 2.98 (t, $J = 7.5$ Hz, 2H), 7.03 (bs, NH), 7.33 (d, $J = 8.2$ Hz, 2H), 7.42 (d, $J = 8.2$ Hz, 2H), 8.01 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 8.13 (d, $J = 1.8$ Hz, 1H), 8.20 (d, $J = 8.3$ Hz, 1H); ^{13}C NMR (acetone- d_6 , 100 MHz) δ 13.4, 25.3, 30.0, 31.2, 37.0, 67.6, 109.0, 114.8, 122.5 (q, $J = 271.5$ Hz), 127.4 (q, $J = 4.7$ Hz), 129.5, 130.0, 131.9 (q, $J = 32.5$ Hz), 133.3, 133.8, 135.6, 138.4, 143.1, 171.7, 175.0, 178.0.

41-6) 3-[4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl]-N-(2-hydroxyethyl)-propionamide, 41f (41-6) [RD135]

To a suspension of 41c (0.094 g, 0.2 mmol) in THF (10 ml) at -5°C was added thionyl chloride (0.019 ml, 0.26 mmol). The medium was stirred at -5°C for one hour. Then 2-aminoethanol (0.0183 g, 0.03 mmol) was added into the mixture at -5°C . After stirring of an additional 30 minutes, the medium was filtered. The filtrate was concentrated and chromatographed (dichloromethane:acetone, 50:50) to yield 41f (0.093 g, 0.18 mmol, 90%) as an off-white powder.



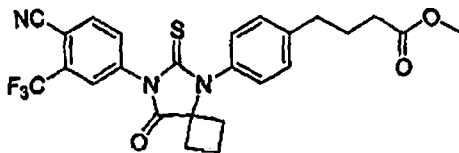
¹H NMR (acetone-*d*₆, 400 MHz) δ 1.51-1.61 (m, 1H), 2.01-2.11 (m, 1H), 2.49-2.66 (m, 6H), 2.99 (t, *J* = 7.5 Hz, 2H), 3.27 (dd, *J*₁ = 11.2 Hz, *J*₂ = 5.6 Hz, 3H), 3.51 (dd, *J*₁ = 11.2 Hz, *J*₂ = 5.6 Hz, 2H), 3.87 (bs, OH), 7.20 (bs, NH), 7.33 (d, *J* = 8.2 Hz, 2H), 7.43 (d, *J* = 8.2 Hz, 2H), 8.02 (dd, *J*₁ = 8.3 Hz, *J*₂ = 1.8 Hz, 1H), 8.14 (d, *J* = 1.8 Hz, 1H), 8.22 (d, *J* = 8.3 Hz, 1H); ¹³C NMR (acetone-*d*₆, 100 MHz) δ 13.4, 31.0, 31.2, 37.1, 42.0, 61.2, 67.6, 109.0, 114.8, 122.5 (q, *J* = 271.5 Hz), 127.4 (q, *J* = 4.7 Hz), 129.6, 130.0, 131.9 (q, *J* = 32.5 Hz), 133.3, 133.8, 135.6, 138.4, 143.0, 171.9, 175.0, 178.1.

42-1) 4-[4-(1-Cyanocyclobutylamino)-phenyl]-butyric acid, 42a

Trimethylsilyl cyanide (0.50 g, 5 mmol) was added dropwise to a mixture of 4-(4-aminophenyl)-butyric acid (0.537 g, 3 mmol), cyclobutanone (0.35 g, 5 mmol) and sodium sulfate (1 g) in 1,4-dioxane (10 ml). The mixture was stirred for 15 hours. After filtration to eliminate sodium sulfate, the medium was concentrated under vacuum to obtain a brown liquid which was subjected to chromatography (dichloromethane:acetone, 50:50) to yield 42a (0.665 g, 2.58 mmol, 86%) as a yellowish solid.

42-2) 4-[4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]-phenyl]-butyric acid methyl ester, 42b [RD129]

A mixture of 1a (0.547 g, 2.4 mmol) and 42a (0.342 g, 1.5 mmol) in dry DMF (2 ml) was stirred at room temperature for 15 hours. To this mixture were added methanol (10 ml) and HCl aq. (5 ml, 2M). The second mixture was refluxed for 3 h. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (3 × 30 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane) to yield 42b (0.594 g, 1.18 mmol, 79%) as a white powder.

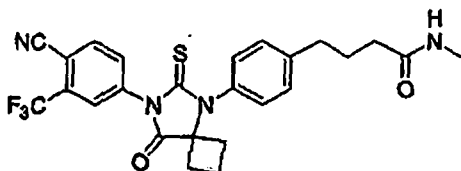


¹H NMR (CDCl₃, 400 MHz) δ 1.60-1.70 (m, 1H), 1.98-2.07 (m, 2H), 2.14-2.26 (m, 1H), 2.40 (t, *J* = 7.4 Hz, 2H), 2.52-2.60 (m, 2H), 2.62-2.68 (m, 2H), 2.74 (t, *J* = 7.4 Hz, 2H), 3.68 (s, 3H), 7.22 (d, *J* = 8.2 Hz, 2H), 7.38 (d, *J* = 8.2 Hz, 2H), 7.86 (dd, *J*₁ = 8.3 Hz, *J*₂ = 1.8 Hz, 1H), 7.95 (d, *J* = 8.3 Hz, 1H), 7.98 (d, *J* = 1.8 Hz, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 13.7, 26.1, 31.4, 33.5, 34.8, 51.7, 67.5, 109.9, 114.9, 121.9 (q, *J* = 272.7 Hz), 127.1 (q, *J* = 4.7 Hz), 129.7, 130.1, 132.3, 133.0, 133.3 (q, *J* = 33.2 Hz), 135.2, 137.2, 143.5, 173.8, 175.0, 179.9.

42-3) 4-[4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]-phenyl]-butyric acid, 42c [RD141]

42-5) 4-{4-[7-(4-Cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl}-N-methyl-butylamide, 42e [RD131]

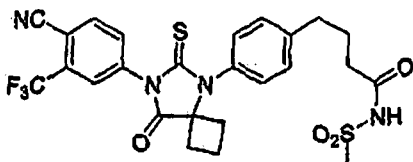
To a suspension of 42c (0.097 g, 0.2 mmol) in THF (10 ml) at -5°C was added thionyl chloride (0.019 ml, 0.26 mmol). The medium was stirred at -5°C for one hour. Then methylamine was bubbled into the mixture at -5°C for 30 minutes. The medium was filtered. The filtrate was concentrated and chromatographed (dichloromethane:acetone, 75:25) to yield 42e (0.095 g, 0.19 mmol, 95%) as an off-white powder.



^1H NMR (CDCl_3 , 400 MHz) δ 1.52-1.64 (m, 1H), 1.94-2.01 (m, 2H), 2.10-2.17 (m, 1H), 2.20 (t, $J = 7.3$ Hz, 2H), 2.46-2.62 (m, 4H), 2.69 (t, $J = 7.3$ Hz, 2H), 2.73 (d, $J = 4.7$ Hz, 3H), 6.09 (bs, 1H), 7.16 (d, $J = 8.2$ Hz, 2H), 7.33 (d, $J = 8.2$ Hz, 2H), 7.82 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 7.91 (d, $J = 8.3$ Hz, 1H), 7.94 (d, $J = 1.8$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.7, 26.2, 26.8, 31.4, 35.0, 35.7, 67.5, 109.7, 114.9, 121.9 (q, $J = 272.7$ Hz), 127.1 (q, $J = 4.7$ Hz), 129.7, 130.0, 132.3, 133.8, 133.3 (q, $J = 33.2$ Hz), 135.2, 137.3, 143.7, 173.3, 174.9, 179.8.

42-6) N-(4-(4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]phenyl)-butanoyl)-methanesulfonamide, 42f [RD157]

[0099] A mixture of 4-(4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]phenyl)butanoic acid (42c) (0.049 g, 0.1 mmol), 2,4,6-trichlorobenzoyl chloride (0.244 g, 1 mmol), 4-dimethylaminopyridine (0.122 g, 1 mmol) and methanesulfonamide (0.019 g, 0.2 mmol) in dichloromethane was stirred at room temperature for 20 hours. The mixture was concentrated and chromatographed (dichloromethane:acetone, 80:20) to yield *N*-(4-(4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]phenyl)-butanoyl)-methanesulfonamide (42f) [RD157] (0.053 g, 0.094 mmol, 94%), the structure of which is illustrated in Formula 8, as a white powder.

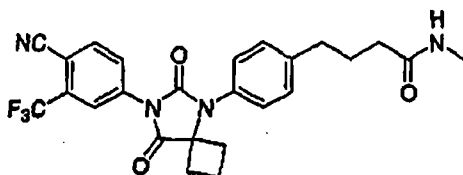


Formula 8

^1H NMR (acetone- d_6 , 400 MHz) δ 1.51-1.60 (m, 1H), 1.96-2.11 (m, 3H), 2.49 (t, $J = 7.3$ Hz, 2H), 2.51-2.57 (m, 2H), 2.61-2.67 (m, 2H), 2.75 (t, $J = 7.5$ Hz, 2H), 2.94 (bs, 1H), 3.24 (s, 3H), 7.33 (d, $J = 8.3$ Hz, 2H), 7.43 (d, $J = 8.2$ Hz, 2H), 8.02 (dd, $J = 8.3, 1.6$ Hz, 1H), 8.02 (d, $J = 1.6$ Hz, 1H), 8.21 (d, $J = 8.3$ Hz, 1H); ^{13}C NMR (acetone- d_6 , 100 MHz) δ 13.4, 25.8, 31.2, 34.3, 35.2, 40.6, 67.6, 109.0, 114.8, 122.5 (q, $J = 271.5$ Hz), 127.5 (q, $J = 4.9$ Hz), 129.6, 130.1, 131.9 (q, $J = 33.6$ Hz), 133.3, 133.9, 135.6, 138.4, 143.1, 171.9, 175.0, 180.5.

42-7) *N*-methyl-4-{4-[7-(4-cyano-3-trifluoromethylphenyl)-6,8-dioxo-5,7-diazaspiro[3.4]oct-5-yl]-phenyl}butyramide, 42g [RD158]

[00100] Hydrogen peroxide (30%, 0.4) was added dropwise to a solution of *N*-methyl-4-{4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]phenyl}butanamide (42e) (0.032 g, 0.064 mmol) in glacial acetic acid (0.5 ml). The mixture was stirred at room temperature for 5 hours and then washed with water and extracted with ethyl acetate. The organic layer was dried over magnesium sulfate, concentrated and chromatographed (dichloromethane:acetone, 80:20) to yield *N*-methyl-4-{4-[7-(4-cyano-3-trifluoromethylphenyl)-6,8-dioxo-5,7-diazaspiro[3.4]oct-5-yl]-phenyl}butyramide (42g) [RD158] (0.029 g, 0.06 mmol, 94%), the structure of which is illustrated in Formula 9, as a white powder.



Formula 9

^1H NMR (CDCl_3 , 400 MHz) δ 1.63-1.71 (m, 1H), 1.93-2.04 (m, 2H), 2.18-2.27 (m, 3H), 2.44-2.53 (m, 2H), 2.57-2.65 (m, 2H), 2.70 (t, $J = 7.3$ Hz, 2H), 2.79 (d, $J = 4.8$ Hz, 3H), 5.79 (bs, 1H), 7.21 (d, $J = 8.2$ Hz, 2H), 7.34 (d, $J = 8.2$ Hz, 2H), 7.92 (d, $J = 8.4$ Hz, 1H), 8.03 (dd, $J = 8.3, 1.8$ Hz, 1H), 8.18 (d, $J = 1.8$ Hz, 1H).

Example 43

43-1) 4-(4-aminophenyl)-piperazine-1-carboxylic acid *tert*-butyl ester, 43a

A mixture of 4-iodoaniline (0.654 g, 3 mmol), piperazine-1-carboxylic acid *tert*-butyl ester (0.67 g, 3.6 mmol), potassium phosphate (1.272 g, 6 mmol), ethylene glycol (0.33 ml) and copper iodide (0.03 g, 0.15 mmol) in 2-propanol (3 ml) was placed under argon in a sealed-tube and heated to 80°C for 30 hours. After being cooled to room temperature, the medium was washed with water (50 ml) and extracted with ethyl acetate (100 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane:acetone, 70:30) to yield 43a (0.36 g, 1.3 mmol, 43%) as a yellow powder.

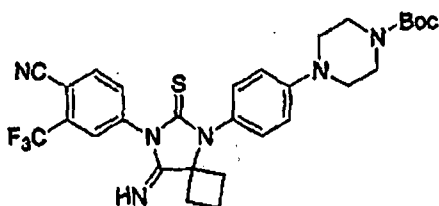
43-2) 4-[4-(1-cyanocyclobutylamino)phenyl]-piperazine-1-carboxylic acid *tert*-butyl ester, 43b

Trimethylsilyl cyanide (0.3 g, 3 mmol) was added dropwise to a mixture of 43a (0.415 g, 1.5 mmol), cyclobutanone (0.21 g, 3 mmol) and sodium sulfate (1 g) in dichloromethane (5 ml). The mixture was stirred for 15 hours. After filtration to eliminate sodium sulfate, the medium was concentrated under vacuum to obtain a brown liquid which was subjected to chromatography (dichloromethane:acetone, 75:25) to yield 43b (0.448 g, 1.26 mmol, 84%) as a yellow solid.

43-3) 4-[4-[7-(4-cyano-3-trifluoromethylphenyl)-8-imino-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]-phenyl]-piperazine-1-carboxylic acid *tert*-butyl ester, 43c.[RD139]

and
4-[4-[7-(4-cyano-3-trifluoromethylphenyl)-8-(4-cyano-3-trifluoromethylphenylthiocarbamoylimino)-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]-phenyl]-piperazine-1-carboxylic acid *tert*-butyl ester, 43d [RD140]

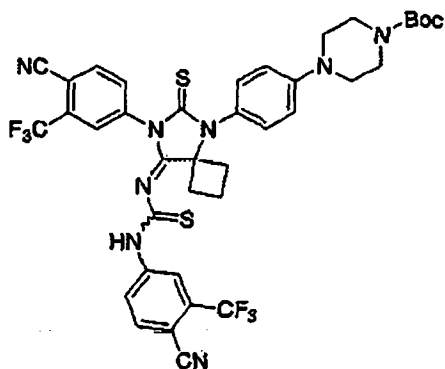
A mixture of 1a (0.228 g, 1 mmol) and 43b (0.472 g, 0.63 mmol) in dry DMF (1 ml) was stirred at room temperature for 20 hours. The mixture was concentrated and chromatographed (dichloromethane:acetone, 90:10) to yield 43c (0.173 g, 0.296 mmol, 47%), the structure of which is illustrated in Formula 10, as a off-white powder and 43d (0.169 g, 0.21 mmol, 33%), the structure of which is illustrated in Formula 11, as a yellow powder.



Formula 10

¹H NMR (CDCl₃, 400 MHz) δ 1.48, (s, 9H), 1.57-1.67 (m, 1H), 2.01-2.09 (m, 1H), 2.59-2.70 (m, 4H), 3.25 (t, J = 5.1 Hz, 4H), 3.59 (t, J = 4.9 Hz, 4H), 7.02 (d, J = 8.9 Hz, 2H), 7.20 (d, J = 8.9 Hz, 2H), 7.81

(d, $J = 7.4$ Hz, 1H), 7.93 (s, 1H), 7.97 (d, $J = 8.1$ Hz, 1H).

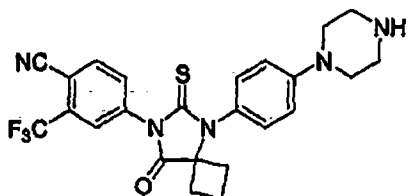


Formula 11

^1H NMR (CDCl_3 , 400 MHz) δ 1.48, (s, 9H), 1.57-1.64 (m, 1H), 2.01-2.10 (m, 1H), 2.60-2.89 (m, 4H), 3.24 (t, $J = 5.1$ Hz, 4H), 3.57 (t, $J = 4.9$ Hz, 4H), 7.02 (d, $J = 8.9$ Hz, 2H), 7.20 (d, $J = 8.9$ Hz, 2H), 7.54-7.98 (m, 4H), 7.97 (d, $J = 8.1$ Hz, 1H).

43-4) 4-[8-Oxo-5-(4-piperazin-1-yl-phenyl)-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl]-2-trifluoromethylbenzonitrile, 43e [RD137]

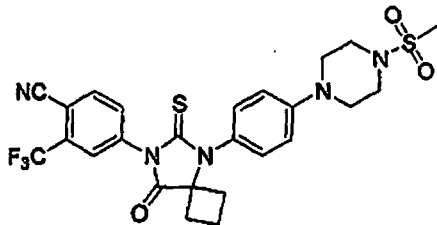
A mixture of 43c (0.117 g, 0.2 mmol), methanol (5 ml) and HCl aq. (2 ml, 2M) was refluxed for 2 hours. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (3×30 ml). The organic layer was dried over MgSO_4 , concentrated and chromatographed (dichloromethane:acetone, 50:50 and then methanol:acetone, 50:50) to yield 43e (0.089 g, 0.184 mmol, 92%) as a white powder.



^1H NMR (CD_3OD , 400 MHz) δ 1.51-1.61 (m, 1H), 2.01-2.11 (m, 1H), 2.48-2.59 (m, 4H), 2.90-2.97 (m, 4H), 3.25-3.30 (m, 4H), 7.03 (d, $J = 8.9$ Hz, 2H), 7.16 (d, $J = 8.9$ Hz, 2H), 7.86 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 8.02 (d, $J = 8.3$ Hz, 1H), 8.07 (d, $J = 1.8$ Hz, 1H); ^{13}C NMR (CD_3OD , 100 MHz) δ 13.2, 30.9, 45.1, 48.9, 67.5, 108.9, 114.8, 115.9, 122.3 (q, $J = 271.7$ Hz), 126.4, 127.3 (q, $J = 4.7$ Hz), 130.4, 132.2 (q, $J = 33.2$ Hz), 133.0, 135.4, 138.1, 152.1, 175.4, 180.4.

43-5) 4-{5-[4-(4-methanesulfonylpiperazin-1-yl)-phenyl]-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl}-2-trifluoromethylbenzonitrile, 43f [RD138]

A mixture of 43e (0.049 g, 0.1 mmol), methanesulfonyl chloride (0.012 ml, 0.15 mmol) and triethylamine (0.15 ml) in dichloromethane was stirred at room temperature for 5 hours. The medium was filtered. The filtrate was concentrated and chromatographed (dichloromethane: acetone, 95:5) to yield 43f (0.042 g, 0.074 mmol, 74%) as a white powder.



$^1\text{H NMR}$ (CDCl_3 , 400 MHz) δ 1.62-1.70 (m, 1H), 2.14-2.23 (m, 1H), 2.51-2.58 (m, 2H), 2.61-2.67 (m, 2H), 2.84 (s, 3H), 3.39 (s, 8H), 7.05 (d, $J = 8.9$ Hz, 2H), 7.20 (d, $J = 8.9$ Hz, 2H), 7.84 (dd, $J_1 = 8.3$ Hz, $J_2 = 1.8$ Hz, 1H), 7.95 (d, $J = 8.3$ Hz, 1H), 7.97 (d, $J = 1.8$ Hz, 1H); $^{13}\text{C NMR}$ (CDCl_3 , 100 MHz) δ 13.7, 31.4, 34.6, 45.7, 48.4, 67.5, 109.8, 114.9, 117.0, 121.9 (q, $J = 272.7$ Hz), 126.8, 127.1 (q, $J = 4.7$ Hz), 130.7, 132.3, 133.4 (q, $J = 33.2$ Hz), 135.2, 137.3, 151.1, 175.0, 180.2.

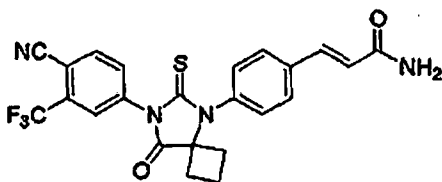
Example 44

44-1) 3-{4-[7-(4-Cyano-3-trifluoromethyl-phenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl}-acrylic acid, 44a

A mixture of 39a (0.025 g, 0.05 mmol) in methanol (2 ml) and solution of sodium hydroxide (2 ml, 2M) was stirred at room temperature for 5 hours. Methanol was evaporated. The residue was adjusted to pH = 5 by HCl aq. (2M) and then extracted with ethyl acetate (3 \times 50 ml). The organic layer was dried over MgSO_4 and concentrated to dryness to obtain 44a (0.02 g, 0.042 mmol, 85%).

44-2) 3-{4-[7-(4-Cyano-3-trifluoromethyl-phenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl}-acrylamide, 44b [RD119]

To a suspension of 44b (0.02 g, 0.042 mmol) in THF (1 ml) at -5°C was added thionyl chloride (0.007 ml, 0.1 mmol). The medium was stirred at -5°C for one hour. Then ammonia was bubbled into the mixture. The excess of ammonia was condensed by reflux condenser at -78°C for 30 minutes and then was allowed to evaporate. The medium was filtered. The filtrate was concentrated and chromatographed (dichloromethane:acetone, 70:30) to yield 44b (0.014 g, 0.03 mmol, 71%) as an off-white powder.

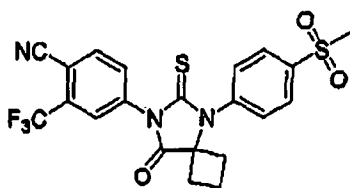


¹H NMR (DMSO-*d*₆, 400 MHz) δ 1.49-1.52 (m, 1H), 1.88-1.93 (m, 1H), 2.37-2.46 (m, 2H), 2.57-2.62 (m, 2H), 6.66 (d, *J* = 15.9 Hz, 1H), 7.16 (bs, 1H), 7.43 (d, *J* = 8.3 Hz, 2H), 7.47 (d, *J* = 15.9 Hz, 1H), 7.58 (bs, 1H), 8.03 (dd, *J*₁ = 8.3 Hz, *J*₂ = 1.8 Hz, 1H), 8.23 (d, *J* = 1.8 Hz, 1H), 8.34 (d, *J* = 8.3 Hz, 1H).

Example 45 [RD145]

[00101] Trimethylsilyl cyanide (0.4 g, 4 mmol) was added dropwise to a mixture of 4-methanesulfonylphenylamine hydrochloride (0.415 g, 2 mmol), cyclobutanone (0.28 g, 4 mmol) and sodium sulfate (1 g) in DMF (3 ml). The mixture was stirred for 15 hours at 120 °C. After filtration to remove the sodium sulfate, the filtrate was washed with brine and extracted with ethyl acetate. The organic layer was concentrated and chromatographed (dichloromethane:acetone, 90:10) to yield 1-(4-methanesulfonylphenylamino)cyclobutanecarbonitrile (45a) (0.116 g, 0.44 mmol, 22%) as a yellowish solid. 4-methanesulfonylphenylamine (0.201g, 1.17 mmol, 59%) was also recovered.

[00102] A mixture of 4-isothiocyanato-2-trifluoromethylbenzonitrile (1a) (0.0141 g, 0.62 mmol) and 1-(4-methanesulfonylphenylamino)cyclobutanecarbonitrile (45a) (0.11 g, 0.42 mmol) in dry DMF (2 ml) was stirred at room temperature for 3 days. To this mixture were added methanol (10 ml) and aq. 2N HCl (5 ml). The second mixture was refluxed for 3 h. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (3 × 30 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane:acetone, 97:3) to yield 4-[5-(4-methanesulfonylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl]-2-trifluoromethylbenzonitrile (45b) [RD145] (0.031 g, 0.065 mmol, 15%), the structure of which is illustrated in Formula 14, as a white powder.



Formula 14

¹H NMR (CDCl₃, 400 MHz) δ 1.63-1.72 (m, 1H), 2.21-2.28 (m, 1H), 2.46-2.54 (m, 2H), 2.68-2.74 (m,

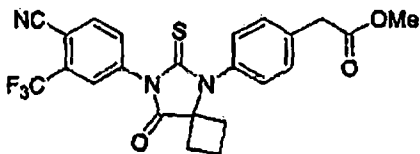
2H), 3.16 (s, 3H), 7.57 (d, $J = 8.3$ Hz, 2H), 7.85 (dd, $J = 8.3, 1.8$ Hz, 1H), 7.97 (d, $J = 1.8$ Hz, 1H), 7.98 (d, $J = 8.3$ Hz, 1H), 8.17 (d, $J = 8.3$ Hz, 2H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.6, 31.8, 44.4, 67.5, 110.2, 114.8, 122.4 (q, $J = 271.5$ Hz), 127.0 (q, $J = 4.9$ Hz), 129.4, 131.4, 132.1, 133.6 (q, $J = 33.3$ Hz), 135.3, 136.8, 140.3, 141.8, 174.4, 179.9.

Example 46

[00103] Trimethylsilyl cyanide (0.69 g, 7 mmol) was added dropwise to a mixture of 4-aminophenylacetic acid (0.755 g, 5 mmol) and cyclobutanone (0.49 g, 7 mmol) in dioxane (20 ml). The mixture was stirred for 8 hours at 80 °C. The mixture was concentrated and chromatographed (dichloromethane:acetone, 60:40) to yield [4-(1-cyanocyclobutylamino)phenyl]acetic acid (46a) (1.138 g, 4.95 mmol, 99%) as a white solid.

46-1) RD146

[00104] A mixture of 4-isothiocyanato-2-trifluoromethylbenzonitrile (1a) (0.638 g, 2.8 mmol) and [4-(1-cyanocyclobutylamino)phenyl]acetic acid (46a) (0.46 g, 2.0 mmol) in DMF (5 ml) was stirred at room temperature for 15 hours. To this mixture were added methanol (20 ml) and aq. 2N HCl (10 ml). The second mixture was refluxed for 1 h. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (3 \times 50 ml). The organic layer was dried over MgSO_4 , concentrated and chromatographed (dichloromethane pure and then dichloromethane:acetone, 95:5) to yield {4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]phenyl}acetic acid methyl ester (46b) [RD146] (0.532 g, 1.124 mmol, 56%), the structure of which is illustrated in Formula 15, as a white powder.

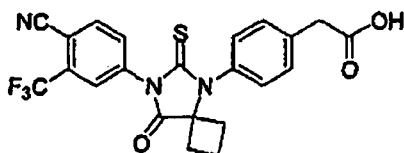


Formula 15

^1H NMR (CDCl_3 , 400 MHz) δ 1.60-1.69 (m, 1H), 2.15-2.25 (m, 1H), 2.50-2.58 (m, 2H), 2.61-2.66 (m, 2H), 3.72 (bs, 5H), 7.27 (d, $J = 8.3$ Hz, 2H), 7.50 (d, $J = 8.3$ Hz, 2H), 7.84 (dd, $J = 8.3, 1.8$ Hz, 1H), 7.94 (d, $J = 8.2$ Hz, 1H), 7.97 (d, $J = 1.6$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.7, 31.4, 44.7, 52.3, 67.4, 109.9, 114.9, 122.0 (q, $J = 272.5$ Hz), 127.0 (q, $J = 4.9$ Hz), 130.0, 131.1, 132.3, 133.0 (q, $J = 33.3$ Hz), 134.1, 135.2, 135.9, 137.2, 171.4, 174.9, 179.9.

46-2) RD147

[00105] A mixture of {4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]phenyl}acetic acid methyl ester (46b) (0.095 g, 0.2 mmol) and a solution of sodium hydroxide (1 ml, 2M) in methanol (2 ml) was stirred at room temperature for 2 hours. The methanol was evaporated. The residue was adjusted to pH 5 by aq. 2M HCl and then the mixture was extracted with ethyl acetate (3 × 10 ml). The organic layer was dried over MgSO₄ and concentrated to dryness to obtain {4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]phenyl}acetic acid (46c) [RD147] (0.087 g, 0.19 mmol, 95%), the structure of which is illustrated in Formula 16.

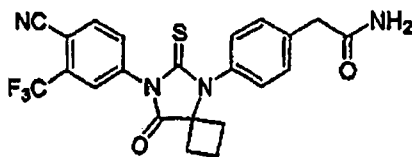


Formula 16

¹H NMR (CDCl₃, 400 MHz) δ 1.60-1.69 (m, 1H), 2.15-2.25 (m, 1H), 2.50-2.64 (m, 4H), 3.73 (s, 2H), 7.26 (d, *J* = 8.3 Hz, 2H), 7.51 (d, *J* = 8.3 Hz, 2H), 7.84 (dd, *J* = 8.3, 1.8 Hz, 1H), 7.95 (d, *J* = 8.2 Hz, 1H), 7.96 (d, *J* = 1.6 Hz, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 13.7, 31.4, 40.2, 40.8, 67.4, 109.9, 114.9, 122.0 (q, *J* = 272.5 Hz), 127.0 (q, *J* = 4.9 Hz), 129.9, 131.2, 132.3, 133.3 (q, *J* = 33.3 Hz), 133.9, 135.2, 136.1, 137.2, 174.1, 174.9, 179.9.

46-3) RD148

[00106] Thionyl chloride (0.238 g, 2 mmol) was added dropwise to a mixture of {4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]phenyl}acetic acid (46c) (0.357 g, 0.777 mmol) in THF (5 ml) cooled to 0 °C. The mixture was stirred for 1 hour at room temperature and then ammonia was bubbled into the mixture. The excess ammonia was condensed by a reflux condenser at -78 °C for 30 minutes and then was allowed to evaporate. The medium was filtered and the filtrate was concentrated and chromatographed (dichloromethane:acetone, 70:30) to yield 2-(4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]phenyl)acetamide (46d) [RD148] (0.345 g, 0.75 mmol, 97%), the structure of which is illustrated in Formula 17, as an off-white powder.

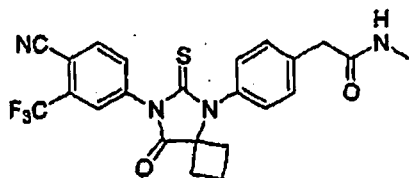


Formula 17

^1H NMR (CDCl_3 , 400 MHz) δ 1.62-1.66 (m, 1H), 2.18-2.23 (m, 1H), 2.49-2.55 (m, 2H), 2.61-2.66 (m, 2H), 3.63 (s, 2H), 5.91 (bs, 1H), 6.10 (bs, 1H), 7.27 (d, $J = 8.1$ Hz, 2H), 7.50 (d, $J = 8.1$ Hz, 2H), 7.83 (dd, $J = 8.3, 1.8$ Hz, 1H), 7.95 (d, $J = 8.2$ Hz, 1H), 7.96 (d, $J = 1.6$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.7, 31.5, 42.5, 67.4, 109.9, 114.9, 121.9 (q, $J = 272.4$ Hz), 127.1 (q, $J = 4.9$ Hz), 130.2, 131.1, 132.2, 133.3 (q, $J = 33.3$ Hz), 134.1, 135.2, 136.8, 137.2, 172.8, 174.8, 180.0.

46-4) RD149

[00107] Thionyl chloride (0.238 g, 2 mmol) was added dropwise to a mixture of {4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]phenyl}acetic acid (46c) (0.357 g, 0.777 mmol) in THF (5 ml) cooled to 0 °C. The mixture was stirred for 1 hour at room temperature and then methylamine (0.5 ml) was added into the mixture. The mixture was stirred for an additional 2 hours. The medium was filtered and the filtrate was concentrated and chromatographed (dichloromethane:acetone, 80:20) to yield *N*-methyl-2-{4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]phenyl}acetamide (46e) [RD149] (0.348 g, 0.738 mmol, 95%), the structure of which is illustrated in Formula 18, as an off-white powder.



Formula 18

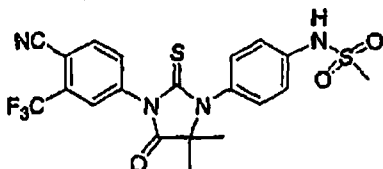
^1H NMR (CDCl_3 , 400 MHz) δ 1.61-1.70 (m, 1H), 2.17-2.31 (m, 1H), 2.50-2.56 (m, 2H), 2.61-2.68 (m, 2H), 2.82 (d, $J = 4.8$ Hz, 3H), 3.62 (s, 2H), 7.27 (d, $J = 8.3$ Hz, 2H), 7.50 (d, $J = 8.3$ Hz, 2H), 7.84 (dd, $J = 8.3, 1.8$ Hz, 1H), 7.95 (d, $J = 8.2$ Hz, 1H), 7.96 (d, $J = 1.6$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.7, 26.6, 31.5, 43.1, 67.4, 110.0, 114.9, 122.0 (q, $J = 272.5$ Hz), 127.1 (q, $J = 4.9$ Hz), 130.2, 131.0, 132.2, 133.3 (q, $J = 33.3$ Hz), 134.1, 135.2, 137.0, 137.1, 170.1, 174.8, 179.9.

Example 47

N-(4-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]phenyl)methanesulfonamide (47a) [RD150]

[00108] A mixture of 4-[3-(4-aminophenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-

trifluoromethylbenzonitrile (2d) (0.02 g, 0.05 mmol), methanesulfonyl chloride (0.009g, 0.075 mmol) and pyridine (0.006 g, 0.075 mmol) in dichloromethane (1 ml) was stirred at room temperature for 15 hours. The medium was washed with water (2 ml) and extracted with ethyl acetate (5 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (HPLC, alumina column) to yield *N*-(4-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxo-imidazolidin-1-yl]phenyl)methanesulfonamide (47a) [RD150] (0.009 g, 0.018 mmol, 36%), the structure of which is illustrated in Formula 2, as a white powder.



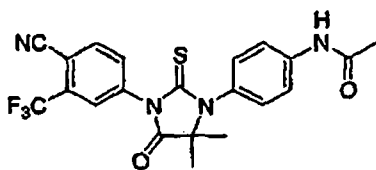
Formula 2

¹H NMR (DMSO-d₆, 400 MHz) δ 1.46 (s, 6H), 3.07 (s, 3H), 7.32 (s, 4H), 8.05 (dd, *J* = 8.2, 1.2 Hz, 1H), 8.26 (d, *J* = 1.2 Hz, 1H), 8.35 (d, *J* = 8.2 Hz, 1H), 10.08 (bs, 1H); ¹³C NMR (DMSO-d₆, 100 MHz) δ 23.3, 40.4, 66.7, 109.0, 115.5, 119.9, 122.6 (q, *J* = 272.2 Hz), 128.5 (q, *J* = 4.7 Hz), 130.8, 131.2, 131.5 (q, *J* = 32.3 Hz), 134.5, 136.6, 138.6, 139.5, 175.4, 180.4.

Example 48

N-(4-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxo-imidazolidin-1-yl]phenyl)acetamide, 48a, [RD151]

[00109] A mixture of 4-[3-(4-aminophenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile (2d) [RD9] (0.008 g, 0.02 mmol), acetyl chloride (0.004g, 0.03 mmol) and triethylamine (0.003 g, 0.03 mmol) in dichloromethane (1 ml) was stirred at 0 °C for 2 hours. The mixture was concentrated and chromatographed (dichloromethane:acetone, 90:10) to yield *N*-(4-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxo-imidazolidin-1-yl]phenyl)acetamide, 48a, [RD151] (0.007 g, 0.016 mmol, 80%), the structure of which is illustrated in Formula 3, as a white powder.



Formula 3

^1H NMR (CDCl_3 , 400 MHz) δ 1.58 (s, 6H), 2.21 (s, 3H), 7.24 (d, $J = 8.6$ Hz, 2H), 7.48 (bs, 1H), 7.69 (d, $J = 8.6$ Hz, 2H), 7.83 (dd, $J = 8.2, 1.9$ Hz, 1H), 7.96 (d, $J = 1.2$ Hz, 1H), 7.97 (d, $J = 8.2$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 23.6, 53.4, 66.4, 110.0, 114.8, 120.7, 122.6 (q, $J = 272.2$ Hz), 127.1 (q, $J = 4.7$ Hz), 129.1, 130.2, 132.2, 133.5 (q, $J = 32.3$ Hz), 135.2, 137.1, 139.2, 168.1, 175.0, 180.0.

Example 49

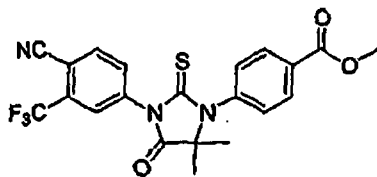
[00110] Concentrated sulfuric acid was slowly added to a mixture of 4-aminobenzoic acid (4 g, 29.2 mmol) in methanol cooled to 0 °C. After the addition, the mixture was stirred at room temperature for 5 hours. The mixture was washed with a saturated solution of sodium bicarbonate and extracted with ethyl acetate. The organic layer was dried over MgSO_4 and concentrated under vacuum to obtain 4-aminobenzoic acid methyl ester (49a) (4.22 g, 27.9 mmol, 96%) as an off-white solid.

[00111] A mixture of 4-aminobenzoic acid methyl ester (0.32 g, 2.12 mmol), acetonecyanohydrin (3ml) and sodium sulfate (1 g) was refluxed for 15 hours. After filtration to remove the sodium sulfate, the filtrate was washed with brine and extracted with ethyl acetate. The organic layer was concentrated and chromatographed (dichloromethane:acetone, 60:40) to yield 4-[(cyanodimethylmethyl)-amino]-benzoic acid methyl ester (49b) (0.398 g, 1.95 mmol, 92%) as a white solid.

49-1) RD152

[00112] A mixture of 4-isothiocyanato-2-trifluoromethylbenzonitrile (1a) (0.228 g, 1 mmol) and 4-[(cyanodimethylmethyl)-amino]-benzoic acid methyl ester (49b) (0.14 g, 0.64 mmol) in DMF (2 ml) was heated under microwave irradiation at 60 °C for 12 hours. To this mixture were added methanol (6 ml) and aq. 2N HCl (2 ml). The second mixture was refluxed for 4 h. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (3 x 30 ml). The organic layer was dried over MgSO_4 , concentrated and chromatographed (dichloromethane; dichloromethane:acetone, 75:25) to yield 4-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxo-imidazolidin-1-yl]benzoic acid methyl ester (49c) [RD152] (0.18 g, 0.4 mmol,

63%), the structure of which is illustrated in Formula 19, as a white powder.

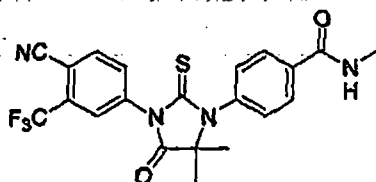


Formula 19

^1H NMR (CDCl_3 , 400 MHz) δ 1.60 (s, 6H), 3.95 (s, 3H), 7.40 (d, $J = 8.6$ Hz, 2H), 7.84 (dd, $J = 8.2, 1.9$ Hz, 1H), 7.96 (d, $J = 1.2$ Hz, 1H), 7.97 (d, $J = 8.2$ Hz, 1H), 8.21 (d, $J = 8.6$ Hz, 2H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 23.8, 52.6, 66.6, 110.3, 114.8, 121.9 (q, $J = 272.7$ Hz), 127.1 (q, $J = 4.7$ Hz), 129.8, 131.2, 131.4, 132.2, 133.5 (q, $J = 32.3$ Hz), 135.3, 137.0, 139.2, 165.9, 174.7, 179.7.

49-2) **RD153**

[00113] A mixture of 4-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]benzoic acid methyl ester (49c) (0.02 g, 0.0435 mmol) and methylamine (2 ml distilled from its 40% aqueous solution) was kept at -20°C for 15 hours. After evaporation of the methylamine, the mixture was chromatographed (dichloromethane:acetone, 80:20) to yield 4-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]-N-methylbenzamide (49d) [RD153] (0.01 g, 0.0224, 51%), the structure of which is illustrated in Formula 20. The ester 4-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]benzoic acid methyl ester (49c) (0.08 g, 0.0179 mmol, 41%) was also recovered.



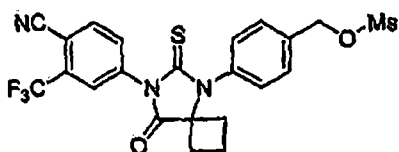
Formula 20

^1H NMR (Acetone- d_6 , 400 MHz) δ 1.60 (s, 6H), 2.90 (d, $J = 4.6$ Hz, 3H), 7.48 (d, $J = 8.6$ Hz, 2H), 7.80 (bs, 1H), 7.99 (d, $J = 8.6$ Hz, 2H), 8.06 (dd, $J = 8.2, 1.8$ Hz, 1H), 8.18 (d, $J = 1.8$ Hz, 1H), 8.25 (d, $J = 8.2$ Hz, 1H); ^{13}C NMR (Acetone- d_6 , 100 MHz) δ 23.8, 54.0, 66.5, 110.3, 114.8, 121.9 (q, $J = 272.7$ Hz), 127.1 (q, $J = 4.7$ Hz), 128.2, 129.9, 133.5 (q, $J = 32.3$ Hz), 135.7, 135.8, 138.2, 138.3, 139.2, 166.0, 174.9, 179.7.

Example 50

50-1) RD154

[00114] A mixture of 4-[8-(4-hydroxymethylphenyl)-5-oxo-7-thioxo-6-azaspiro[3.4]oct-6-yl]-2-trifluoromethyl-benzonitrile (36b) (0.086 g, 0.2 mmol) and methanesulfonyl anhydride (0.07 g, 0.4 mmol) in dichloromethane (1 ml) was stirred at room temperature for 15 hours. The mixture was concentrated and chromatographed (dichloromethane:acetone, 98:2) to yield Methanesulfonic acid 4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]phenylmethyl ester (50a) [RD154] (0.089 g, 0.175 mmol, 88%), the structure of which is illustrated in Formula 22, as a white powder.

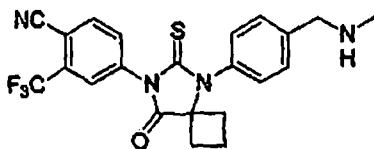


Formula 22

$^1\text{H NMR}$ (CDCl_3 , 400 MHz) δ 1.63-1.70 (m, 1H), 2.17-2.31 (m, 1H), 2.48-2.57 (m, 2H), 2.64-2.70 (m, 2H), 3.04 (s, 3H), 5.30 (s, 2H), 7.37 (d, $J = 8.3$ Hz, 2H), 7.62 (d, $J = 8.3$ Hz, 2H), 7.84 (dd, $J = 8.3, 1.8$ Hz, 1H), 7.97 (d, $J = 8.2$ Hz, 1H), 7.98 (d, $J = 1.6$ Hz, 1H).

50-2) RD155

[00115] Methylamine (0.5 ml) was bubbled into a mixture of Methanesulfonic acid 4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]phenylmethyl ester (50a) (0.059 g, 0.115 mmol) in THF (3 ml) cooled to -78°C . After 1 hour of reaction at -78°C , the mixture was concentrated and chromatographed (dichloromethane:acetone, 95:5; methanol) to yield 4-[5-(4-methylaminomethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl]-2-trifluoromethylbenzonitrile (50b) [RD155] (0.042 g, 0.095 mmol, 82%), the structure of which is illustrated in Formula 23, as a white powder.

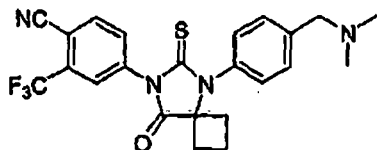


Formula 23

¹H NMR (CDCl₃, 400 MHz) δ 1.57-1.70 (m, 1H), 2.16-2.24 (m, 1H), 2.52 (s, 3H), 2.53-2.57 (m, 2H), 2.60-2.68 (m, 2H), 3.85 (s, 2H), 7.27 (d, *J* = 8.3 Hz, 2H), 7.55 (d, *J* = 8.3 Hz, 2H), 7.84 (dd, *J* = 8.3, 1.8 Hz, 1H), 7.95 (d, *J* = 8.2 Hz, 1H), 7.97 (d, *J* = 1.6 Hz, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 13.7, 31.5, 36.4, 55.6, 67.4, 110.0, 114.9, 122.0 (q, *J* = 272.5 Hz), 127.0 (q, *J* = 4.9 Hz), 129.1, 129.6, 129.8, 132.2, 133.3 (q, *J* = 33.3 Hz), 133.7, 135.2, 142.4, 174.8, 179.9.

50-3) RD156

[00116] A mixture of Methanesulfonic acid 4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]phenylmethyl ester (50a) (0.02 g, 0.039 mmol) and dimethylamine (0.5 ml; distilled from its 40% aqueous solution) in THF (1 ml) was stirred for 2 hours at -78 °C. The mixture was concentrated and chromatographed (dichloromethane:acetone, 95:5;acetone) to yield 4-[5-(4-dimethylaminomethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl]-2-trifluoromethylbenzonitrile (50c) [RD156] (0.017 g, 0.037 mmol, 95%), the structure of which is illustrated in Formula 24, as a white powder.



Formula 24

¹H NMR (CDCl₃, 400 MHz) δ 1.57-1.70 (m, 1H), 2.16-2.24 (m, 1H), 2.32 (s, 6H), 2.55-2.60 (m, 2H), 2.63-2.69 (m, 2H), 3.53 (s, 2H), 7.27 (d, *J* = 8.3 Hz, 2H), 7.55 (d, *J* = 8.3 Hz, 2H), 7.84 (dd, *J* = 8.3, 1.8 Hz, 1H), 7.95 (d, *J* = 8.2 Hz, 1H), 7.97 (d, *J* = 1.6 Hz, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 13.7, 31.5, 45.5, 63.7, 67.4, 110.0, 114.9, 122.0 (q, *J* = 272.5 Hz), 127.0 (q, *J* = 4.9 Hz), 129.1, 129.6, 129.8, 132.2, 133.3 (q, *J* = 33.3 Hz), 133.7, 135.2, 142.4, 174.8, 179.9.

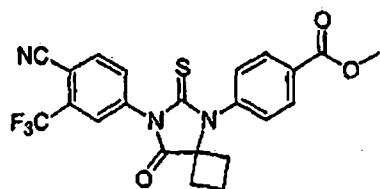
Example 51

[00117] Sodium cyanide (0.245 g, 5 mmol) was added to a mixture of 4-aminobenzoic acid (0.274 g, 2 mmol) and cyclobutanone (0.21 g, 3 mmol) in 90% acetic acid (4.5 ml). The reaction mixture was stirred at room temperature for 15 hours. The mixture was washed with aqueous HCl (pH 2) and extracted with ethyl acetate. The organic layer was dried over magnesium sulfate and concentrated to

dryness under vacuum to yield 4-(1-cyanocyclobutylamino)benzoic acid (51a) (0.426 g, 1.97 mmol, 99%) as a white solid.

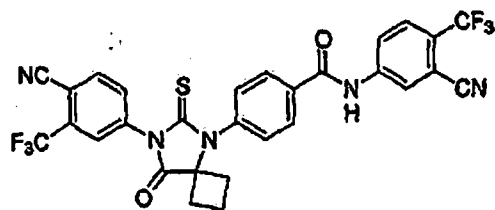
51-1) RD159 and RD160

[00118] A mixture of 4-isothiocyanato-2-trifluoromethylbenzonitrile (1a) (0.51 g, 2.22 mmol) and 4-(1-cyanocyclobutylamino)benzoic acid (51a) (0.343 g, 1.59 mmol) in DMF (2 ml) was heated under microwave irradiation at 60 °C and stirred for 16 hours. To this mixture were added methanol (10 ml) and aq. 2M HCl (5 ml). The second mixture was refluxed for 12 hours. After being cooled to room temperature, the reaction mixture was poured into cold water (20 ml) and extracted with ethyl acetate (3 × 30 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane:acetone, 95:5) to yield 4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]-benzoic acid methyl ester (51b) [RD159] (0.09 g, 0.196 mmol, 12%), the structure of which is illustrated in Formula 25, as a white powder and *N*-(3-cyano-4-trifluoromethylphenyl)-4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]benzamide (51b') [RD160] (0.28 g, 0.45 mmol, 29%), the structure of which is illustrated in Formula 26, as a white powder.



Formula 25

¹H NMR (CDCl₃, 400 MHz) δ 1.67-1.71 (m, 1H), 2.20-2.26 (m, 1H), 2.49-2.57 (m, 2H), 2.66-2.73 (m, 2H), 3.96 (s, 3H), 7.42 (d, *J* = 8.4 Hz, 2H), 7.85 (dd, *J* = 8.3, 1.7 Hz, 1H), 7.97 (d, *J* = 8.3 Hz, 1H), 7.98 (d, *J* = 1.7 Hz, 1H), 8.26 (d, *J* = 8.3 Hz, 2H); ¹³C NMR (CDCl₃, 100 MHz) δ 13.7, 31.6, 52.6, 67.5, 110.1, 114.8, 121.8 (q, *J* = 272.7 Hz), 127.0 (q, *J* = 4.7 Hz), 130.2, 131.4, 131.5, 132.2, 133.4 (q, *J* = 33.2 Hz), 135.2, 137.0, 139.2, 165.9, 174.6, 179.7.

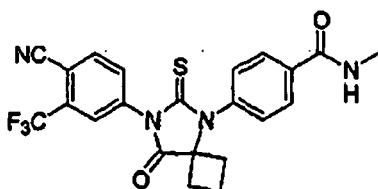


Formula 26

^1H NMR (CDCl_3 , 400 MHz) δ 1.67-1.71 (m, 1H), 2.18-2.26 (m, 1H), 2.50-2.58 (m, 2H), 2.68-2.74 (m, 2H), 7.47 (d, $J = 8.5$ Hz, 2H), 7.83 (d, $J = 8.7$ Hz, 1H), 7.84 (dd, $J = 8.3, 1.9$ Hz, 1H), 7.96 (d, $J = 8.0$ Hz, 1H), 9.97 (d, $J = 1.9$ Hz, 1H), 8.10-8.14 (m, 3H), 8.21 (d, $J = 1.9$ Hz, 1H), 8.88 (s, 1H).

51-2) RD161

[00119] A mixture of 4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]-benzoic acid methyl ester (51b) (0.046 g, 0.1 mmol) and methylamine (1 ml distilled from its 40% aqueous solution) was kept at -20 °C for 15 hours. After evaporation of the methylamine, the mixture was chromatographed (dichloromethane:acetone, 80:20) to yield *N*-methyl-4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]benzamide (51c) [RD161] (0.041 g, 0.085, 84%), the structure of which is illustrated in Formula 27.



Formula 27

^1H NMR (CDCl_3 , 400 MHz) δ 1.63-1.70 (m, 1H), 2.18-2.26 (m, 1H), 2.48-2.56 (m, 2H), 2.65-2.71 (m, 2H), 3.05 (d, $J = 4.8$ Hz, 3H), 6.32 (bs, 1H), 7.39 (d, $J = 8.3$ Hz, 2H), 7.84 (dd, $J = 8.3, 1.7$ Hz, 1H), 7.95-7.98 (m, 4H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.6, 27.0, 31.6, 67.4, 110.3, 114.8, 121.8 (q, $J = 272.7$ Hz), 127.0 (q, $J = 4.7$ Hz), 128.7, 130.3, 132.1, 133.3 (q, $J = 33.2$ Hz), 135.2, 136.3, 137.0, 137.8, 167.2, 174.6, 179.8.

Example 52 [RD162]

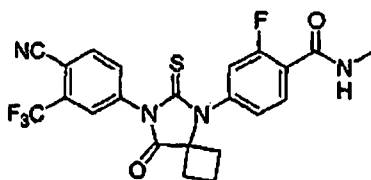
[00120] Thionyl chloride (2.38 g, 20 mmol) was added slowly to a solution of 2-fluoro-4-nitrobenzoic acid (2.97 g, 16 mmol) in DMF (50 ml) cooled at -5 °C. The mixture was stirred for an additional 1 hour at -5 °C. Methylamine (0.62 g, 20 mmol; freshly distilled from its 40% aqueous solution) was added to the reaction medium. The second mixture was stirred for an additional 1 hour. Ethyl acetate (300 ml) was added to the mixture, which was washed with brine (3×150 ml). The organic layer was dried over MgSO_4 , and concentrated to yield *N*-methyl-2-fluoro-4-nitrobenzamide (52a) (2.89

g, 14.6 mmol, 91%) as a yellow solid. ¹H NMR (Acetone d₆, 400 MHz) δ 3.05 (d, *J* = 4.3 Hz, 3H), 6.31 (dd, *J* = 13.5, 2.1 Hz, 1H), 6.40 (dd, *J* = 8.5, 2.1 Hz, 1H), 7.64 (dd, *J* = 8.6, 8.6 Hz, 1H).

[00121] A mixture of *N*-methyl-2-fluoro-4-nitrobenzamide (52a) (2.89 g, 14.6 mmol) and iron (5.04 g, 90 mmol) in ethyl acetate (40 ml) and acetic acid (40 ml) was refluxed for 1 hour. The solid particles were filtered off. The filtrate was washed with water and extracted with ethyl acetate. The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane:acetone, 95:5) to yield *N*-methyl-2-fluoro-4-aminobenzamide (52b) (2.3 g, 13.7 mmol, 94%) as an off-white solid. ¹H NMR (acetone-d₆, 400 MHz) δ 2.86 (d, *J* = 4.3 Hz, 3H), 5.50 (bs, 2H), 6.37 (dd, *J*₁ = 14.7 Hz, *J*₂ = 2.1 Hz, 1H), 6.50 (dd, *J* = 8.5, 2.1 Hz, 1H), 7.06 (bs, 1H), 7.68 (dd, *J* = 8.8 8.8 Hz, 1H); ¹³C NMR (acetone-d₆, 100 MHz) δ 25.8, 99.6 (d, *J* = 13.8 Hz), 109.2 (d, *J* = 12.8 Hz), 110.0 (d, *J* = 1.6 Hz), 132.5 (d, *J* = 4.8 Hz), 153.5 (d, *J* = 12.6 Hz), 162.2 (d, *J* = 242.5 Hz), 164.0 (d, *J* = 3.1 Hz).

[00122] Sodium cyanide (1.47 g, 30 mmol) was added to a mixture of *N*-methyl-2-fluoro-4-aminobenzamide (52b) (1.68 g, 10 mmol) and cyclobutanone (1.4 g, 20 mmol) in 90% acetic acid (20 ml). The reaction mixture was stirred at 80 °C for 24 hours. The mixture was washed with water and extracted with ethyl acetate. The organic layer was dried over magnesium sulfate and concentrated to dryness under vacuum. The solid was washed with a 50:50 mixture of ethyl ether and hexane (10 ml) to remove cyclobutanone cyanohydrin to afford after filtration *N*-methyl-4-(1-cyanocyclobutylamino)-2-fluorobenzamide (52c) (2.19 g, 8.87 mmol, 89%). ¹H NMR (CDCl₃, 400 MHz) δ 1.87-1.95 (m, 1H), 2.16-2.27 (m, 1H), 2.35-2.41 (m, 2H), 2.76-2.83 (m, 2H), 2.97 (d, *J* = 4.4 Hz, 3H), 4.68 (bs, 1H), 6.29 (dd, *J* = 14.3, 1.8 Hz, 1H), 6.48 (dd, *J* = 8.3, 1.8 Hz, 1H), 6.75 (q, *J* = 4.4 Hz, 1H), 7.90 (dd, *J* = 8.3, 8.3 Hz, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 15.7, 26.7, 33.9, 49.4, 100.2 (d, *J* = 29.5 Hz), 110.6, 111.0 (d, *J* = 11.8 Hz), 133.1 (d, *J* = 4.2 Hz), 148.4 (d, *J* = 12.0 Hz), 162.0 (d, *J* = 244.1 Hz), 164.4 (d, *J* = 3.6 Hz).

[00123] A mixture of 4-isothiocyanato-2-trifluoromethylbenzonitrile (1a) (2.16 g, 9.47 mmol) and *N*-methyl-4-(1-cyanocyclobutylamino)-2-fluorobenzamide (52c) (1.303 g, 5.27 mmol) in DMF (20 ml) was heated under microwave irradiation at 80 °C for 16 hours. To this mixture was added methanol (50 ml) and aq. 2N HCl (20 ml). The second mixture was refluxed for 3 hours. After being cooled to room temperature, the reaction mixture was poured into cold water (100 ml) and extracted with ethyl acetate (150 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane:acetone, 95:5) to yield *N*-methyl-4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-2-fluorobenzamide (52d) [RD162] (1.43 g, 3.0 mmol, 57%), the structure of which is illustrated in Formula 28, as a yellow powder.



Formula 28

^1H NMR (CDCl_3 , 400 MHz) δ 1.65-1.75 (m, 1H), 2.18-2.30 (m, 1H), 2.49-2.57 (m, 2H), 2.67-2.73 (m, 2H), 3.07 (d, $J = 4.4$ Hz, 3H), 6.75 (q, $J = 4.6$ Hz, 1H), 7.17 (dd, $J = 11.5, 1.9$ Hz, 1H), 7.26 (dd, $J = 8.3, 1.9$ Hz, 1H), 7.83 (dd, $J = 8.2, 2.0$ Hz, 1H), 7.95 (d, $J = 1.8$ Hz, 1H), 7.97 (d, $J = 8.3$ Hz, 1H), 8.30 (dd, $J = 8.3, 8.3$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 13.6, 27.0, 31.7, 67.4, 110.3, 114.8, 118.2, 118.5, 121.9 (q, $J = 272.7$ Hz), 126.6, 127.0 (q, $J = 4.8$ Hz), 132.1, 133.3 (q, $J = 33.2$ Hz), 133.8, 135.3, 136.8, 139.1 (d, $J = 10.9$ Hz), 160.5 (d, $J = 249.1$ Hz), 162.7 (d, $J = 3.3$ Hz), 174.3, 179.8; ^{19}F NMR (CDCl_3 , 100 MHz) δ -111.13, -62.58.

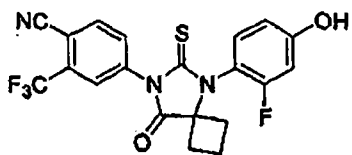
Example 53 [RD163]

[00124] A mixture of 4-nitro-3-fluorophenol (0.314 g, 2 mmol) and iron (0.56 g, 10 mmol) in ethyl acetate (4 ml) and acetic acid (2 ml) was refluxed for 3 hour. The solid particles were filtered off. The filtrate was washed with water and extracted with ethyl acetate. The organic layer was dried over MgSO_4 , concentrated to yield 4-amino-3-fluorophenol (53a) (0.25 g, 19.6 mmol, 98%) as a brown solid. ^1H NMR (CDCl_3 , 400 MHz) δ 6.48-6.58 (m, 2H), 6.61-6.70 (m, 1H), 7.87 (bs, 3H).

[00125] Sodium cyanide (0.194 g, 4 mmol) was added to a mixture of 4-amino-3-fluorophenol (0.29 g, 2.28 mmol) and cyclobutanone (0.175 g, 2.5 mmol) in 90% acetic acid (3 ml). The reaction mixture was stirred at room temperature for 15 hours. The medium was washed with water and extracted with ethyl acetate. The organic layer was dried over magnesium sulfate, concentrated and chromatographed (dichloromethane:acetone, 90:10) to yield 1-(2-fluoro-4-hydroxyphenylamino)-cyclobutanecarbonitrile (53b) (0.271 g, 1.31 mmol, 58%) as an off-white solid. ^1H NMR (CDCl_3 , 400 MHz) δ 2.13-2.20 (m, 2H), 2.36-2.41 (m, 2H), 2.70-2.75 (m, 2H), 4.00 (bs, 1H), 6.46 (bs, 1H), 6.52 (ddd, $J_1 = 2.2$ Hz, $J_2 = 0.65$ Hz, $J_3 = 0.22$ Hz, 1H), 6.57 (d, $J = 2.3$ Hz), 6.62 (dd, $J_1 = 3.0$ Hz, $J_2 = 0.67$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 15.7, 34.1, 50.9, 104.0 (d, $J = 21.9$ Hz), 111.0 (d, $J = 3.4$ Hz), 115.8 (d, $J = 3.7$ Hz), 121.8, 125.3 (d, $J = 12.3$ Hz), 150.1 (d, $J = 10.4$ Hz), 152.8 (d, $J = 239.3$ Hz).

[00126] A mixture of 4-isothiocyanato-2-trifluoromethylbenzonitrile (1a) (0.228 g, 1.0 mmol) and 1-(2-fluoro-4-hydroxyphenylamino)-cyclobutanecarbonitrile (53b) (0.145 g, 0.7 mmol) in dry DMF (2 ml) was stirred at room temperature for 24 hours. To this mixture were added methanol (10 ml) and

aq. 2M HCl (2 ml). The second mixture was refluxed for 1 hour. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (50 ml). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane pure and then dichloromethane:acetone, 90:10) to yield 4-[5-(2-fluoro-4-hydroxyphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl]-2-trifluoromethylbenzonitrile (53c) [RD163] (0.17 g, 0.39 mmol, 56%), the structure of which is illustrated in Formula 29, as a off-white powder.



Formula 29

¹H NMR (CDCl₃, 400 MHz) δ 1.66-1.75 (m, 1H), 2.18-2.28 (m, 1H), 2.42-2.50 (m, 1H), 2.54-2.67 (m, 3H), 6.76 (d, *J* = 2.2 Hz, 2H), 7.15 (t, *J* = 2.1 Hz, 1H), 7.35 (bs, 1H), 7.87 (dd, *J*₁ = 8.2 Hz, *J*₂ = 1.8 Hz, 1H), 7.97 (d, *J* = 8.2 Hz, 1H), 7.98 (d, *J* = 1.8 Hz, 1H); ¹³C NMR (CDCl₃, 100 MHz) δ 13.8, 31.0, 67.6, 104.8 (d, *J* = 22.3 Hz), 109.8, 112.6, 114.4 (d, *J* = 13.1 Hz), 114.9, 121.9 (q, *J* = 272.8 Hz), 127.1 (q, *J* = 4.8 Hz), 132.0, 132.3, 133.5 (q, *J* = 33.3 Hz), 135.3, 137.2, 159.3 (d, *J* = 11.2 Hz), 159.6 (d, *J* = 249.7 Hz), 175.2, 180.5; ¹⁹F NMR (CDCl₃, 100 MHz) δ -117.5, -62.49.

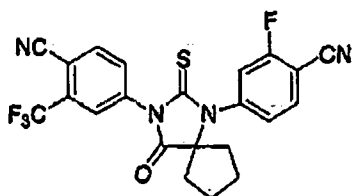
Example 54 [RD168]

[00127] A mixture of 4-nitro-2-fluorobenzonitrile (1.83 g, 5 mmol) and iron (1.68 g, 6 mmol) in a mixture of acetic acid (40 ml) and ethyl acetate (40 ml) was refluxed for 2 hours. The solid was filtered off and the filtrate was washed with water and extracted with ethyl acetate. The organic layer was dried over magnesium sulfate, concentrated and chromatographed (dichloromethane:acetone, 95:5) to yield 4-amino-2-fluorobenzonitrile (54a) (0.653 g, 4.8 mmol, 96%).

[00128] Sodium cyanide (0.74 g, 15 mmol) was added to a mixture of 4-amino-2-fluorobenzonitrile (1.36 g, 10 mmol) and cyclopentanone (1.26 g, 15 mmol) in 90% acetic acid (10 ml). The reaction mixture was stirred at room temperature for 3 hours and then the medium was heated to 80 °C and stirred for an additional 5 hours. The medium was washed with water and extracted with ethyl acetate. The organic layer was dried over magnesium sulfate, concentrated and chromatographed (dichloromethane:acetone, 97:3) to yield 4-(1-cyanocyclopentylamino)-2-fluorobenzonitrile (54b) (2.07 g, 9.03 mmol, 90%) as a yellow solid. ¹H NMR (CDCl₃, 400 MHz) δ 1.69-1.91 (m, 4H), 2.13-2.18 (m,

2H), 2.37-2.42 (m, 2H), 5.08 (bs, 1H), 6.54-6.62 (m, 2H), 7.39 (t, $J = 7.3$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 23.7, 39.8, 56.8, 89.6 (d, $J = 15.8$ Hz), 101.2 (d, $J = 23.8$ Hz), 110.9, 115.2, 120.8, 134.1 (d, $J = 2.4$ Hz), 150.3 (d, $J = 11.2$ Hz), 164.5 (d, $J = 254.1$ Hz).

[00129] A mixture of 4-isothiocyanato-2-trifluoromethylbenzonitrile (1a) (0.171 g, 0.75 mmol) and 4-(1-cyanocyclopentylamino)-2-fluorobenzonitrile (54b) (0.115 g, 0.5 mmol) in dry DMF (1 ml) was heated under microwave irradiation at 60 °C for 48 hours. To this mixture were added methanol (3 ml) and aq 2M HCl (2 ml). The second mixture was refluxed for 1 hour. After being cooled to room temperature, the reaction mixture was poured into cold water (10 ml) and extracted with ethyl acetate (15 ml). The organic layer was dried over MgSO_4 , concentrated and chromatographed (dichloromethane:acetone, 98:2) to yield 4-[1-(4-cyano-3-fluorophenyl)-4-oxo-2-thioxo-1,3-diazaspiro[4.4]non-3-yl]-2-trifluoromethylbenzonitrile (54c) [RD168] (0.017 g, 0.037 mmol, 7%), of which the structure is illustrated in Formula 30, as an off-white powder.

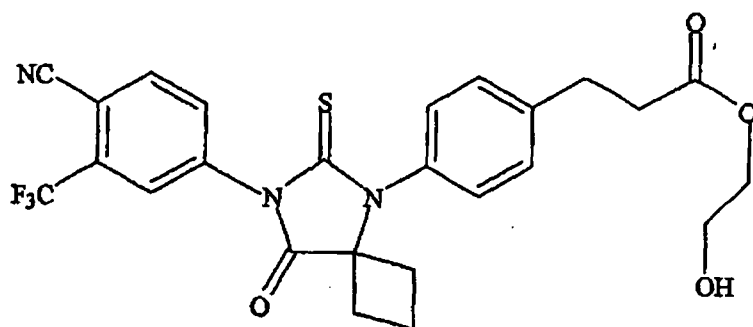


Formula 30

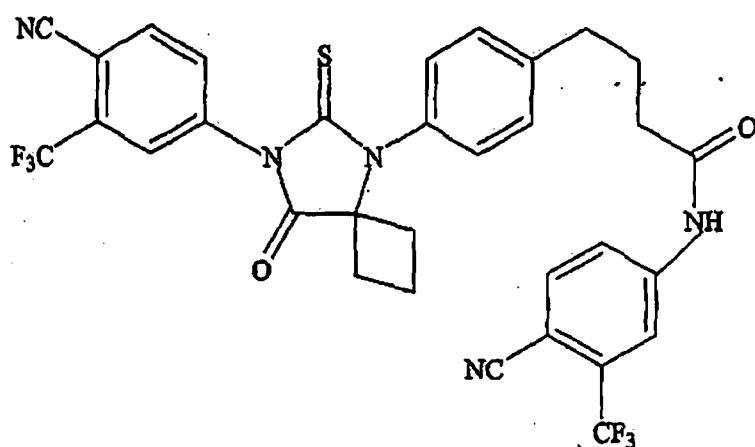
^1H NMR (CDCl_3 , 400 MHz) δ 1.53-1.63 (m, 2H), 1.89-2.00 (m, 2H), 2.09-2.16 (m, 2H), 2.35-2.42 (m, 2H), 7.27-7.37 (m, 2H), 7.78-7.90 (m, 3H), 7.95 (d, $J = 1.8$ Hz, 1H), 7.97 (d, $J = 8.3$ Hz, 1H); ^{13}C NMR (CDCl_3 , 100 MHz) δ 25.2, 36.5, 75.3, 103.2 (d, $J = 15.3$ Hz), 110.4, 112.8, 114.7, 119.2 (d, $J = 20.7$ Hz), 121.9 (q, $J = 272.8$ Hz), 127.0 (q, $J = 4.8$ Hz), 132.1, 133.7 (q, $J = 33.2$ Hz), 134.6, 135.3, 135.8, 136.8, 141.8 (d, $J = 9.5$ Hz), 163.4 (d, $J = 261.5$ Hz), 175.3, 180.1.

Example 55 [RD136 and RD142]

[00130] Additional diarylhydantoin compounds can be synthesized, including the following compounds illustrated in Formulas 35 and 36.



Formula 35 [RD136]

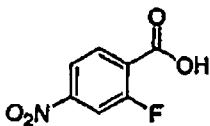


Formula 36 [RD142]

Example 56 [RD162]

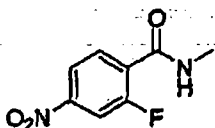
[00131] In the following, air or moisture sensitive reactions were conducted under argon atmosphere using oven-dried glassware and standard syringe/septa techniques. The reactions were monitored with a SiO₂ TLC plate under UV light (254 nm) followed by visualization with a *p*-anisaldehyde or ninhydrin staining solution. Column chromatography was performed on silica gel 60. ¹H NMR spectra were measured at 400 MHz in CDCl₃ unless stated otherwise and data were reported as follows in ppm (δ) from the internal standard (TMS, 0.0 ppm): chemical shift (multiplicity, integration,

coupling constant in Hz.).



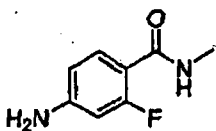
Formula 37

[00132] Periodic acid (1.69 g, 7.41 mmol) was dissolved in acetonitrile (25 mL) by vigorous stirring, and then chromium trioxide (0.16 g, 1.60 mmol) was dissolved into the solution. 2-Fluoro-4-nitrotoluene (0.33 g, 2.13 mmol) was added to the above solution with stirring. A white precipitate formed immediately with exothermic reaction. After 1 h of stirring, the supernatant liquid of the reaction mixture was decanted to a flask, and the solvent was removed by evaporation. The residues were extracted with methylene chloride (2×30 mL) and water (2×30 mL). The organic layer was dried over MgSO₄, and concentrated to give 2-Fluoro-4-nitrobenzoic acid (Formula 37) (0.32 mg, 81%) as a white solid. ¹H NMR δ 8.06 (ddd, 1 H, J=9.9, 2.2 and 0.3), 8.13 (ddd, 1 H, J=8.6, 2.2 and 0.9), 8.25 (ddd, 1 H, J=8.6, 7.0 and 0.3).



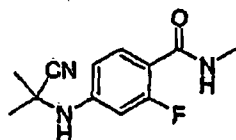
Formula 38

[00133] Thionyl chloride (0.15 g, 1.30 mmol) was added slowly to a solution of 2-fluoro-4-nitrobenzoic acid (Formula 37) (0.20 g, 1.10 mmol) in DMF (5 mL) cooled at -5 °C. The mixture was stirred for an additional 1 hour at -5 °C. Excess methylamine (freshly distilled from its 40% aqueous solution) was added to the reaction medium. The second mixture was stirred for an additional 1 hour. Ethyl acetate (50 mL) was added to the mixture, which was washed with brine (2 × 50 ml). The organic layer was dried over MgSO₄, and concentrated to yield *N*-Methyl-2-fluoro-4-nitrobenzamide (Formula 38) (0.18 g, 85%) as a yellowish solid. ¹H NMR (acetone-*d*₆) δ 3.05 (d, 3 H, J=4.3), 6.31 (dd, 1 H, J=13.5 and 2.1), 6.40 (dd, 1H, J=8.6 and 2.1), 7.64 (dd, 1H, J= 8.6 and 8.6).



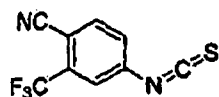
Formula 39

[00134] A mixture of *N*-Methyl-2-fluoro-4-nitrobenzamide (Formula 38) (0.18 g, 0.91 mmol) and iron (0.31 g, 5.60 mmol) in ethyl acetate (5 mL) and acetic acid (5 mL) was refluxed for 1 h. The solid particles were filtered off. The filtrate was washed with water and extracted with ethyl acetate. The organic layer was dried over MgSO₄, concentrated and the residue was purified with SiO₂ column chromatography (dichloromethane:acetone, 95:5) to give *N*-Methyl-2-fluoro-4-aminobenzamide (Formula 39) (0.14 g, 92%) as an off-white solid. ¹H NMR (acetone-*d*₆) δ 2.86 (d, 3 H, *J*=4.3), 5.50 (br s, 2 H), 6.37 (dd, 1 H, *J*=14.7 and 2.1), 6.50 (dd, 1H, *J*=8.6 and 2.1), 7.06 (br s, 1H), 7.68 (dd, 1H, *J*=8.8 and 8.8).



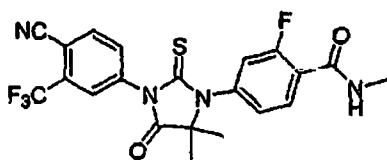
Formula 40

[00135] A mixture of *N*-Methyl-2-fluoro-4-aminobenzamide (Formula 39) (96 mg, 0.57 mmol), acetone cyanohydrin (0.3 mL, 3.14 mmol) and magnesium sulfate (50 mg) was heated to 80 °C and stirred for 12 h. To the medium was added ethyl acetate (25 mL) and then washed with water (2 × 25 mL). The organic layer was dried over MgSO₄ and concentrated and the residue was purified with SiO₂ column chromatography (dichloromethane:acetone, 95:5) to give *N*-Methyl-2-fluoro-4-(1,1-dimethylcyanomethyl)-aminobenzamide (Formula 40) (101 mg, 75%) as a white solid. ¹H NMR δ 1.74 (s, 6 H), 2.98 (dd, 3 H, *J*=4.8 and 1.1), 6.58 (dd, 1 H, *J*=14.6 and 2.3), 6.63 (dd, 1 H, *J*=8.7 and 2.3), 6.66 (br s, 1 H), 7.94 (dd, 1 H, *J*=8.7 and 8.7).



Formula 41

[00136] 4-Amino-2-trifluoromethylbenzonitrile (2.23 g, 12 mmol) was added portionwise over 15 min into a well-stirred heterogeneous mixture of thiophosgene (1 mL, 13 mmol) in water (22 mL) at room temperature. Stirring was continued for an additional 1 h. The reaction medium was extracted with chloroform (3 × 15 mL). The combined organic phase was dried over MgSO₄ and evaporated to dryness under reduced pressure to yield desired product 4-Isothiocyanato-2-trifluoromethylbenzonitrile (Formula 41) as brownish solid and was used as such for the next step (2.72 g, 11.9 mmol, 99%). ¹H NMR δ 7.49 (dd, 1 H, *J*=8.3 and 2.1), 7.59 (d, 1 H, *J*=2.1), 7.84 (d, 1 H, *J*=8.3).

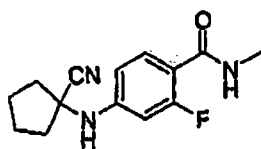


RD162' (Formula 42)

56-1) RD162'

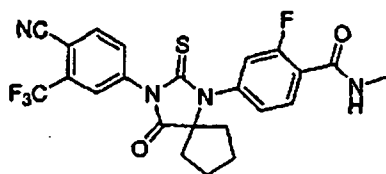
[00137] A mixture of *N*-Methyl-2-fluoro-4-(1,1-dimethyl-cyanomethyl)-aminobenzamide (Formula 40) (30 mg, 0.13 mmol) and 4-Isothiocyanato-2-trifluoromethylbenzonitrile (Formula 41) (58 mg, 0.26 mmol) in DMF (1 mL) was heated under microwave irradiation at 100 °C for 11 hours. To this mixture was added methanol (20 mL) and aq. 1 N HCl (5 mL). The second mixture was refluxed for 1.5 h. After being cooled to room temperature, the reaction mixture was poured into cold water (50 mL) and extracted with ethyl acetate (50 mL). The organic layer was dried over MgSO₄, concentrated and the residue was purified with SiO₂ column chromatography (dichloromethane:acetone, 95:5) to give RD162' (Formula 42) (15 mg, 25%) as a colorless crystal. ¹H NMR δ 1.61 (s, 6 H), 3.07 (d, 3 H, *J*=4.1), 6.71 (m, 1 H), 7.15 (dd, 1H, *J*=11.7 and 2.0), 7.24 (dd, 1H, *J*=8.4 and 2.0), 7.83 (dd, 1H, *J*=8.2 and 2.1), 7.95 (d, 1H, *J*=2.1), 7.99 (d, 1H, *J*=8.2), 8.28 (dd, 1H, *J*=8.4 and 8.4).

Example 57



Formula 43

[00138] A mixture of *N*-Methyl-2-fluoro-4-aminobenzamide (Formula 39) (62 mg, 0.37 mmol), cyclopentanone (0.07 mL, 0.74 mmol) and TMS-CN (0.1 mL, 0.74 mmol) was heated to 80 °C and stirred for 13 h. To the medium was added ethyl acetate (2 × 20 mL) and then washed with water (2 × 20 mL). The organic layer was dried over MgSO₄ and concentrated and the residue was purified with silica gel column chromatography (dichloromethane:acetone, 95:5) to give *N*-Methyl 2-fluoro-4-(1-cyanocyclopentyl)aminobenzamide (Formula 43) (61 mg, 63%) as a white solid. ¹H NMR δ 7.95 (dd, 1H, *J* = 8.8, 8.8 Hz), 6.65 (br s, 1H), 6.59 (dd, 1H, *J* = 8.8, 2.3 Hz), 6.50 (dd, 1H, *J* = 14.6, 2.3 Hz), 4.60 (br s, 1H), 2.99 (dd, 3H, *J* = 4.8, 1.1 Hz), 2.36-2.45 (m, 2H), 2.10-2.18 (m, 2H), 1.82-1.95 (m, 4H).

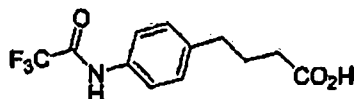


RD162" (Formula 44)

57-1) RD162"

[00139] A mixture of *N*-Methyl 2-fluoro-4-(1-cyanocyclopentyl)aminobenzamide (Formula 43) (57 mg, 0.22 mmol) and 4-isothiocyanato-2-trifluoromethyl benzonitrile (0.15 g, 0.65 mmol) in DMF (3 mL) was heated under microwave irradiation (open vessel) at 130 °C for 12 hours. To this mixture was added methanol (20 mL) and *aq.* 1 N HCl (5 mL). The second mixture was refluxed for 1.5 h. After being cooled to room temperature, the reaction mixture was poured into cold water (50 mL) and extracted with ethyl acetate (50 mL). The organic layer was dried over MgSO₄, concentrated and the residue was purified with silica gel column chromatography (dichloromethane:acetone, 95:5) to give 4-(3-(4-Cyano-3-(trifluoromethyl)phenyl)-4-oxo-2-thioxo-1,3-diazaspiro[4.4]nonan-1-yl)-2-fluoro-*N*-methylbenzamide, RD162" (Formula 44) (8 mg, 7%) as a pale yellowish solid. ¹H NMR δ 8.28 (dd, 1H, *J* = 8.4, 8.4 Hz), 7.98 (d, 1H, *J* = 8.3 Hz), 7.96 (d, 1H, *J* = 1.8 Hz), 7.84 (dd, 1H, *J* = 8.3, 1.8 Hz), 7.27 (dd, 1H, *J* = 8.4, 1.8 Hz), 7.17 (dd, 1H, *J* = 11.7, 1.8 Hz), 6.67-6.77 (m, 1H), 3.07 (d, 3H, *J* = 4.3 Hz), 2.32-2.41 (m, 2H), 2.13-2.21 (m, 2H), 1.85-1.96 (m, 2H), 1.49-1.59 (m, 2H).

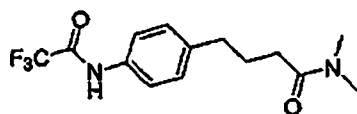
Example 58



Formula 45

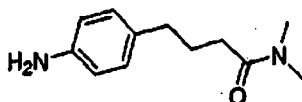
[00140] Trifluoroacetic anhydride (0.85 mL, 6.14 mmol) was added to a solution of 4-(4-aminophenyl)butyric acid (0.5 g, 2.79 mmol) in chloroform (10 mL) at 0 °C. The mixture was warmed to room temperature and stirred for 3 hours. The mixture was partitioned with chloroform (20 mL) and water (20 mL). The organic layer was dried over MgSO₄, concentrated and the residue was purified with silica gel column chromatography (dichloromethane:acetone, 9:1) to give 4-[4-(2,2,2-Trifluoroacetylamino)phenyl]butanoic acid (Formula 45) (0.53 g, 69%). ¹H NMR δ 7.81 (br s, 1H), 7.48

(d, 2H, $J = 8.5$ Hz), 7.22 (d, 2H, $J = 8.5$ Hz), 2.68 (t, 2H, $J = 7.5$ Hz), 2.38 (t, 2H, $J = 7.5$ Hz), 1.96 (p, 2H, $J = 7.5$ Hz).



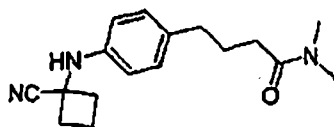
Formula 46

[00141] Thionyl chloride (71 mg, 0.60 mmol) was added slowly to a solution of 4-[4-(2,2,2-Trifluoroacetyl-amino)phenyl]butanoic acid (Formula 45) (0.15 g, 0.55 mmol) in DMF (5 mL) cooled at -5 °C. The mixture was stirred for an additional 1 hour at -5 °C. Excess dimethylamine (freshly distilled from its 40% aqueous solution) was added to the reaction medium. The second mixture was stirred for an additional 1 hour. Ethyl acetate (50 mL) was added to the mixture, which was washed with brine (2×50 mL). The organic layer was dried over $MgSO_4$, and concentrated to yield *N,N*-Dimethyl 4-[4-(2,2,2-Trifluoroacetyl-amino)phenyl]butanamide (Formula 46) (0.17 g, quant.) as a yellowish solid. 1H NMR δ 9.70 (br s, 1H), 7.55 (d, 2H, $J = 8.6$ Hz), 7.11 (d, 2H, $J = 8.6$ Hz), 2.91 (s, 3H), 2.89 (s, 3H), 2.60 (t, 2H, $J = 7.7$ Hz), 2.27 (t, 2H, $J = 7.7$ Hz), 1.89 (p, 2H, $J = 7.7$ Hz).



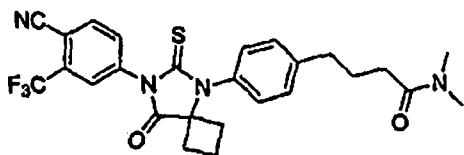
Formula 47

[00142] 1 N NaOH solution (3 mL) was added to a solution of *N,N*-Dimethyl 4-[4-(2,2,2-Trifluoroacetyl-amino)phenyl]butanamide (Formula 46) (0.17 g, 0.55 mmol) in methanol (2 mL) at room temperature. The mixture was stirred for 14 hour. The mixture was partitioned with chloroform (25 mL) and water (25 mL). The organic layer was dried over $MgSO_4$, and concentrated and the residue was purified with silica gel column chromatography (dichloromethane:acetone, 9:1) to give *N,N*-Dimethyl 4-(4-aminophenyl)butanamide (Formula 47) (74 mg, 66%) as a white solid. 1H NMR δ 6.97 (d, 2H, $J = 8.3$ Hz), 6.61 (d, 2H, $J = 8.3$ Hz), 3.56 (br s, 2H), 2.92 (s, 6 H), 2.56 (t, 2H, $J = 7.7$ Hz), 2.28 (t, 2H, $J = 7.7$ Hz), 1.91 (p, 2H, $J = 7.7$ Hz).



Formula 48

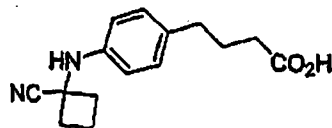
[00143] A mixture of *N,N*-Dimethyl 4-(4-aminophenyl)butanamide (Formula 47) (74 mg, 0.36 mmol), cyclobutanone (54 mg, 0.78 mmol) and TMSCN (77 mg, 0.78 mmol) was heated to 80 °C and stirred for 15 h. To the medium was added ethyl acetate (2 × 20 mL) and then washed with water (2 × 20 mL). The organic layer was dried over MgSO₄ and concentrated and the residue was purified with silica gel column chromatography (dichloromethane:acetone, 9:1) to give *N,N*-Dimethyl 4-[4-(1-cyanocyclobutylamino)phenyl]butanamide (Formula 48) (58 mg, 57%) as a white solid. ¹H NMR δ 7.07 (d, 2H, *J* = 8.5 Hz), 6.59 (d, 2H, *J* = 8.5 Hz), 3.94 (br s, 1H), 2.94 (s, 3H), 2.93 (s, 3H), 2.75-2.83 (m, 2H), 2.60 (t, 2H, *J* = 7.6 Hz), 2.33-2.42 (m, 2H), 2.30 (t, 2H, *J* = 7.6 Hz), 2.11-2.28 (m, 2H), 1.93 (p, 2H, *J* = 7.6 Hz).



RD169 Formula 49

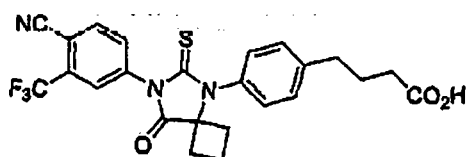
[00144] A mixture of *N,N*-Dimethyl 4-[4-(1-cyanocyclobutylamino)phenyl]butanamide (Formula 48) (58 mg, 0.20 mmol) and 4-isothiocyanato-2-trifluoromethyl benzonitrile (74 mg, 0.32 mmol) in DMF (3 mL) was heated under reflux for 2 hours. To this mixture was added methanol (20 mL) and aq. 1 N HCl (5 mL). The second mixture was refluxed for 1.5 h. After being cooled to room temperature, the reaction mixture was poured into cold water (50 mL) and extracted with ethyl acetate (50 mL). The organic layer was dried over MgSO₄, concentrated and the residue was purified with silica gel column chromatography (dichloromethane:acetone, 95:5) to give 4-(4-(7-(4-Cyano-3-(trifluoromethyl)phenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]octan-5-yl)phenyl)-*N,N*-dimethylbutanamide, RD169 (Formula 49) (44 mg, 42%) as a pale yellowish solid. ¹H NMR δ 7.98 (s, 1H), 7.97 (d, 1H, *J* = 8.2 Hz), 7.86 (d, 1H, *J* = 8.2 Hz), 7.42 (d, 2H, *J* = 8.3 Hz), 7.22 (d, 2H, *J* = 8.3 Hz), 2.99 (s, 3H), 2.96 (s, 3H), 2.78 (t, 2H, *J* = 7.5 Hz), 2.62-2.70 (m, 2H), 2.52-2.63 (m, 2H), 2.40 (t, 2H, *J* = 7.5 Hz), 2.15-2.30 (m, 1H), 2.04 (p, 2H, *J* = 7.5 Hz), 1.62-1.73 (m, 1H).

Example 59



Formula 50

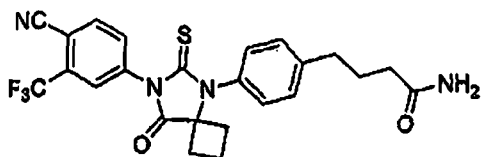
[00145] A mixture of 4-(4-aminophenyl)butyric acid (0.20 g, 1.12 mmol), cyclobutanone (0.17 mL, 2.23 mmol) and TMSCN (0.30 mL, 2.23 mmol) was heated to 80 °C and stirred for 13 h. To the medium was added ethyl acetate (2 × 30 mL) and then washed with water (2 × 30 mL). The organic layer was dried over MgSO₄ and concentrated and the residue was purified with silica gel column chromatography (dichloromethane:acetone, 9:1) to give 4-[4-(1-Cyanocyclobutylamino)phenyl]butanoic acid (Formula 50) (0.21 g, 74%) as a yellowish solid. ¹H NMR δ 7.06 (d, 2H, *J* = 8.6 Hz), 6.59 (d, 2H, *J* = 8.6 Hz), 2.75-2.83 (m, 2H), 2.59 (t, 2H, *J* = 7.5 Hz), 2.37 (t, 2H, *J* = 7.5 Hz), 2.33-2.42 (m, 2H), 2.11-2.28 (m, 2H), 1.92 (p, 2H, *J* = 7.5 Hz).



Formula 51

[00146] A mixture of 4-[4-(1-Cyanocyclobutylamino)phenyl]butanoic acid (Formula 50) (0.21 g, 0.83 mmol) and 4-isothiocyanato-2-trifluoro benzonitrile (0.25 g, 1.08 mmol) in toluene (10 mL) was heated under reflux for 1 hours. To this mixture was added *aq.* 1 N HCl (5 mL). The second mixture was refluxed for 1.5 h. After being cooled to room temperature, the reaction mixture was poured into cold water (50 mL) and extracted with ethyl acetate (50 mL). The organic layer was dried over MgSO₄, concentrated and the residue was purified with silica gel column chromatography (dichloromethane:acetone, 95:5) to give 4-(4-(7-(4-Cyano-3-(trifluoromethyl)phenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]octan-5-yl)phenyl)butanoic acid, RD141 (Formula 51) (60 mg, 15%). ¹H NMR δ 7.98 (d, 1H, *J* = 1.8 Hz), 7.97 (d, 1H, *J* = 8.3 Hz), 7.86 (dd, 1H, *J* = 8.3, 1.8 Hz), 7.42 (d, 2H, *J* = 8.5 Hz), 7.24 (d, 2H, *J* = 8.5 Hz), 2.79 (t, 2H, *J* = 7.5 Hz), 2.62-2.68 (m, 2H), 2.51-2.59 (m, 2H), 2.47 (t, 2H, *J* = 7.5 Hz), 2.14-2.26 (m, 1H), 2.06 (p, 2H, *J* = 7.5 Hz), 1.60-1.70 (m, 1H).

Example 60

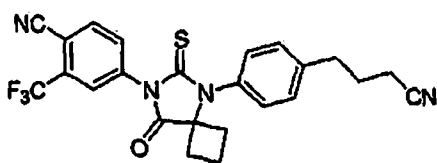


RD130 Formula 52

[00147] To a solution of 4-(4-(7-(4-Cyano-3-(trifluoromethyl)phenyl)-8-oxo-6-thioxo-5,7-

diazaspiro[3.4]octan-5-yl)phenyl)butanoic acid, RD141 (Formula 51) (60 mg, 0.12 mmol) in DMF (3 mL) was added thionyl chloride (0.01 mL, 0.15 mmol) at 0 °C. The mixture was stirred at 0 °C for 1 hour. Then ammonia was bubbled into the mixture. The mixture was partitioned with ethyl acetate (25 mL) and water (25 mL). The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane:acetone, 70:30) to yield 4-(4-(7-(4-Cyano-3-(trifluoromethyl)phenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]octan-5-yl)phenyl)butanamide, RD130 (Formula 52) (37 mg, 61%) as a white powder. ¹H NMR δ 7.97 (d, 1H, *J* = 1.8 Hz), 7.95 (d, 1H, *J* = 8.3 Hz), 7.85 (dd, 1H, *J* = 8.3 Hz), 7.39 (d, 2H, *J* = 8.3 Hz), 7.22 (d, 2H, *J* = 8.3 Hz), 5.59 (br s, 2H), 2.77 (t, 2H, *J* = 7.5 Hz), 2.62-2.68 (m, 2H), 2.51-2.59 (m, 2H), 2.31 (t, 2H, *J* = 7.5 Hz), 2.16-2.25 (m, 1H), 2.05 (p, 2H, *J* = 7.5 Hz), 1.57-1.70 (m, 1H).

Example 61

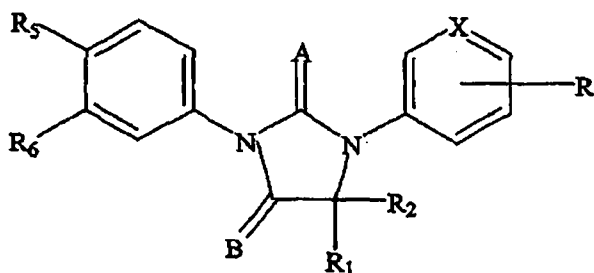


RD170 Formula 53

A solution of DMSO (0.01 mL, 0.12 mmol) in dry dichloromethane (1 mL) was added to a stirred solution of oxalyl chloride (0.01 mL, 0.09 mmol) in dry dichloromethane (2 mL) at -78 °C. After 15 min, a dichloromethane solution of 4-(4-(7-(4-Cyano-3-(trifluoromethyl)phenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]octan-5-yl)phenyl)butanamide, RD130 (Formula 52) (35 mg, 0.07 mmol) was added to the reaction mixture. Stirring was continued for 20 min at -78 °C, and then triethylamine (0.03 mL, 0.22 mmol) was added. After 30 min at -78 °C, the reaction mixture was warmed to room temperature and then reaction was quenched with saturated aq. NH₄Cl solution. The reaction mixture was diluted with dichloromethane, and extracted with dichloromethane. The organic layer was dried over MgSO₄, concentrated and chromatographed (dichloromethane:acetone, 95:5) to yield 4-(5-(4-(3-Cyanopropyl)phenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]octan-7-yl)-2-(trifluoromethyl)benzonitrile, RD170 (Formula 53) (29 mg, 87%) as a viscous oil. ¹H NMR δ 7.98 (d, 1H, *J* = 1.8 Hz), 7.98 (d, 1H, *J* = 8.3 Hz), 7.86 (dd, 1H, *J* = 8.3, 1.8 Hz), 7.43 (d, 2H, *J* = 8.4 Hz), 7.27 (d, 2H, *J* = 8.4 Hz), 2.90 (t, 2H, *J* = 7.3 Hz), 2.63-2.73 (m, 2H), 2.52-2.62 (m, 2H), 2.42 (t, 2H, *J* = 7.3 Hz), 2.18-2.30 (m, 1H), 2.07 (p, 2H, *J* = 7.3 Hz), 1.63-1.73 (m, 1H).

[00148] One skilled in the art could modify and/or combine the syntheses described herein to make other diarylhydantoin compounds.

[00149] Inventive compounds also include those with the following formulas.



Where R is selected from hydrogen, aryl, substituted aryl, alkyl, substituted alkyl, alkenyl, substituted alkenyl, alkynyl, substituted alkynyl, halogenated alkyl, halogenated alkenyl, halogenated alkynyl, arylalkyl, arylalkenyl, arylalkynyl, heterocyclic aromatic or non-aromatic, substituted heterocyclic aromatic or non-aromatic, cycloalkyl, substituted cycloalkyl, halogen, SO_2R_{11} , $\text{NR}_{11}\text{R}_{12}$, $\text{NR}_{12}(\text{CO})\text{OR}_{11}$, $\text{NH}(\text{CO})\text{NR}_{11}\text{R}_{12}$, $\text{NR}_{12}(\text{CO})\text{R}_{11}$, $\text{O}(\text{CO})\text{R}_{11}$, $\text{O}(\text{CO})\text{OR}_{11}$, $\text{O}(\text{CS})\text{R}_{11}$, $\text{NR}_{12}(\text{CS})\text{R}_{11}$, $\text{NH}(\text{CS})\text{NR}_{11}\text{R}_{12}$, $\text{NR}_{12}(\text{CS})\text{OR}_{11}$.

R_1 and R_2 are independently selected from hydrogen, aryl, alkyl, substituted alkyl, alkenyl, substituted alkenyl, alkynyl, substituted alkynyl, halogenated alkyl, halogenated alkenyl, halogenated alkynyl, arylalkyl, arylalkenyl, arylalkynyl, heterocyclic aromatic or non-aromatic, substituted heterocyclic aromatic or non-aromatic, cycloalkyl, substituted cycloalkyl.

R_1 and R_2 can be connected to form a cycle which can be heterocyclic, substituted heterocyclic, cycloalkyl, substituted cycloalkyl.

R_3 is selected from aryl, substituted aryl, alkyl, substituted alkyl, alkenyl, substituted alkenyl, alkynyl, substituted alkynyl, arylalkyl, arylalkenyl, arylalkynyl, heterocyclic aromatic or non-aromatic, substituted heterocyclic aromatic or non-aromatic, cycloalkyl, substituted cycloalkyl, SO_2R_{11} , $\text{NR}_{11}\text{R}_{12}$, $(\text{CO})\text{OR}_{11}$, $(\text{CO})\text{NR}_{11}\text{R}_{12}$, $(\text{CO})\text{R}_{11}$, $(\text{CS})\text{R}_{11}$, $(\text{CS})\text{R}_{12}$, $(\text{CS})\text{NR}_{11}\text{R}_{12}$, $(\text{CS})\text{OR}_{11}$.

R_5 is CN or NO_2 or SO_2R_{11}

R_6 is CF_3 , alkyl, substituted alkyl, alkenyl, substituted alkenyl, alkynyl, substituted alkynyl, halogenated alkyl, halogenated alkenyl, halogenated alkynyl, halogen.

A is sulfur atom (S) or oxygen atom (O).

B is O or S or NR_3

X is carbon or nitrogen and can be at any position in the ring.

R_{11} and R_{12} are independently selected from hydrogen, aryl, alkyl, substituted alkyl, substituted alkyl, alkenyl, substituted alkenyl, alkynyl, substituted alkynyl, halogenated alkyl, halogenated alkenyl, halogenated alkynyl, arylalkyl, arylalkenyl, arylalkynyl, heterocyclic aromatic or non-aromatic, substituted heterocyclic aromatic or non-aromatic, cycloalkyl, substituted cycloalkyl.

R₁₁ and R₁₂ can be connected to form a cycle which can be heterocyclic aromatic or non-aromatic, substituted heterocyclic aromatic, cycloalkyl, substituted cycloalkyl.

[00150]

Pharmacological examination of the compounds

[00151] Compounds for which synthetic routes are described above were identified through screening on hormone refractory prostate cancer cells for antagonistic and agonistic activities against AR utilizing screening procedures similar to those in PCT applications US04/42221 and US05/05529, which are hereby incorporated by reference. A number of compounds exhibited potent antagonistic activities with minimal agonistic activities for over expressed AR in hormone refractory prostate cancer.

In vitro biological assay

Effect of compounds on AR by a reporter assay

[00152] The compounds were subjected to tests using an artificial AR response reporter system in a hormone refractory prostate cancer cell line. In this system, the prostate cancer LNCaP cells were engineered to stably express about 5-fold higher level of AR than endogenous level. The exogenous AR has similar properties to endogenous AR in that both are stabilized by a synthetic androgen R1881. The AR-over expressed cells were also engineered to stably incorporate an AR response reporter and the reporter activity of these cells shows features of hormone refractory prostate cancer. It responds to low concentration of a synthetic androgen R1881, is inhibited only by high concentrations of bicalutamide (see Table 1), and displays agonistic activity with bicalutamide (Figure 1 and Table 2). Consistent with published data, bicalutamide inhibited AR response reporter and did not have agonistic activity in hormone sensitive prostate cancer cells (Figure 2).

[00153] We examined the antagonistic activity of the compounds for which the synthesis is described above in the presence of 100 pM of R1881. Engineered LNCaP cells (LNCaP-AR, also abbreviated LN-AR) were maintained in Iscove's medium containing 10% fetal bovine serum (FBS). Two days prior to drug treatment, the cells were grown in Iscove's medium containing 10% charcoal-stripped FBS (CS-FBS) to deprive of androgens. The cells were split and grown in Iscove's medium containing 10% CS-FBS with 100 pM of R1881 and increasing concentrations of test compounds. After two days of incubation, reporter activities were assayed.

[00154] Table 1 lists the IC₅₀ of these compounds to inhibit AR in hormone refractory prostate

cancer. The control substance bicalutamide has an IC₅₀ of 889 nM. Most of the compounds identified (diarylthiohydantoins) have IC₅₀s between 100 to 200 nM in inhibiting AR in hormone refractory prostate cancer. In contrast, antiandrogenic compounds listed as examples in US patent no. 5,705,654, such as examples 30-2, 30-3, 31-2, 31-3, and 24-3 (RD73-RD77) have no inhibitory activities on AR in this system.

Table 1

Antagonistic activities against AR in hormone refractory prostate cancer,
measured by an AR response reporter and by endogenous PSA expression.

Example	Name	IC ₅₀ (nM)	IC ₅₀ (nM)
		Reporter	PSA
Bicalutamide Comparative	N-[4-cyano-3-(trifluoromethyl)phenyl]-3-[(4-fluorophenyl)sulfonyl]-2-hydroxy-2-methylpropanamide	889	>1000
29 Comparative	4-[3-(4-hydroxybutyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzotrile	No(*)	No
6-2 (6b) [RD10]	4-[3-phenyl-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzotrile	149	n/a (**)
5-3b (5c) [RD7]	4-[3-(4-methylphenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzotrile	125	132
3-3 (3c) [RD8]	4-[3-(4-hydroxyphenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzotrile	137	122
2-4 (2d) [RD9]	4-[3-(4-aminophenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzotrile	273	n/a
4 (4a) [RD13]	Chloroacetic acid 4-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]phenyl ester	131	n/a
8-2 (8b) [RD35]	4-(4-Oxo-2-thioxo-1-(4-methylphenyl)-1,3-diazaspiro[4.4]non-3-yl)-2-trifluoromethylbenzotrile	147	n/a
7-3b (7c) [RD37]	4-(8-oxo-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzotrile	124	128

9-3 (9c) [RD48]	4-(4-Oxo-2-thioxo-1-(4-methylphenyl)-1,3-diazaspiro[4.5]dec-3-yl)-2-trifluoromethylbenzotrile	194	n/a
10-3 (10c) [RD49]	4-(4-oxo-2-thioxo-1-(4-methylphenyl)-1,3-diazaspiro[4.5]undec-3-yl)-2-trifluoromethylbenzotrile	232	n/a
28 Comparative (28a) [RD52]	4-(8-methyl-4-oxo-2-thioxo-1,3,8-triazaspiro[4.5]dec-3-yl)-2-trifluoromethylbenzotrile	No	n/a
27-3 (27c) [RD53]	4-(8-methyl-4-oxo-2-thioxo-1-(4-methylphenyl)-1,3,8-triazaspiro[4.5]dec-3-yl)-2-trifluoromethylbenzotrile	638	n/a
26 (26a) [RD54]	4-[1-(4-cyanophenyl)-4-oxo-2-thioxo-1,3-diazaspiro[4.4]non-3-yl]-2-trifluoromethylbenzotrile	469	n/a
25 (25a) [RD55]	4-[1-(4-nitrophenyl)-4-oxo-2-thioxo-1,3-diazaspiro[4.4]non-3-yl]-2-trifluoromethylbenzotrile	498	n/a
12-2 (12b) [RD57]	4-(8-oxo-6-thioxo-5-(4-biphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzotrile	283	n/a
11-2 (11b) [RD58]	4-(8-oxo-6-thioxo-5-(4-hydroxyphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzotrile	162	n/a
17 (17a) [RD59]	4-[3-(4-hydroxyphenyl)-4,4-dimethyl-2,5-dithioxoimidazolidin-1-yl]-2-trifluoromethylbenzotrile	278	287
18 (18a) [RD60]	4-[3-(4-hydroxyphenyl)-4,4-dimethyl-2,5-dioxoimidazolidin-1-yl]-2-trifluoromethylbenzotrile	369	511
22-2 (22b) [RD65]	2-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-4-oxo-2-thioxoimidazolidin-1-yl]benzoic acid	523	>500
20-2 (20b) [RD66]	4-(4,4-dimethyl-5-oxo-2-thioxo-3-(4-trifluoromethylphenyl)imidazolidin-1-yl)-2-trifluoromethylbenzotrile	143	144
21-2 (21b) [RD67]	4-(4,4-bischloromethyl-5-oxo-2-thioxo-3-(4-methylphenyl)imidazolidin-1-yl)-2-trifluoromethylbenzotrile	521	>500
19-2 (19b) [RD68]	4-(4-fluoromethyl-4-methyl-5-oxo-2-thioxo-3-(4-methylphenyl)imidazolidin-1-yl)-2-trifluoromethylbenzotrile	126	129

23-2 (23b) [RD71]	4-(8-oxo-6-thioxo-5-(2-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile	258	232
30-2 Comparative (30b) [RD73]	4-(5-methyl-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile	No	No
30-3 Comparative (30c) [RD74]	4-(5-methyl-6,8-dioxo-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile	No	No
31-2 Comparative (31b) [RD75]	4-(1-methyl-4-oxo-2-thioxo-1,3-diazaspiro[4.4]non-3-yl)-2-trifluoromethylbenzonitrile	No	No
31-3 Comparative (31c) [RD76]	4-(1-methyl-2,4-dioxo-1,3-diazaspiro[4.4]non-3-yl)-2-trifluoromethylbenzonitrile	No	No
24-3 Comparative (24c) [RD77]	4-(4-oxo-2-thioxo-1,3-diazaspiro[4.4]non-3-yl)-2-trifluoromethylbenzonitrile	No	No
15-2 (15b) [RD82]	4-[4,4-dimethyl-3-(4-pyridin-2-yl)-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile	723	n/a
14-2 (14b) [RD83]	4-[4,4-dimethyl-3-(4-methylpyridin-2-yl)-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile	457	n/a
16-2 Comparative (16b) [RD84]	4-[5-(5-methyl-2H-pyrazol-3-yl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl]-2-trifluoromethylbenzonitrile	>1000	n/a
13-2 (12b) [RD85]	4-(8-oxo-6-thioxo-5-(4-biphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile	>1000	n/a
32 (32a) [RD90]	4-(8-methylimino-6-thioxo-5-p-tolyl-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile	222	421
33 (33a) [RD91]	1-[3-(4-cyano-3-trifluoromethylphenyl)-5,5-dimethyl-2-thioxo-1-p-tolyl-imidazolidin-4-ylidene]-3-ethylthiourea	157	239
34 (34a) [RD92]	1-[7-(4-cyano-3-trifluoromethylphenyl)-6-thioxo-5-p-tolyl-5,7-diazaspiro[3.4]oct-8-ylidene]-3-phenylthiourea	176	276

35 (35a) [RD93]	1-(4-Cyano-3-trifluoromethyl-phenyl)-3-[7-(4-cyano-3-trifluoromethyl-phenyl)-6-thioxo-5-p-tolyl-5,7-diaza-spiro[3.4]oct-8-ylidene]-thiourea	144	158
36-2 (36b) [RD110]	4-[8-(4-hydroxymethyl-phenyl)-5-oxo-7-thioxo-6-aza-spiro[3.4]oct-6-yl]-2-trifluoromethyl-benzonitrile	311	337
37 (37a) [RD114]	4-[5-(4-formylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl]-2-trifluoromethyl-benzonitrile	n/a	263
38 (38a) [RD116]	4-[5-[4-(1-hydroxyethyl)-phenyl]-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl]-2-trifluoromethyl-benzonitrile	n/a	187
39 (39a) [RD117]	3-{4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]-phenyl}-acrylic acid ethyl ester	n/a	197
40 (40a) [RD120]	4-[5-[4-(3-hydroxypropenyl)-phenyl]-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl]-2-trifluoromethylbenzonitrile	n/a	114
41-2 (41b) [RD128]	3-{4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]-phenyl}-propionic acid methyl ester	No	n/a
41-4 (41d) [RD133]	3-{4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl}-propionamide	224	n/a
41-5 (41e) [RD134]	3-{4-[7-(4-Cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl}-N-methyl-propionamide	234	n/a
41-6 (41f) [RD135]	3-{4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl}-N-(2-hydroxyethyl)-propionamide	732	n/a
42-2 (42b) [RD129]	4-[4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-5-yl]-phenyl]-butyric acid methyl ester	432	n/a
42-4	4-[4-[7-(4-Cyano-3-trifluoromethylphenyl)-8-oxo-6-	112	n/a

(42d) [RD130]	thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl)- butyramide		
42-5 (42e) [RD131]	4-{4-[7-(4-Cyano-3-trifluoromethylphenyl)-8-oxo-6- thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl}-N- methyl-butyramide	92	n/a
43-4 (43e) [RD137]	4-[8-Oxo-5-(4-piperazin-1-yl-phenyl)-6-thioxo-5,7- diazaspiro[3.4]oct-7-yl]-2-trifluoromethylbenzotrile	718	n/a
43-5 (43f) [RD138]	4-(5-[4-(4-methanesulfonylpiperazin-1-yl)-phenyl]-8- oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl)-2- trifluoromethylbenzotrile	138	n/a
44-2 (44b) [RD119]	44-2) 3-{4-[7-(4-Cyano-3-trifluoromethyl-phenyl)-8- oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-phenyl}- acrylamide,		113

(* No: the compound did not inhibit AR response reporter; (** n/a: the compound was not examined in this assay.

[00155] One previously unrecognized property of AR overexpression in hormone refractory prostate cancer is its ability to switch antagonists to agonists. Therefore, only those compounds with minimal or no agonistic activities are qualified to be anti-androgens for this disease. To determine agonistic activities of different compounds, we examined their stimulating activities on AR using the AR response reporter as the measure in the LN-AR system in the absence of R1881. Table 2 lists the agonistic activities of different compounds. Consistent with previous results, bicalutamide activated AR in hormone refractory prostate cancer. The diarylthiohydantoin derivatives such as examples 7-3b (RD37), 33 (RD91), 34 (RD92), and 35 (RD93) have no agonistic activity. In contrast, RU59063, and other anti-androgenic compounds listed as examples in US Patent Number 5,705,654, such as examples 30-2, 30-3, 31-2, 31-3, and 24-3 (RD73-RD77) strongly activated AR in hormone refractory prostate cancer.

Table 2
Agonistic activities of selective test substances on
AR response reporter in hormone refractory prostate cancer
Fold induction by increasing

Example	Name	concentrations of compounds		
		0.1 μ M	1 μ M	10 μ M
DMSO	Dimethyl sulfoxide	1.00 (*)	1.00	1.00
R1881	methyltrienolone	44.33	n/a(**)	n/a
Bicaluta mide	N-[4-cyano-3-(trifluoromethyl)phenyl]-3-[(4-fluorophenyl)sulfonyl]-2-hydroxy-2-methylpropanamide	1.66	3.04	10.40
29 Comp.	4-[3-(4-hydroxybutyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzotrile	10.99	20.84	34.62
7-3b (7c) [RD37]	4-(8-oxo-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzotrile	0.87	1.19	0.89
33 (33a) [RD91]	1-[3-(4-cyano-3-trifluoromethyl-phenyl)-5,5-dimethyl-2-thioxo-1-p-tolyl-imidazolidin-4-ylidene]-3-ethyl-thiourea	1.30	1.18	1.28
34 (34a) [RD92]	1-[7-(4-cyano-3-trifluoromethyl-phenyl)-6-thioxo-5-p-tolyl-5,7-diaza-spiro[3.4]oct-8-ylidene]-3-phenyl-thiourea	1.19	1.41	1.17
35 (35a) [RD93]	1-(4-Cyano-3-trifluoromethyl-phenyl)-3-[7-(4-cyano-3-trifluoromethyl-phenyl)-6-thioxo-5-p-tolyl-5,7-diaza-spiro[3.4]oct-8-ylidene]-thiourea	1.26	1.10	1.30
30-2 Comp. (30b) [RD73]	4-(5-methyl-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzotrile	14.88	19.41	35.22
30-3 Comp. (30c) [RD74]	4-(5-methyl-6,8-dioxo-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzotrile	11.39	14.26	30.63
31-2 Comp. (31b) [RD76]	4-(1-methyl-4-oxo-2-thioxo-1,3-diazaspiro[4.4]non-3-yl)-2-trifluoromethylbenzotrile	17.03	16.63	33.77
31-3	4-(1-methyl-2,4-dioxo-1,3-diaza-spiro[4.4]non-	11.99	19.77	38.95

Comp. (31c) [RD76]	3-yl)-2-trifluoromethylbenzotrile			
24-3 Comp. (24c) [RD77]	4-(4-oxo-2-thioxo-1,3-diazaspiro[4.4]non-3-yl)- 2-trifluoromethylbenzotrile	14.88	22.48	37.09

(*) Fold induction: activities induced by a specific test substance over activities in DMSO vehicle; (**)
n/a: the compound was not examined in this assay.

[00156] To examine the specificity of AR inhibitors, selective compounds were tested in LNCaP cells with an over expression of glucocorticoid receptor (GR), the closest member of AR in the nuclear receptor family. These cells also carry a GR response reporter and the reporter activity was induced by dexamethasone, a GR agonist and the induction was blocked by RU486, a GR inhibitor. Example 7-3b (RD37) (4-(8-oxo-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethyl benzotrile) had no effect on GR in this system.

Effect of compounds on AR by measuring secreted levels of
prostate specific antigen (PSA)

[00157] It is well established that PSA levels are indicators of AR activities in prostate cancer. To examine if the compounds affect AR function in a physiological environment, we determined secreted levels of endogenous PSA induced by R1881 in the AR-overexpressed LNCaP cells (LNCaP-AR, also abbreviated LN-AR). The LNCaP-AR cells are a line of lymph node carcinoma of prostate cells transduced with a plasmid that makes express androgen receptors. LNCaP-AR cells were maintained in Iscove's medium containing 10% FBS. Two days prior to drug treatment, the cells were grown in Iscove's medium containing 10% CS-FBS to deprive of androgens. The cells were split and grown in Iscove's medium containing 10% CS-FBS with appropriate concentrations of R1881 and the test compounds. After four days incubation, secreted PSA levels were assayed using PSA ELISA kits (American Qualex, San Clemente, CA)

[00158] The secreted PSA level of LNCaP-AR cells was strongly induced by 25 pM of R1881. In contrast, PSA was not induced in the parental LNCaP cells until concentration of R1881 reached 100 pM. This is consistent with our previous report that the AR in hormone refractory prostate cancer is hyper-

sensitive to androgens. A dose-dependent inhibition on AR activity was carried out to determine the IC50s of different compounds in inhibiting PSA expression, and the results were listed in Table 1. IC50s of the selective compounds on PSA expression closely resemble those measured by the reporter assay, confirming that the diarylhydantoin derivatives are strong inhibitors of AR in hormone refractory prostate cancer.

[00159] We also examined agonistic activities of selective compounds on AR in hormone refractory prostate cancer using secreted PSA as the surrogate marker. To do this, androgen-starved AR over expressed LNCaP cells were incubated with increasing concentrations of the compounds for which a synthesis is described above in the absence of R1881 and secreted PSA in the culture medium was measured 4 days later.

[00160] Table 3 lists the agonistic activities of the selective compounds. Consistent with the results obtained from the reporter assay, the diarylthiohydantoin derivatives such as examples 7-3b (RD37), 33 (RD91), 34 (RD92), and 35 (RD93) have no agonistic activities. In contrast, RUS9063, and other antiandrogenic compounds listed as examples in US patent no. 5,705,654, such as examples 30-2 (RD73), 30-3 (RD74), and 31-2 (RD75) stimulated PSA expression in hormone refractory prostate cancer.

Table 3

Agonistic activities of selective test substances on endogenous PSA in hormone refractory prostate cancer

Fold induction by increasing concentrations of compounds

Example	Name	0.1 μ M	1 μ M	10 μ M
DMSO	Dimethyl sulfoxide	1.00 (*)	1.00	1.00
R1881	methyltrienolone	20.69	n/a(**)	n/a
Bicaluta mide	N-[4-cyano-3-(trifluoromethyl)phenyl]-3-[(4-fluorophenyl)sulfonyl]-2-hydroxy-2-methylpropanamide	2.00	2.55	5.55
29 Comp.	4-[3-(4-hydroxybutyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzotrile	6.88	11.50	21.50
7-3b (7c) [RD37]	4-(8-oxo-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzotrile	1.25	1.20	1.15
33	1-[3-(4-cyano-3-trifluoromethyl-phenyl)-5,5-	1.06	1.30	0.85

(33a) [RD91]	dimethyl-2-thioxo-1-p-tolyl-imidazolidin-4-ylidene]-3-ethyl-thiourea			
34 (34a) [RD92]	1-[7-(4-cyano-3-trifluoromethyl-phenyl)-6-thioxo-5-p-tolyl-5,7-diaza-spiro[3.4]oct-8-ylidene]-3-phenyl-thiourea	1.31	1.05	0.90
35 (35a) [RD93]	1-(4-Cyano-3-trifluoromethyl-phenyl)-3-[7-(4-cyano-3-trifluoromethyl-phenyl)-6-thioxo-5-p-tolyl-5,7-diaza-spiro[3.4]oct-8-ylidene]-thiourea	1.44	1.30	1.05
30-2 Comp. (30b) [RD73]	4-(5-methyl-8-oxo-6-thioxo-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzotrile	6.25	17.95	25.65
30-3 Comp. (30c) [RD74]	4-(5-methyl-6,8-dioxo-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzotrile	7.50	15.20	23.75
31-2 Comp. (31b) [RD75]	4-(1-methyl-4-oxo-2-thioxo-1,3-diazaspiro[4.4]non-3-yl)-2-trifluoromethylbenzotrile	8.13	18.20	17.50

(*) Fold induction: activities induced by a specific test substance over activities in DMSO vehicle; (**)

n/a: the compound was not examined in this assay.

[00161]

Effect of compounds on AR mitochondrial activity by MTS assay

[00162] LNCaP-AR cells were maintained in Iscove's medium containing 10% FBS. The compounds were examined for their effect on growth of hormone refractory prostate cancer cells. Overexpressed LNCaP cells were used because these cells behave as hormone refractory prostate cancer cells in vitro and in vivo (1). We measured mitochondria activity by MTS assay, a surrogate for growth. LNCaP cells with overexpressed AR (LN-AR) were maintained in Iscove's medium containing 10% FBS. Two days prior to drug treatment, the cells were grown in Iscove's medium containing 10% CS-FBS to deprive of androgens. The cells were then split and grown in Iscove's medium containing 10% CS-FBS with appropriate concentrations of R1881 and increasing concentrations of the test compounds. After four days incubation, cell growth was monitored by MTS (Promega, Madison, WI).

[00163] Consistent with the reporter assay and PSA assay, growth of the AR-overexpressed LNCaP was stimulated by 25 microM of R1881, but the parental cells were not stimulated until R1881 concentration reached 100 microM. Figure 2 shows the inhibitory effect of selected compounds on growth of hormone refractory prostate cancer in the presence of 100 pM of R1881. The current clinical drug bicalutamide did not inhibit hormone refractory prostate cancer. In contrast, example 5-3b (RD7) (4-[3-(4-methylphenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethyl-benzonitrile) and example 7-3b (RD37) (4-(8-oxo-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile) inhibited hormone refractory prostate cancer with high potency.

[00164] We examined if growth inhibition in the MTS assay occurs by targeting AR, example 5-3b (RD7) (4-[3-(4-methylphenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethylbenzonitrile) and example 7-3b (RD37) (4-(8-oxo-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile) were tested in DU-145 cells, a prostate cancer cell line that lacks AR expression. These compounds had no growth inhibitory effect on DU-145 cells. The compounds did not inhibit cells other than AR-expressed prostate cancer cells, as they had no growth effect on MCF7 and SkBr3, two commonly used breast cancer cells, or 3T3, a normal mouse fibroblast cell line.

[00165] Examples of in vitro biological activity of diarylthiohydantoin derivatives are shown in the Figures 3, 4 and 5. For example, based on relative luciferase activity, Fig. 3 indicates that at a concentration of 500 nM the compounds ranked, in order of most active to least active as follows: RD152 > RD153 > RD145 > RD163 > RD161 = RD162 > bicalutamide. For example, based on relative PSA level, Fig 4 indicates that at a concentration of 500 nM the compounds ranked, in order of most active to least active as follows: RD138 > RD131 > RD37 > RD133 > RD134 > RD137 > RD138 > RD135 > bicalutamide. For example, based on relative MTS units, Fig. 5 indicates that at a concentration of 500 nM the compounds ranked, in order of most active to least active as follows: RD168 > RD37 > RD141 > RD162 > bicalutamide.

Inhibitory effect on hormone refractory prostate cancer xenograft tumors.

[00166] Example 7-3b (RD37) (4-(8-oxo-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzonitrile) was used to examine if the diarylthiohydantoin derivatives have in vivo effects on hormone refractory prostate cancer. First we examined this compound on xenograft tumors established from AR-overexpressed LNCaP cells. The engineered cells in Matrigel (Collaborative Biomedical) were injected subcutaneously into the flanks of the castrated male SCID mice. Tumor size was measured weekly in three dimensions using calipers. After xenograft tumors established (tumor size reached at least 40 mm³), mice with tumors were randomized and treated with different doses of

compounds orally once daily. Consistent with clinical observation, current clinical drug bicalutamide did not inhibit growth of hormone refractory prostate cancer (same as vehicle) (Figure 7a). In contrast, example 7-3b (RD37) (4-(8-oxo-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzotrile) strongly inhibited growth of these tumors (Figure 7a) and the inhibition is dose-dependent (Figure 7b). Furthermore, example 7-3b (RD37) inhibited PSA expression (Figure 8), the clinical marker for hormone refractory prostate cancer.

[00167] Example 7-3b (RD37) (4-(8-oxo-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzotrile) was also tested in another xenograft model of hormone refractory prostate cancer, hormone refractory LAPC4. This model was established from passaging of hormone sensitive prostate cancer in castrated mice, which mimics the clinical progression of prostate cancer (2). Similar to the finding using AR-overexpressed LNCaP xenograft model, current clinical drug bicalutamide did not inhibit growth and PSA expression in hormone refractory LAPC4 xenograft model (same as vehicle) (Figure 9a and 9b). In contrast, example 7-3b (RD37) strongly inhibited growth and PSA expression of these tumors (Figure 9a and 9b).

Inhibitory effect on growth of hormone sensitive prostate cancer cells.

[00168] To determine if the diarylthiahydantoin derivatives also inhibit hormone sensitive prostate cancer cells, we tested some selective compounds on growth of LNCaP cells by measuring MTS of mitochondria activities. In contrast to have no effect on growth of hormone refractory prostate cancer, the current clinical drug bicalutamide mildly inhibited hormone sensitive LNCaP cells in a dose-dependent manner. Example 5-3b (RD7) (4-[3-(4-methylphenyl)-4,4-dimethyl-5-oxo-2-thioxoimidazolidin-1-yl]-2-trifluoromethyl-benzotrile) and example 7-3b (RD37) (4-(8-oxo-6-thioxo-5-(4-methylphenyl)-5,7-diazaspiro[3.4]oct-7-yl)-2-trifluoromethylbenzotrile) inhibited hormone sensitive prostate cancer with a 10-fold higher potency than bicalutamide (Figure 10).

In vivo biological assay

[00169] All animal experiments were performed in compliance with the guidelines of the Animal Research Committee of the University of California at Los Angeles. Animals were bought from Taconic and maintained in a laminar flow tower in a defined flora colony. LNCaP-AR and LNCaP-vector cells were maintained in RPMI medium supplemented with 10% FBS. 10^6 cells in 100 μ l of 1:1 Matrigel to RPMI medium were injected subcutaneously into the flanks of intact or castrated male SCID mice. Tumor size was measured weekly in three dimensions (length x width x depth) using calipers. Mice were randomized to treatment groups when tumor size reached approximately 100 mm^3 . Drugs were given orally every day at 10 mg/kg and 50 mg/kg. To obtain pharmacodynamic readout, the animals were

imaged via an optical CCD camera, 3 hours after last dose of the treatment. A ROI is drawn over the tumor for luciferase activity measurement in photon/second. The right panels were a representation of the ROIs measurements. Data are shown in figures 11 and 12. Over 18 days RD162 was effective to prevent tumor growth and even to cause tumor shrinkage, and was distinctly more effective than bicalutamide.

[00170] The pharmacokinetics of bicalutamide, 4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-toluene [RD37], *N*-methyl-4-(4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]phenyl)butanamide [RD131], and *N*-methyl-4-[7-(4-cyano-3-trifluoromethylphenyl)-8-oxo-6-thioxo-5,7-diaza-spiro[3.4]oct-5-yl]-2-fluorobenzamide (52d) [RD162] were evaluated in vivo using 8 week-old FVB mice which were purchased from Charles River Laboratories. Mice were divided into groups of three for each time points. Two mice were not treated with drug and two other mice were treated with vehicle solution. Each group was treated with 10 mg per kilogram of body weight.

[00171] The drug was dissolved in a mixture 1:5:14 of DMSO : PEG400 : H₂O. (Vehicle solution) and was administered into mice through the tail vein. The animals are warmed under a heat lamp for approximately 20 minutes prior to treatment to dilate their tail vein. Each mouse was placed into a mouse restrainer (Fisher Sci. Cat# 01-288-32A) and was injected with 200 µl of drug in vehicle solution into the dilated tail vein. After drug administration, the animals were euthanized via CO₂ inhalation at different timepoints: 5 mn, 30 mn, 2 h, 6 h, 16 h. Animals were immediately bled after exposure to CO₂ via cardiac puncture (1 ml BD syringe + 27G 5/8 needle). For oral dosage, the drug was dissolved in a mixture 50:10:1:989 of DMSO : Carboxymethylcellulose : Tween80:H₂O before oral administration via a feeding syringe.

[00172] The serum samples were analyzed to determine the drug's concentration by the HPLC which (Waters 600 pump, Waters 600 controller and Waters 2487 detector) was equipped with an Alltima C18 column (3µ, 150 mm×4.6 mm). The RD37, RD131, and RD162 compounds were detected at 254 nm wave length and bicalutamide was detected at 270 nm wave length.

[00173] The samples for HPLC analysis were prepared according to the following procedure:

- Blood cells were separated from serum by centrifugation.
- To 400 µl of serum were added 80 µl of a 10 µM solution of an internal standard and 520 µl of acetonitrile. Precipitation occurred.
- The mixture was vortexed for 3 minutes and then placed under ultrasound for 30 minutes.

- The solid particles were filtered off or were separated by centrifugation.
- The filtrate was dried under an argon flow to dryness. The sample was reconstructed to 80 μ l with acetonitrile before analyzing by HPLC to determine the drug concentration.
- Standard curve of drug was used to improve accuracy.

[00174] The concentration of RD162 in plasma as a function of time resulting from intravenous and from oral administration is shown in figure 13. The steady state concentration (C_{ss}) of bicalutamide, RD131, and RD162 is shown in Table 4. The concentration at steady state of RD162 is essentially as good as that of bicalutamide, and substantially better than RD131.

Name	IC50 [nM]	LogP	C_{ss} , 10 mg/kg [μ M]	C_{ss} , 25 mg/kg [μ M]	C_{ss} , 50 mg/kg [μ M]
Bic.	1000	2.91	10.0	11.4	11.9
RD131	92	3.44	0.39	0.43	0.40
RD162	122	3.20	9.9	10.7	10.2

Table 4. Steady-state concentration of bicalutamide, RD131, and RD162 in mice plasma.

Ranking of Compounds in Tiers

[00175] Tables 5 – 10 present diarylhydantoin compounds grouped into Tiers 1-6. Table 11 presents diarylhydantoin compounds which have not been placed into a tier. The placement of compounds into tiers was based on available data coupled with analytical judgment. Data considered included in vitro assays (AR response reporter system in LNCaP cell line, PSA level measurement, MTS mitochondrial assay) and in vivo experiments (tumor size measured directly or by emission induced by luciferase reporter gene, pharmacokinetic assays based on blood plasma levels). Not every compound was subjected to each assay. Not all data that was generated is shown. Judgment was applied in ranking compounds relative to each other for their utility in treating prostate cancer, in particular when ranking two compounds for which the same experiments were not performed. Characteristics considered in establishing the ranking include AR antagonism activity, lack of AR agonism in hormone refractory cells, prevention of tumor growth, tumor shrinkage, and pharmacokinetic behavior, with a longer residence time in blood being advantageous.

Tier 1

[00176] Generally, Tier 1 compounds are diarylthiohydantoin with a disubstituted left hand aryl ring that are disubstituted on the right hydantoin carbon, and have either an oxygen or N substituent on the left hydantoin carbon. It is expected that the amido substituent hydrolyzes to an oxygen in aqueous solutions such as encountered in biological systems, in vitro and in vivo. RD100 has good activity with an iodine instead of a CF_3 substituent on the left hand aryl ring.

[00177] Tier 1 compounds (see Table 5) were judged to be much better than bicalutamide for treating prostate cancer. However, RD37 and RD131 were found to metabolize fast, that is, have a short residence time in blood. RD162 had desirable pharmacokinetics.

[00178] Figure 17 shows that under treatment with bicalutamide, PSA levels for LNCaP cells stayed the same or increased relative to treatment with vehicle solution, whereas under treatment with RD162, PSA levels decreased. Figure 18 illustrates that under treatment with vehicle solution, tumors continued to increase in size. By contrast, under treatment with RD162 at a dose of 1 mg per kg body weight per day, the rate of tumor increase decreased, and the size of the tumor appeared to be stabilizing after about 17 days. Under treatment with RD162 at a dose of 10 mg per kg body weight per day, tumor size decreased with time. Figure 19 illustrates that under treatment with RD162 at a dose of 10 mg per kg body weight per day, photon emission associated with luciferase activity decreased. Figure 20 shows that treatment with RD162 at this dose resulted in a decrease or stabilization of tumor size and a decrease in photon emission associated with luciferase activity.

[00179] Figure 21 shows that under treatment with RD162, RD162', RD162'', RD169, and RD170 at doses of 100, 200, 500, and 1000 nM, PSA levels of LN-AR cells decreased. Moreover, the higher the dose, the lower the PSA level. Figure 23 presents urogenital tract weight and rate of photon emission associated with luciferase activity initially and after 14 days of treatment with bicalutamide or with RD162 for intact and castrated mice. The weight and rate of photon emission increased for both intact and castrated mice. Treatment of castrated mice with RD162 resulted in a decrease in weight and photon emission with respect to the untreated castrated mice, as did treatment with bicalutamide.

[00180] Thus, Tier 1 compounds are particularly advantageous for use as AR antagonists, and as therapeutic agents for hormone refractory prostate cancer. They may be useful to treat other AR related diseases or conditions such as benign prostate hyperplasia, hair loss, and acne. These and related compounds may also be useful as modulators of other nuclear receptors, such as glucocorticoid receptor, estrogen receptor, and peroxisome proliferator-activated receptor, and as therapeutic agents for diseases in which nuclear receptors play a role, such as breast cancer, ovarian cancer, diabetes, cardiac diseases, and metabolism related diseases. They may be useful in assays e.g. as standards, or as intermediates or prodrugs.

TABLE 5

TIER 1 COMPOUNDS	
RD7	RD8
RD10	RD35
RD36	RD37
RD57	RD58
RD90	RD91
RD92	RD93

RD 9 RD 10 RD 11 RD 12 RD 13 RD 14 RD 15 RD 16 RD 17 RD 18 RD 19 RD 20 RD 21 RD 22 RD 23 RD 24 RD 25 RD 26 RD 27 RD 28 RD 29 RD 30 RD 31 RD 32 RD 33 RD 34 RD 35 RD 36 RD 37 RD 38 RD 39 RD 40 RD 41 RD 42 RD 43 RD 44 RD 45 RD 46 RD 47 RD 48 RD 49 RD 50 RD 51 RD 52 RD 53 RD 54 RD 55 RD 56 RD 57 RD 58 RD 59 RD 60 RD 61 RD 62 RD 63 RD 64 RD 65 RD 66 RD 67 RD 68 RD 69 RD 70 RD 71 RD 72 RD 73 RD 74 RD 75 RD 76 RD 77 RD 78 RD 79 RD 80 RD 81 RD 82 RD 83 RD 84 RD 85 RD 86 RD 87 RD 88 RD 89 RD 90 RD 91 RD 92 RD 93 RD 94 RD 95 RD 96 RD 97 RD 98 RD 99 RD 100

TIER 1 COMPOUNDS

<p>RD94</p>	<p>RD95</p>
<p>RD96</p>	<p>RD97</p>
<p>RD100</p>	<p>RD102</p>
<p>RD119</p>	<p>RD120</p>
<p>RD130</p>	<p>RD131</p>
<p>RD145</p>	<p>RD152</p>
<p>RD153</p>	<p>RD163</p>

TIER 1 COMPOUNDS	
RD182	RD162
RD182*	RD168
RD189	RD170

Tier 2

[00181] Tier 2 compounds (see Table 6) were significantly better than bicalutamide for treating prostate cancer, although there were indications that RD54 could act as an agonist. Figure 3 illustrates that compounds RD145, RD152, RD153, RD162, and RD163 in Tier 1 and RD161 in Tier 2 dosed at concentrations ranging from 125 nM to 1000 nM acted to reduce luciferase activity in LNCaP-AR cells whereas control solutions of DMSO and of bicalutamide had little or no effect. Figure 4 illustrates, for example, that at concentrations of 1000 nM, compounds RD37 and RD131, in Tier 1, caused a greater decrease in PSA level of LNCaP-AR cells than RD133, RD134, and RD138 in Tier 2. Figure 11 presents tumor volume over time, and illustrates that under treatment with bicalutamide or vehicle solution, tumors continued to grow, whereas under treatment with RD162, in Tier 1, tumors decreased in size. Figure 12 illustrates that photon emission associated with luciferase activity remained about the same or increased under treatment with bicalutamide relative to treatment with vehicle solution, whereas photon emission decreased under treatment with RD162. Figure 14 illustrates that under treatment with bicalutamide, there was little or no decrease in PSA levels, whereas under treatment with RD131 and RD162, PSA levels decreased. Figure 15 illustrates that the IC_{50} for RD37, RD 131, and RD162, in Tier 1, was much lower than the IC_{50} for bicalutamide.

[00182] Generally, Tier 2 compounds are structurally similar to Tier 1 compounds, but with different substituents on the right hand aryl ring. Tier 2 compounds are advantageous for use as AR antagonists, and as therapeutic agents for hormone refractory prostate cancer. They may be useful to treat other AR related diseases or conditions such as benign prostate hyperplasia, hair loss, and acne.

These and related compounds may also be useful as modulators of other nuclear receptors, such as estrogen receptor and peroxisome proliferator-activated receptor, and as therapeutic agents for diseases in which nuclear receptors play a role, such as breast cancer, ovarian cancer, diabetes, cardiac diseases, and metabolism related diseases. They may be useful in assays e.g. as standards, or as intermediates or prodrugs.

TABLE 6

TIER 2 COMPOUNDS	
<p>RD6</p> <p>(comparative)</p>	<p>RD13</p>
<p>RD48</p>	<p>RD49</p>
<p>RD51</p>	<p>RD53</p>
<p>RD54</p>	<p>RD55</p>
<p>RD63</p>	<p>RD66</p>

TIER 2 COMPOUNDS	
RD88	RD71
RD87	RD103
RD110	RD111
RD114	RD116
RD133	RD134
RD138	RD161

Tier 3

[00183] Tier 3 compounds (see Table 7) were judged to be slightly better than bicalutamide for treating prostate cancer. RD133, RD134, and RD138 (in Tier 2) caused a greater decrease in PSA level of LNCaP-AR cells than RD135 and RD137, in Tier 3. All of these compounds caused a greater decrease in PSA level than bicalutamide.

[00184] Other Tier 3 compounds (not shown) were not diarylthiohydantoin, and were comparable in activity to prior art monoarylthiohydantoin compounds RD2, RD4, and RD5.

[00185] Thus, Tier 3 compounds are useful as AR antagonists, and as therapeutic agents for hormone refractory prostate cancer. They may be useful to treat other AR related diseases or conditions such as benign prostate hyperplasia, hair loss, and acne. These and related compounds may also be useful as modulators of other nuclear receptors, such as estrogen receptor and peroxisome proliferator-activated receptor, and as therapeutic agents for diseases in which nuclear receptors play a role, such as breast cancer, ovarian cancer, diabetes, cardiac diseases, and metabolism related diseases. They may be useful in assays e.g. as standards, or as intermediates or prodrugs.

TABLE 7

TIER 3 COMPOUNDS	
<p>RD3</p> <p>(comparative)</p>	<p>RD4</p> <p>(comparative)</p>
<p>RD5</p> <p>(comparative)</p>	<p>RD89</p>
<p>RD127</p>	<p>RD128</p>
<p>RD129</p>	<p>RD135</p>
<p>RD137</p>	

Tier 4

[00186] Tier 4 compounds (see Table 8) were judged to be no better than bicalutamide for treating prostate cancer. Tier 4 RD 39 and RD40 and Tier 1 RD37, for example, differ only in the substituent on the lower right carbon of the hydantoin ring. The substituents on the right hand aryl ring may also affect activity.

[00187] Some Tier 4 compounds (including those shown and others that are not shown) were not diaryl compounds (lacking the right hand aryl ring), were not thiohydantoins, were not disubstituted on the carbon on the lower right hand of the hydantoin ring, and/or had substituents other than oxygen or amido on the lower left hand carbon of the hydantoin ring. This provides evidence of the surprising advantages of diarylthiohydantoins that are disubstituted on the lower right hand carbon of the hydantoin ring and have oxygen or amido on the lower left hand carbon of the hydantoin ring.

Thus, Tier 4 compounds may be useful as AR antagonists, and as therapeutic agents for hormone refractory prostate cancer, at least to the extent that they are comparable to bicalutamide. They may be useful to treat other AR related diseases or conditions such as benign prostate hyperplasia, hair loss, and acne. These and related compounds may also be useful as modulators of other nuclear receptors, such as estrogen receptor and peroxisome proliferator-activated receptor, and as therapeutic agents for diseases in which nuclear receptors play a role, such as breast cancer, ovarian cancer, diabetes, cardiac diseases, and metabolism related diseases. They may be useful in assays e.g. as standards, or as intermediates or prodrugs.

TABLE 8

TIER 4 COMPOUNDS	
<p>RD2</p> <p>(comparative)</p>	<p>RD9</p>
<p>RD21</p>	<p>RD22</p>

TIER 4 COMPOUNDS

RD23		RD24	
RD25		RD26	
RD27		RD30	
RD31		RD39	
RD40		RD44	
RD59		RD60	
RD67		RD82	
RD83		RD117	
RD118		RD148	

TIER 4 COMPOUNDS	
RD149	RD150
RD151	

Tier 5

[00188] Tier 5 compounds (see Table 9) were inactive or nearly inactive, and thus, were worse than bicalutamide for treating prostate cancer. The substituents on the right hand aryl ring are important to determining activity.

[00189] Some Tier 5 compounds (some of which are shown and some that are not shown) were not diaryl compounds (lacking the right hand aryl ring), were not thiohydantoin, were not disubstituted on the carbon on the lower right hand of the hydantoin ring, and/or had substituents other than oxygen or amido on the lower left hand carbon of the hydantoin ring. This provides evidence of the surprising advantages of diarylthiohydantoin that are disubstituted on the lower right hand carbon of the hydantoin ring and have oxygen or amido on the lower left hand carbon of the hydantoin ring. In particular, the terminal substituent in RD155, RD 156, and 158 ($\text{CH}_2\text{NR}_x\text{R}_y$, where $\text{R}_{x,y} = \text{H}$ or methyl) is not seen as contributing to activity in these compounds.

[00190] Tier 5 compounds would not be desirable for treatment of prostate cancer or as AR antagonists, although these and related compounds may be useful as modulators of other nuclear receptors, such as estrogen receptor and peroxisome proliferator-activated receptor, and as therapeutic agents for diseases in which nuclear receptors play a role, such as breast cancer, ovarian cancer, diabetes, cardiac diseases, and metabolism related diseases. They may be useful in assays e.g. as standards, or as intermediates or prodrugs.

TABLE 9

TIER 5 COMPOUNDS

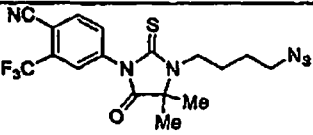
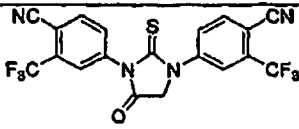
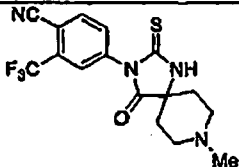
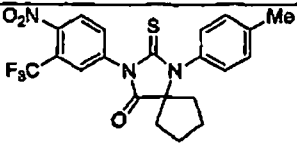
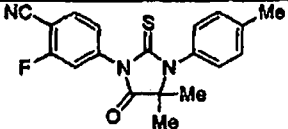
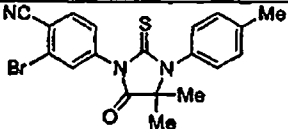
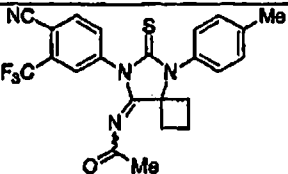
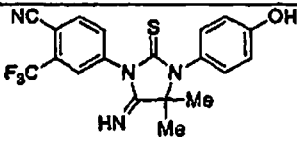
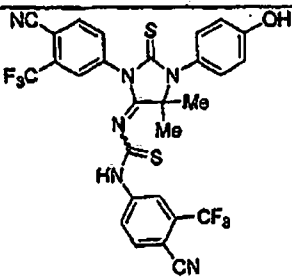
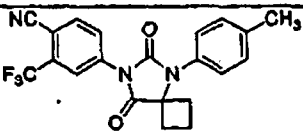
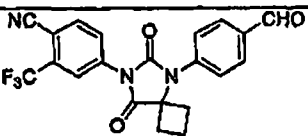
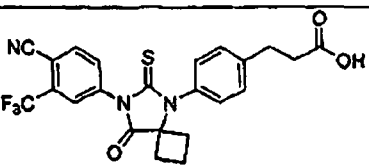
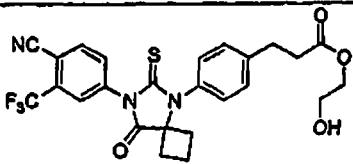
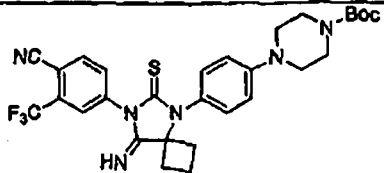
TIER 5 COMPOUNDS	
RD32	RD33
RD85	RD84
RD85	RD155
RD158	RD157
RD158	

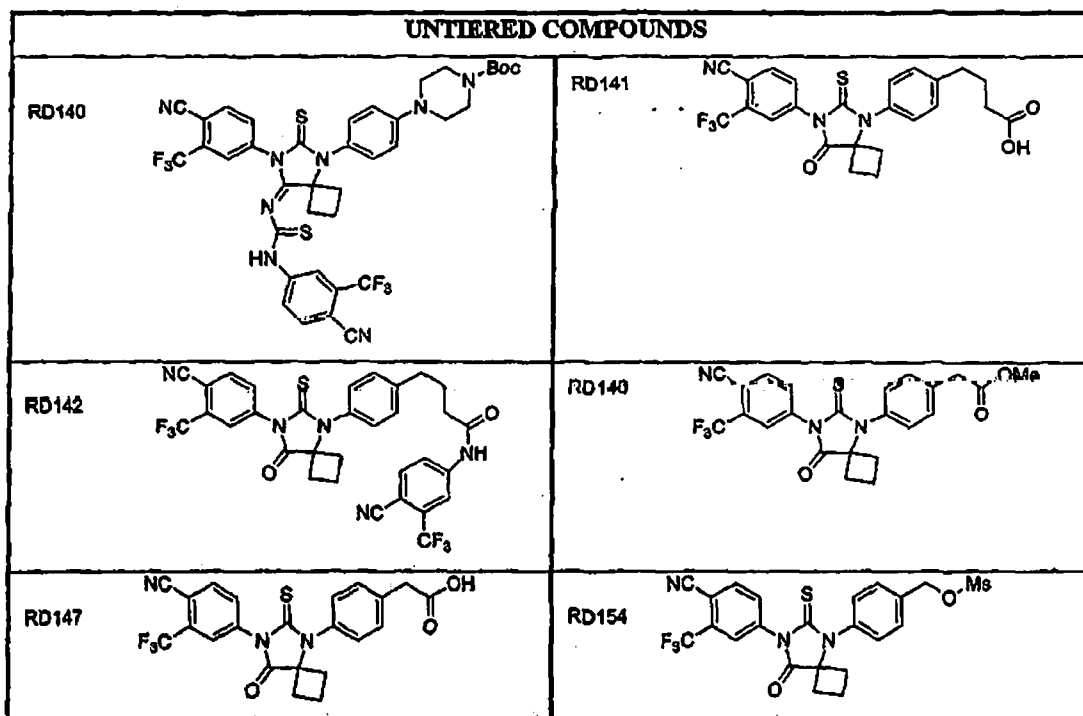
Tier 6

[00191] Tier 6 compounds (see Table 10) were inactive or nearly inactive, and furthermore were strong agonists, and thus were much worse than bicalutamide for treating prostate cancer. The comparative compounds ranked very poor relative to the inventive compounds. Notably, RD72 had very poor activity, with a chlorine substituent on the left hand aryl ring, whereas RD7, with a trifluoromethane, and RD100, with iodine, ranked in Tier 1. The results for the Tier 6 compounds provide evidence of the surprising advantages of diarylthiohydantoin that are disubstituted on the lower right hand carbon of the hydantoin ring and have oxygen or amide on the lower left hand carbon of the hydantoin ring, and have certain substituents on the left hand aryl ring.

[00192] Tier 6 compounds would not be desirable for treatment of prostate cancer or as AR antagonists.

UNTIERED COMPOUNDS

<p>RD1</p>  <p>(comparative)</p>	<p>RD19</p> 
<p>RD52</p> 	<p>RD79</p> 
<p>RD80</p> 	<p>RD81</p> 
<p>RD89</p> 	<p>RD104</p> 
<p>RD105</p> 	<p>RD106</p> 
<p>RD115</p> 	<p>RD132</p> 
<p>RD138</p> 	<p>RD139</p> 



[00195] In short, novel compounds which show evidence of being far superior to bicalutamide in treating prostate cancer were identified and produced.

Sensitivity of Anti-Cancer Activity of Compounds to Structural Differences

[00196] The inventors have determined that what might appear to be a small change in the structure of hydantoin compounds may result in a large change in that compound's performance in treating prostate cancer. For example, RD161 and RD162 differ only by a single fluorine substituent on an aryl ring, and RD162 is in Tier 1, while RD161 is in Tier 2, both being better than bicalutamide for the treatment of prostate cancer, but RD162 being superior. However, RD149, which differs from RD 161 only in having an additional carbon atom between the methylcarbamoyl group and the aryl ring, is no better than bicalutamide for the treatment of prostate cancer and is ranked in Tier 4. The effect of RD161, RD162, and RD149 on luciferase activity can be seen in Figure 24. At a given concentration of compound, the luciferase activity upon exposure to RD161 and RD162 is less than the luciferase activity upon exposure to RD149.

[00197] RD9 differs from RD8 only in that an amino group is substituted for a hydroxyl group. However, whereas RD8 is in Tier 1, much better than bicalutamide for the treatment of prostate cancer, RD9 is in Tier 4, no better than bicalutamide. The effect of RD8 and RD9 on luciferase activity in the 1AR cell line can be seen in Figure 27. For a given dose, the luciferase activity upon exposure to RD8 is less than the luciferase activity upon exposure to RD9. The effect of RD8 and RD9 on luciferase activity in the 4AR cell line can be seen in Figure 26. For a given dose, the luciferase activity upon exposure to RD8 is less than the luciferase activity upon exposure to RD9. The effect of RD8 and RD9 on PSA levels in the LN/AR cell line can be seen in Figure 25. For a given dose, the PSA level upon exposure to RD8 is less than the PSA level upon exposure to RD9.

[00198] RD130 and RD131 differ from each other only by a methyl substituent on the end of a carbamoyl group and both compounds are ranked in Tier 1, although RD131 has been found to be particularly advantageous. RD129 is the same as RD130, with the exception of a methoxy group being substituted for an amino group. However, RD129 is ranked in Tier 3. RD128 is similar to RD129, but has one less carbon in the chain linking the ester group to the aryl ring; RD128 is ranked in Tier 3. The effect of RD130, RD131, RD128, and RD129 on PSA levels in the LN/AR cell line can be seen in Figure 28. For a given concentration, the PSA level upon exposure to RD130 and RD131 is less than the PSA level upon exposure to RD128 and RD129.

[00199] RD153 and RD155 differ from each other in that the former has a methylcarbamoyl group attached to an aryl ring and a dimethyl substituent attached to the thiohydantoin group, whereas the latter has a methylamino group attached to the right hand aryl ring and a cyclobutyl substituent attached to the thiohydantoin group. Whereas RD153 is in Tier 1, much better than bicalutamide for the treatment of prostate cancer, RD155 is in Tier 5, inactive or nearly inactive in the treatment of prostate cancer. The effect of RD153 and RD155 on luciferase activity in the LN/AR cell line can be seen in Figure 29. For a given concentration, the luciferase activity upon exposure to RD153 is less than the luciferase activity upon exposure to RD155.

[00200] RD58 and RD60 differ from each other in the substitution of a thio for an oxo group and a dimethyl substituent for a cyclobutyl substituent. Whereas RD58 is in Tier 1, RD60 is in Tier 4.

Pharmaceutical Compositions and Administration

[00201] The compounds of the invention are useful as pharmaceutical compositions prepared with a therapeutically effective amount of a compound of the invention, as defined herein, and a pharmaceutically acceptable carrier or diluent.

[00202] The diarylhydantoin compounds of the invention can be formulated as pharmaceutical

compositions and administered to a subject in need of treatment, for example a mammal, such as a human patient, in a variety of forms adapted to the chosen route of administration, for example, orally, nasally, intraperitoneally, or parenterally, by intravenous, intramuscular, topical or subcutaneous routes, or by injection into tissue.

[00203] Thus, diarylhydantoin compounds of the invention may be systemically administered, e.g., orally, in combination with a pharmaceutically acceptable vehicle such as an inert diluent or an assimilable edible carrier, or by inhalation or insufflation. They may be enclosed in hard or soft shell gelatin capsules, may be compressed into tablets, or may be incorporated directly with the food of the patient's diet. For oral therapeutic administration, the diarylhydantoin compounds may be combined with one or more excipients and used in the form of ingestible tablets, buccal tablets, troches, capsules, elixirs, suspensions, syrups, wafers, and the like. The diarylhydantoin compounds may be combined with a fine inert powdered carrier and inhaled by the subject or insufflated. Such compositions and preparations should contain at least 0.1% diarylhydantoin compounds. The percentage of the compositions and preparations may, of course, be varied and may conveniently be between about 2% to about 60% of the weight of a given unit dosage form. The amount of diarylhydantoin compounds in such therapeutically useful compositions is such that an effective dosage level will be obtained.

[00204] The tablets, troches, pills, capsules, and the like may also contain the following: binders such as gum tragacanth, acacia, corn starch or gelatin; excipients such as dicalcium phosphate; a disintegrating agent such as corn starch, potato starch, alginic acid and the like; a lubricant such as magnesium stearate; and a sweetening agent such as sucrose, fructose, lactose or aspartame or a flavoring agent such as peppermint, oil of wintergreen, or cherry flavoring may be added. When the unit dosage form is a capsule, it may contain, in addition to materials of the above type, a liquid carrier, such as a vegetable oil or a polyethylene glycol. Various other materials may be present as coatings or to otherwise modify the physical form of the solid unit dosage form. For instance, tablets, pills, or capsules may be coated with gelatin, wax, shellac or sugar and the like. A syrup or elixir may contain the active compound, sucrose or fructose as a sweetening agent, methyl and propylparabens as preservatives, a dye and flavoring such as cherry or orange flavor. Of course, any material used in preparing any unit dosage form should be pharmaceutically acceptable and substantially non-toxic in the amounts employed. In addition, the diarylhydantoin compounds may be incorporated into sustained-release preparations and devices. For example, the diarylhydantoin compounds may be incorporated into time release capsules, time release tablets, and time release pills.

[00205] The diarylhydantoin compounds may also be administered intravenously or intraperitoneally by infusion or injection. Solutions of the diarylhydantoin compounds can be prepared

in water, optionally mixed with a nontoxic surfactant. Dispersions can also be prepared in glycerol, liquid polyethylene glycols, triacetin, and mixtures thereof and in oils. Under ordinary conditions of storage and use, these preparations can contain a preservative to prevent the growth of microorganisms.

[00206] The pharmaceutical dosage forms suitable for injection or infusion can include sterile aqueous solutions or dispersions or sterile powders comprising the diarylhydantoin compounds which are adapted for the extemporaneous preparation of sterile injectable or infusible solutions or dispersions, optionally encapsulated in liposomes. In all cases, the ultimate dosage form should be sterile, fluid and stable under the conditions of manufacture and storage. The liquid carrier or vehicle can be a solvent or liquid dispersion medium comprising, for example, water, ethanol, a polyol (for example, glycerol, propylene glycol, liquid polyethylene glycols, and the like), vegetable oils, nontoxic glyceryl esters, and suitable mixtures thereof. The proper fluidity can be maintained, for example, by the formation of liposomes, by the maintenance of the required particle size in the case of dispersions or by the use of surfactants. The prevention of the action of microorganisms can be brought about by various antibacterial and antifungal agents, for example, parabens, chlorobutanol, phenol, sorbic acid, thimerosal, and the like. In many cases, it will be preferable to include isotonic agents, for example, sugars, buffers or sodium chloride. Prolonged absorption of the injectable compositions can be brought about by the use in the compositions of agents delaying absorption, for example, aluminum monostearate and gelatin.

[00207] Sterile injectable solutions are prepared by incorporating the diarylhydantoin compounds in the required amount in the appropriate solvent with various of the other ingredients enumerated above, as required, followed by filter sterilization. In the case of sterile powders for the preparation of sterile injectable solutions, the preferred methods of preparation are vacuum drying and freeze drying techniques, which yield a powder of the active ingredient plus any additional desired ingredient present in the previously sterile-filtered solutions.

[00208] For topical administration, the diarylhydantoin compounds may be applied in pure form. However, it will generally be desirable to administer them to the skin as compositions or formulations, in combination with a dermatologically acceptable carrier, which may be a solid or a liquid.

[00209] Useful solid carriers include finely divided solids such as talc, clay, microcrystalline cellulose, silica, alumina and the like. Other solid carriers include nontoxic polymeric nanoparticles or microparticles. Useful liquid carriers include water, alcohols or glycols or water/alcohol/glycol blends, in which the diarylhydantoin compounds can be dissolved or dispersed at effective levels, optionally with the aid of non-toxic surfactants. Adjuvants such as fragrances and additional antimicrobial agents can be added to optimize the properties for a given use. The resultant liquid compositions can be applied from absorbent pads, used to impregnate bandages and other dressings, or sprayed onto the affected area using

pump-type or aerosol sprayers.

[00210] Thickeners such as synthetic polymers, fatty acids, fatty acid salts and esters, fatty alcohols, modified celluloses or modified mineral materials can also be employed with liquid carriers to form spreadable pastes, gels, ointments, soaps, and the like, for application directly to the skin of the user.

[00211] Examples of useful dermatological compositions which can be used to deliver the diarylhydantoin compounds to the skin are known to the art; for example, see Jacquet et al. (U.S. Pat. No. 4,608,392), Geria (U.S. Pat. No. 4,992,478), Smith et al. (U.S. Pat. No. 4,559,157) and Wortzman (U.S. Pat. No. 4,820,508), all of which are hereby incorporated by reference.

[00212] Useful dosages of the compounds of formula I can be determined by comparing their in vitro activity, and in vivo activity in animal models. Methods for the extrapolation of effective dosages in mice, and other animals, to humans are known to the art; for example, see U.S. Pat. No. 4,938,949, which is hereby incorporated by reference.

[00213] For example, the concentration of the diarylhydantoin compounds in a liquid composition, such as a lotion, can be from about 0.1-25% by weight, or from about 0.5-10% by weight. The concentration in a semi-solid or solid composition such as a gel or a powder can be about 0.1-5% by weight, or about 0.5-2.5% by weight.

[00214] The amount of the diarylhydantoin compounds required for use in treatment will vary not only with the particular salt selected but also with the route of administration, the nature of the condition being treated and the age and condition of the patient and will be ultimately at the discretion of the attendant physician or clinician.

[00215] Effective dosages and routes of administration of agents of the invention are conventional. The exact amount (effective dose) of the agent will vary from subject to subject, depending on, for example, the species, age, weight and general or clinical condition of the subject, the severity or mechanism of any disorder being treated, the particular agent or vehicle used, the method and scheduling of administration, and the like. A therapeutically effective dose can be determined empirically, by conventional procedures known to those of skill in the art. See, e.g., *The Pharmacological Basis of Therapeutics*, Goodman and Gilman, eds., Macmillan Publishing Co., New York. For example, an effective dose can be estimated initially either in cell culture assays or in suitable animal models. The animal model may also be used to determine the appropriate concentration ranges and routes of administration. Such information can then be used to determine useful doses and routes for administration in humans. A therapeutic dose can also be selected by analogy to dosages for comparable

therapeutic agents.

[00216] The particular mode of administration and the dosage regimen will be selected by the attending clinician, taking into account the particulars of the case (*e.g.*, the subject, the disease, the disease state involved, and whether the treatment is prophylactic). Treatment may involve daily or multi-daily doses of compound(s) over a period of a few days to months, or even years.

[00217] In general, however, a suitable dose will be in the range of from about 0.001 to about 100 mg/kg, *e.g.*, from about 0.01 to about 100 mg/kg of body weight per day, such as above about 0.1 mg per kilogram, or in a range of from about 1 to about 10 mg per kilogram body weight of the recipient per day. For example, a suitable dose may be about 1 mg/kg, 10 mg/kg, or 50 mg/kg of body weight per day.

[00218] The diarylhydantoin compounds are conveniently administered in unit dosage form; for example, containing 0.05 to 10000 mg, 0.5 to 10000 mg, 5 to 1000 mg, or about 100 mg of active ingredient per unit dosage form.

[00219] The diarylhydantoin compounds can be administered to achieve peak plasma concentrations of, for example, from about 0.5 to about 75 μM , about 1 to 50 μM , about 2 to about 30 μM , or about 5 to about 25 μM . Exemplary desirable plasma concentrations include at least or no more than 0.25, 0.5, 1, 5, 10, 25, 50, 75, 100 or 200 μM . For example, plasma levels may be from about 1 to 100 micromolar or from about 10 to about 25 micromolar. This may be achieved, for example, by the intravenous injection of a 0.05 to 5% solution of the diarylhydantoin compounds, optionally in saline, or orally administered as a bolus containing about 1-100 mg of the diarylhydantoin compounds. Desirable blood levels may be maintained by continuous infusion to provide about 0.00005 - 5 mg per kg body weight per hour, for example at least or no more than 0.00005, 0.0005, 0.005, 0.05, 0.5, or 5 mg/kg/hr. Alternatively, such levels can be obtained by intermittent infusions containing about 0.0002 - 20 mg per kg body weight, for example, at least or no more than 0.0002, 0.002, 0.02, 0.2, 2, 20, or 50 mg of the diarylhydantoin compounds per kg of body weight.

[00220] The diarylhydantoin compounds may conveniently be presented in a single dose or as divided doses administered at appropriate intervals, for example, as two, three, four or more sub-doses per day. The sub-dose itself may be further divided, *e.g.*, into a number of discrete loosely spaced administrations; such as multiple inhalations from an insufflator.

[00221] A number of the above-identified compounds exhibit little or no agonistic activities with respect to hormone refractory prostate cancer cells. Because these compounds are strong AR inhibitors, they can be used not only in treating prostate cancer, but also in treating other AR related diseases or conditions such as benign prostate hyperplasia, hair loss, and acne. Because AR belongs to

the family of nuclear receptors, these compounds may serve as scaffolds for drug synthesis targeting other nuclear receptors, such as estrogen receptor and peroxisome proliferator-activated receptor. Therefore, they may be further developed for other diseases such as breast cancer, ovarian cancer, diabetes, cardiac diseases, and metabolism related diseases, in which nuclear receptors play a role.

[00222] The embodiments illustrated and discussed in this specification are intended only to teach those skilled in the art the best way known to the inventors to make and use the invention. Nothing in this specification should be considered as limiting the scope of the present invention. All examples presented are representative and non-limiting. The above-described embodiments of the invention may be modified or varied, without departing from the invention, as appreciated by those skilled in the art in light of the above teachings. It is therefore to be understood that, within the scope of the claims and their equivalents, the invention may be practiced otherwise than as specifically described.

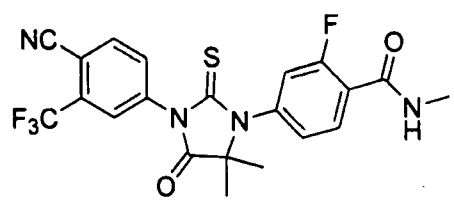
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We Claim:

1. A compound having the formula



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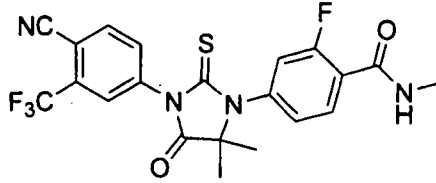
or a pharmaceutically acceptable salt thereof.

2. A compound as claimed in claim 1, for treatment of a hyperproliferative disorder.
3. A pharmaceutical composition comprising a compound as claimed in claim 1 or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier or diluent.
4. A pharmaceutical composition as claimed in claim 3, wherein said compound is in an amount equivalent to a dosage amount of from about 0.001 mg per kg body weight per day to about 100 mg per kg body weight per day for treatment of a hyperproliferative disorder.
5. A pharmaceutical composition as claimed in claim 3, wherein said compound is in an amount equivalent to a dosage amount of from about 0.01 mg per kg body weight per day to about 100 mg per kg body weight per day for treatment of a hyperproliferative disorder.
6. A pharmaceutical composition as claimed in claim 3, wherein said compound is in an amount equivalent to a dosage amount of from about 0.1 mg per kg body weight per day to about 10 mg per kg body weight per day for treatment of a hyperproliferative disorder.

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mixing N-Methyl-2-fluoro-4-(1,1-dimethyl-cyanomethyl)-aminobenzamide and 4-Isothiocyanto-2-trifluoromethylbenzonitrile in DMF and heating to form a first mixture;

adding an alcohol and an acid to the first mixture to form a second mixture;

refluxing the second mixture; and

cooling the second mixture, combining the second mixture with water and extracting an organic layer;

isolating the compound from the organic layer.

Dated this 13th day of December, 2007

Archana Shanker
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Agent for the Applicant

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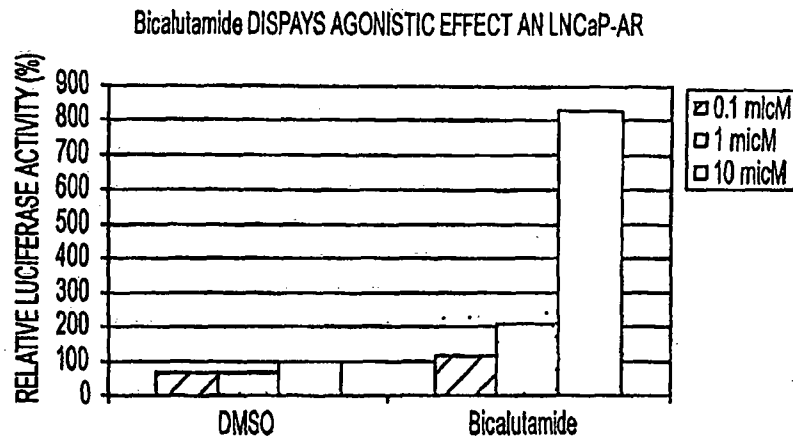


FIG. 1.

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EFFECT OF COMPOUNDS ON LNCaP-AR

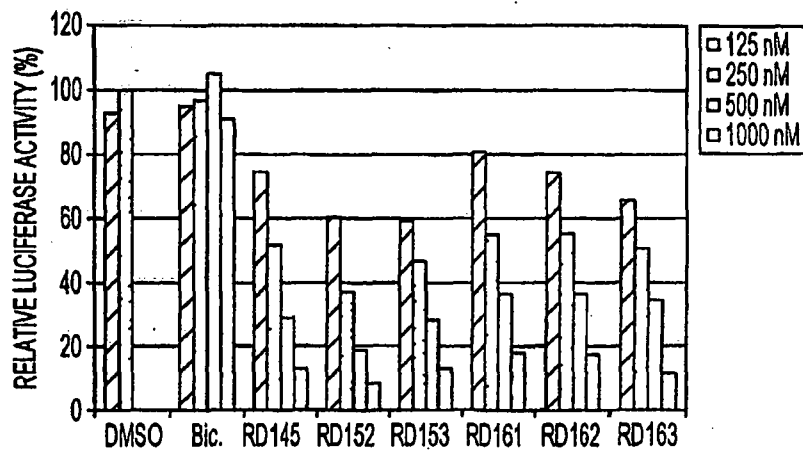


FIG. 3

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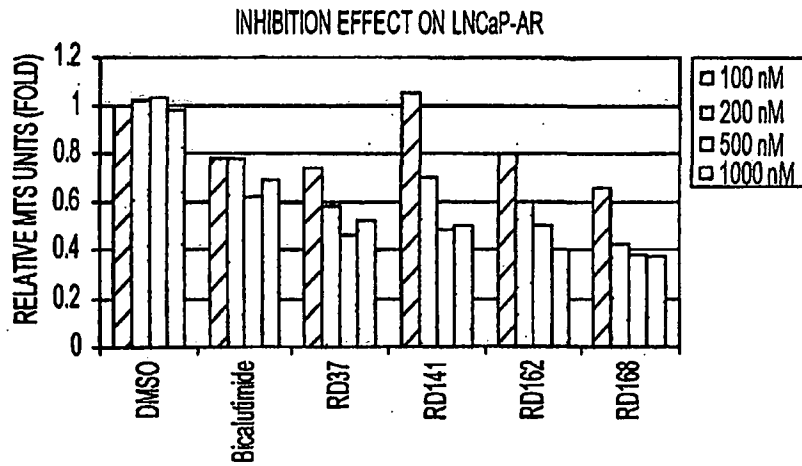


FIG. 5

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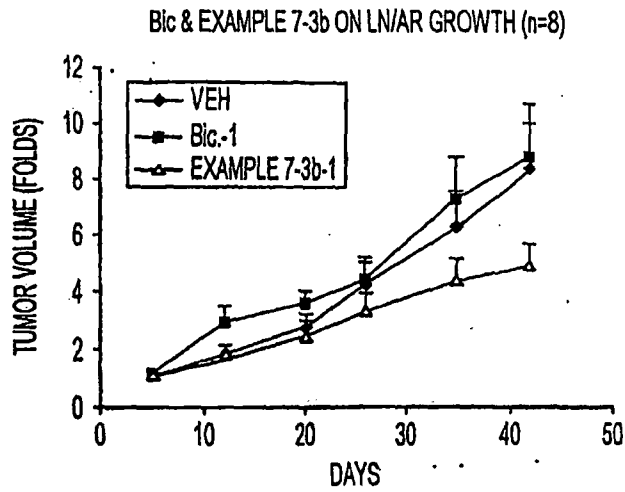


FIG. 7A

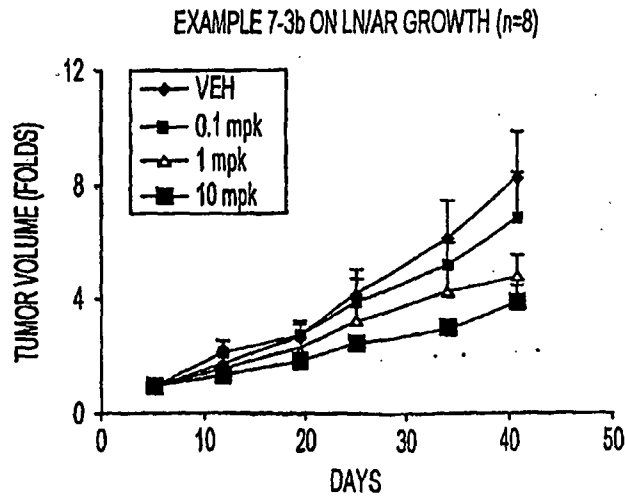


FIG. 7B

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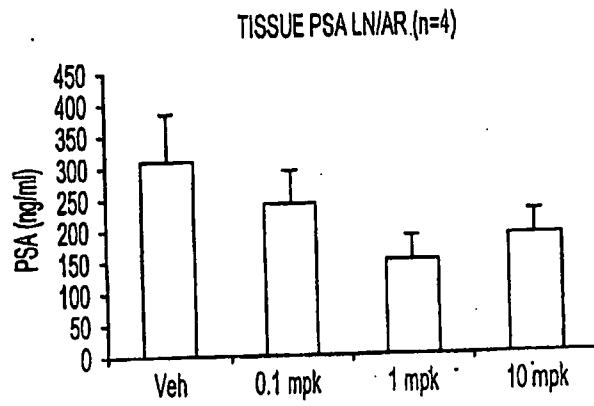


FIG. 8

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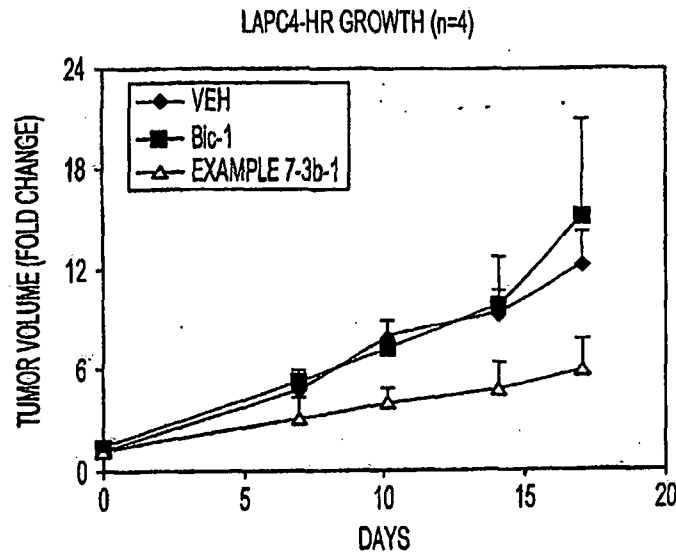


FIG. 9A

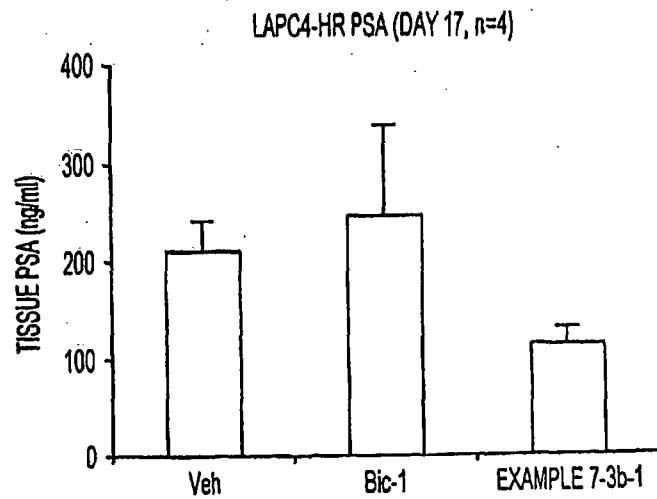


FIG. 9B

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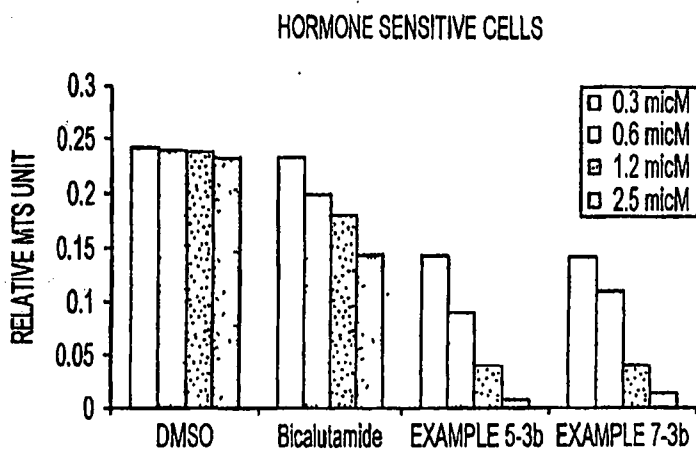


FIG. 10

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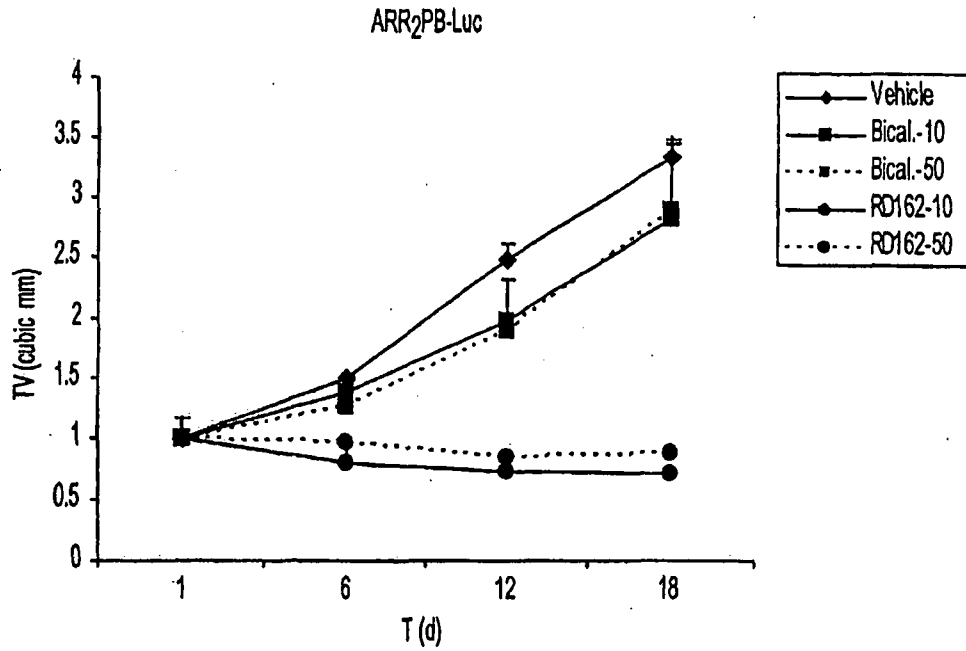


FIG. 11

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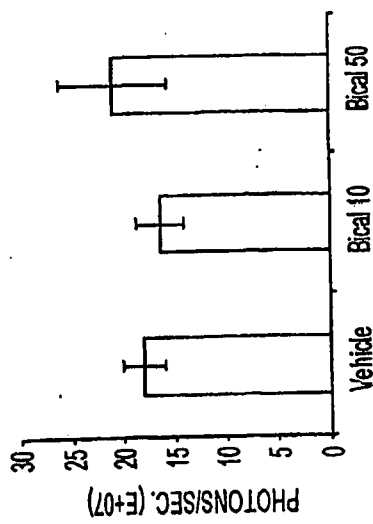
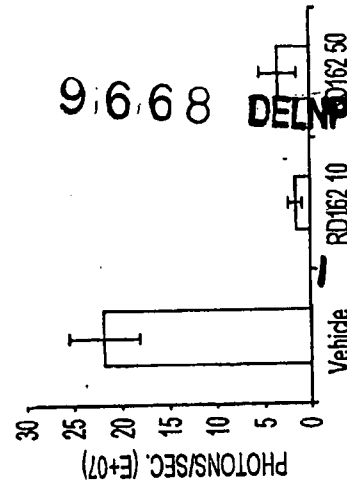


FIG. 12C



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 FIG. 12C

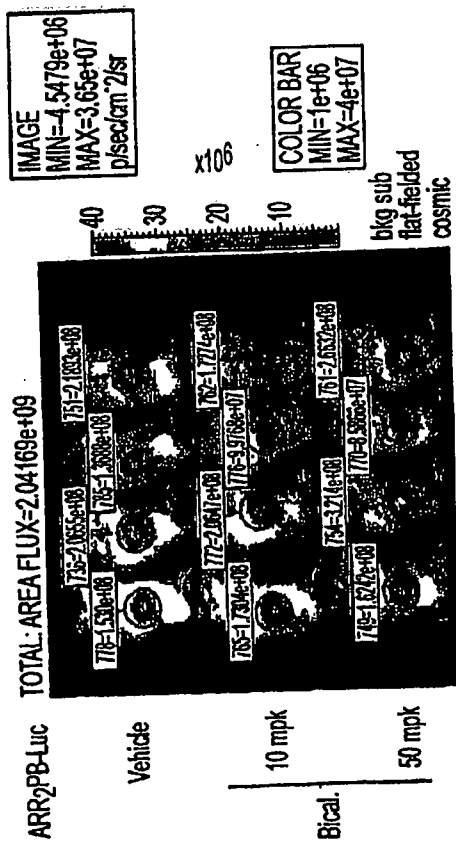


FIG. 12A

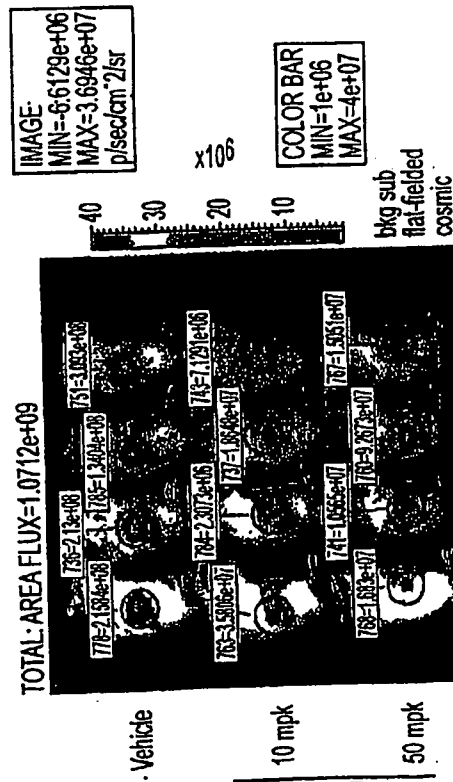


FIG. 12B

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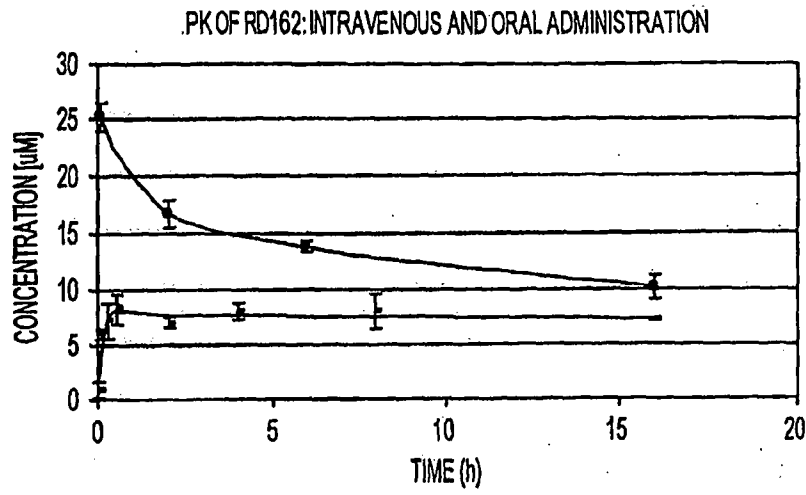


FIG. 13

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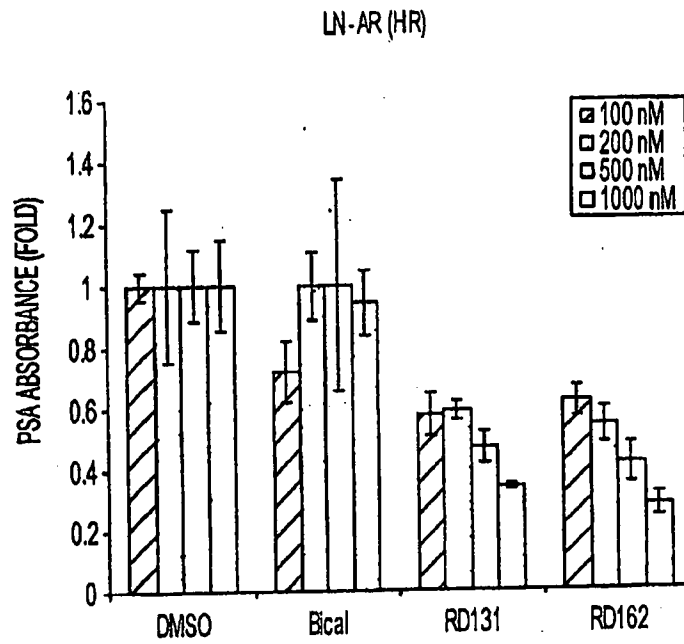


FIG. 14

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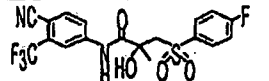
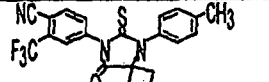
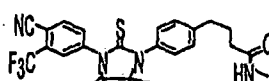
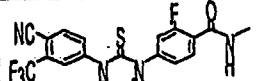
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CHARACTERISTICS OF Bicalutamide, RD37, RD131 AND RD162

NAME	STRUCTURE	IC ₅₀ [nM]	LogP	C _{SS} , 10mpk [μM]	C _{SS} , 25mpk [μM]	C _{SS} , 50mpk [μM]
Bic.		1000	2.91	10.0	11.4	11.9
RD37		124	4.20	NA	NA	NA
RD131		92	3.44	0.39	0.43	0.40
RD162		122	3.20	9.9	10.7	10.2

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FIG. 15A

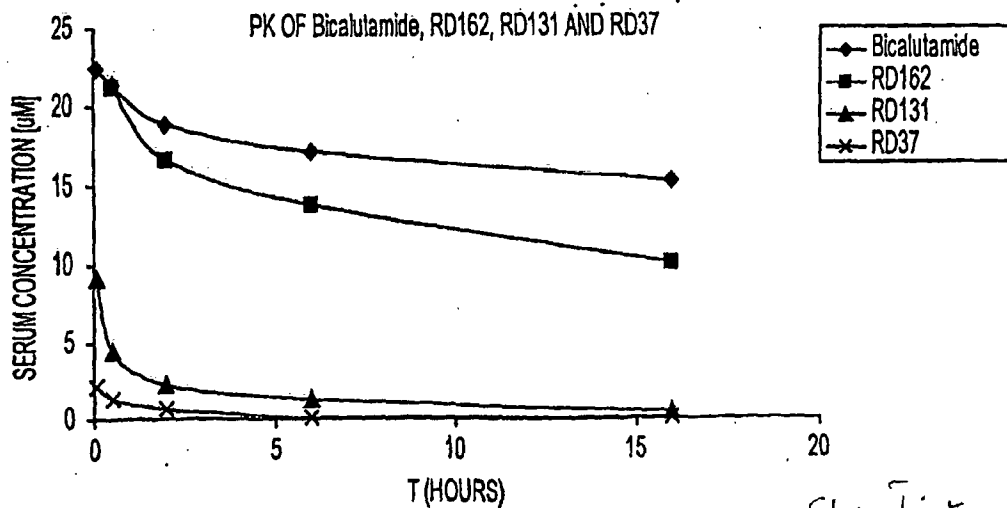


FIG. 15B

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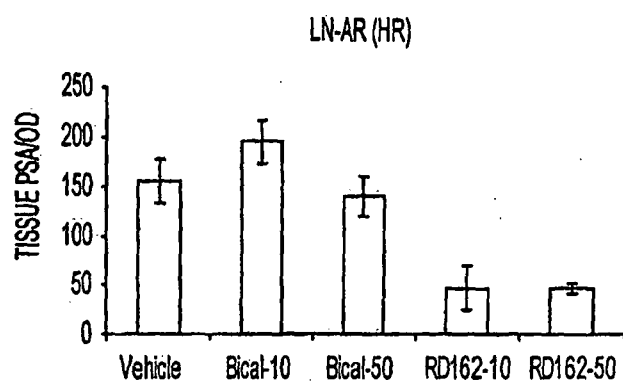


FIG. 17

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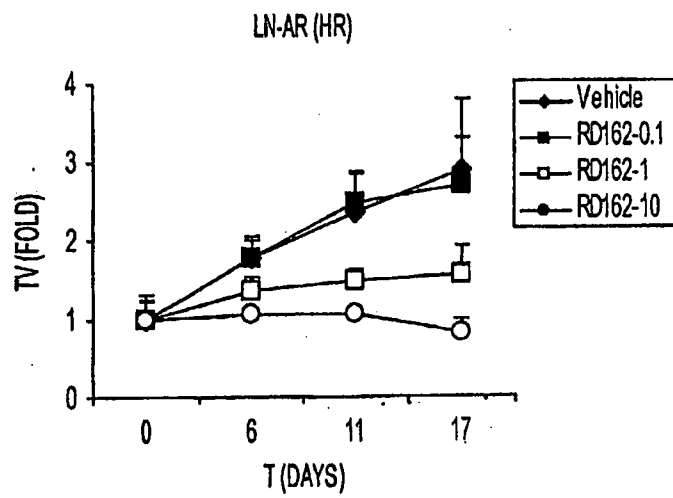


FIG. 18

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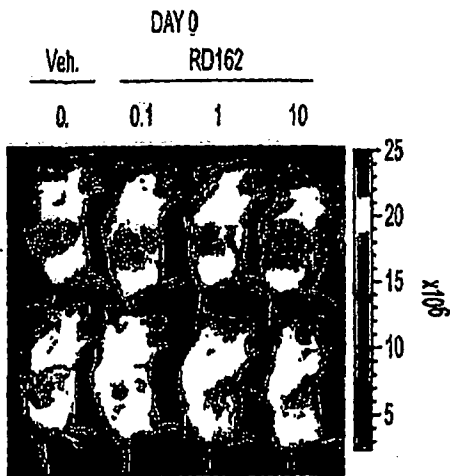


FIG. 19A

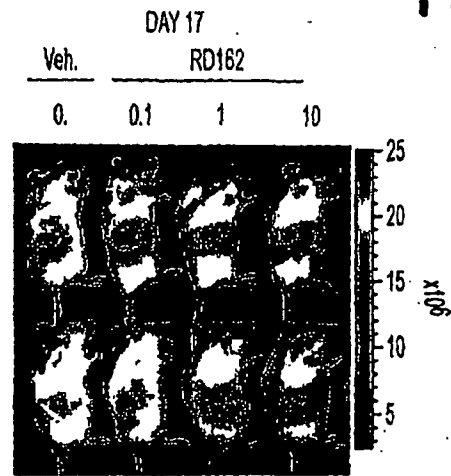


FIG. 19B

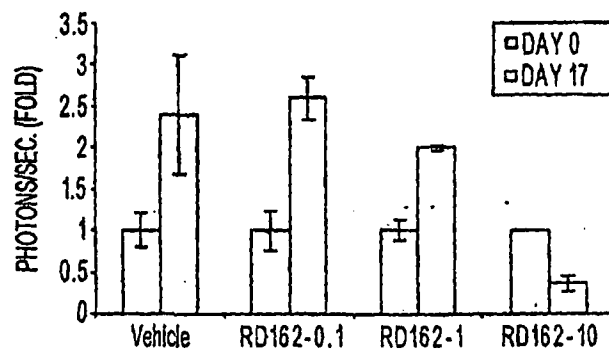


FIG. 19C

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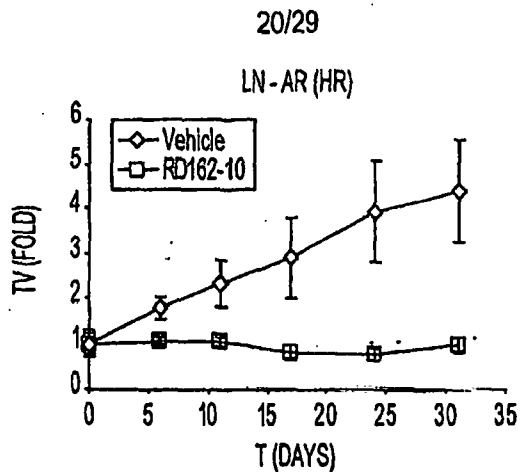


FIG. 20A

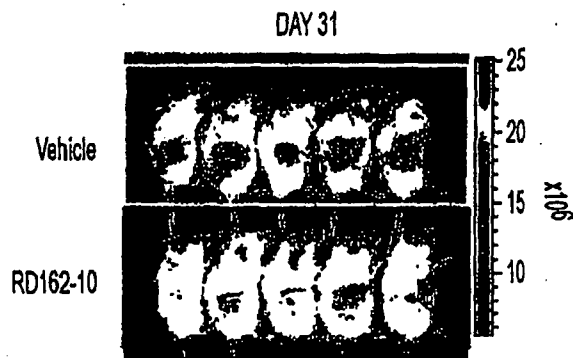


FIG. 20B

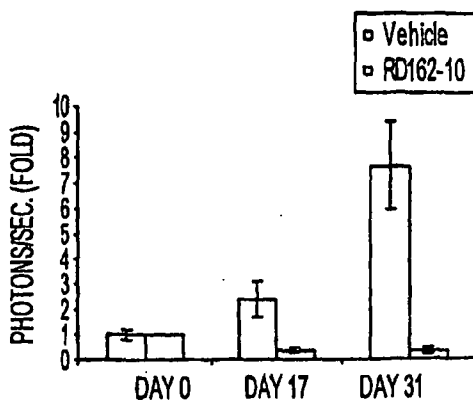


FIG. 20C

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11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41 42 43 44 45 46 47 48 49 50

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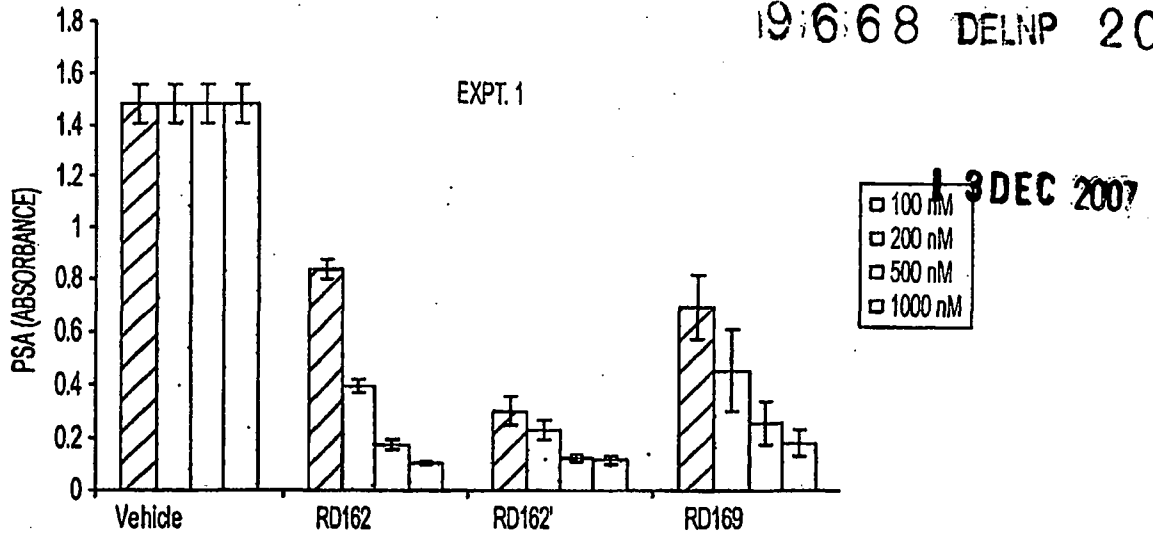


FIG. 21A

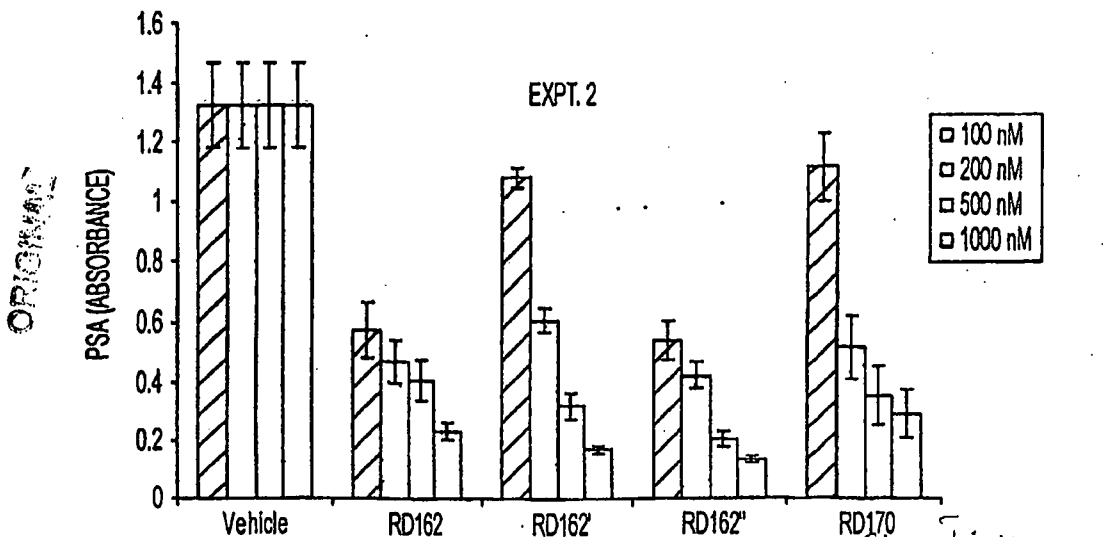


FIG. 21B

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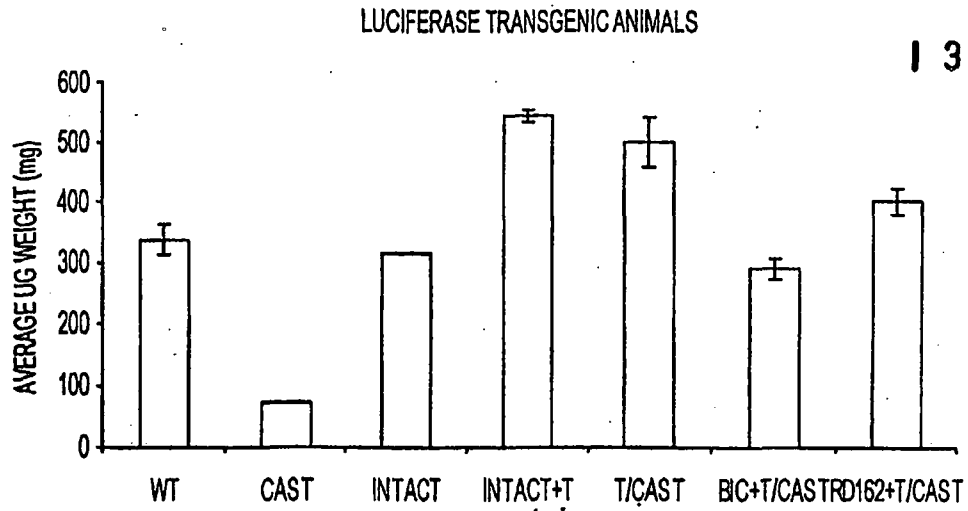


FIG. 23A

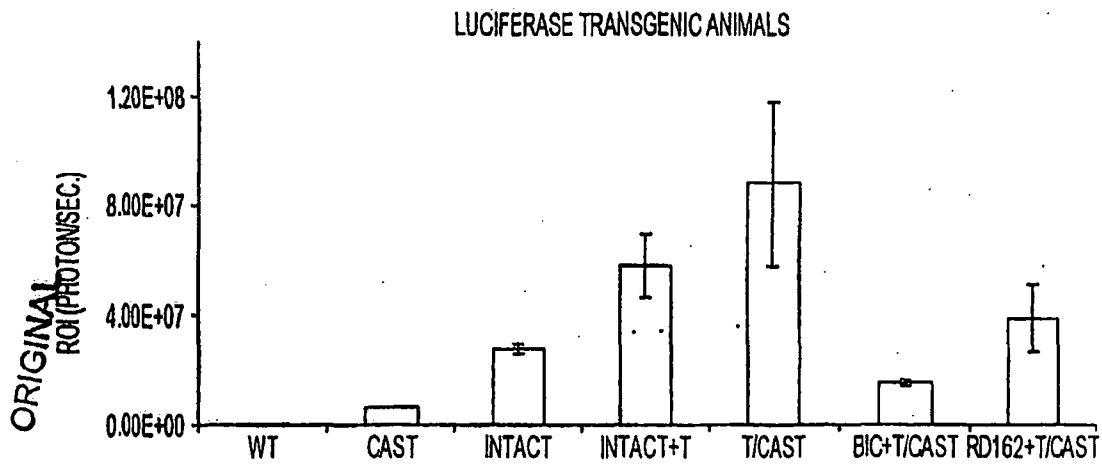


FIG. 23B

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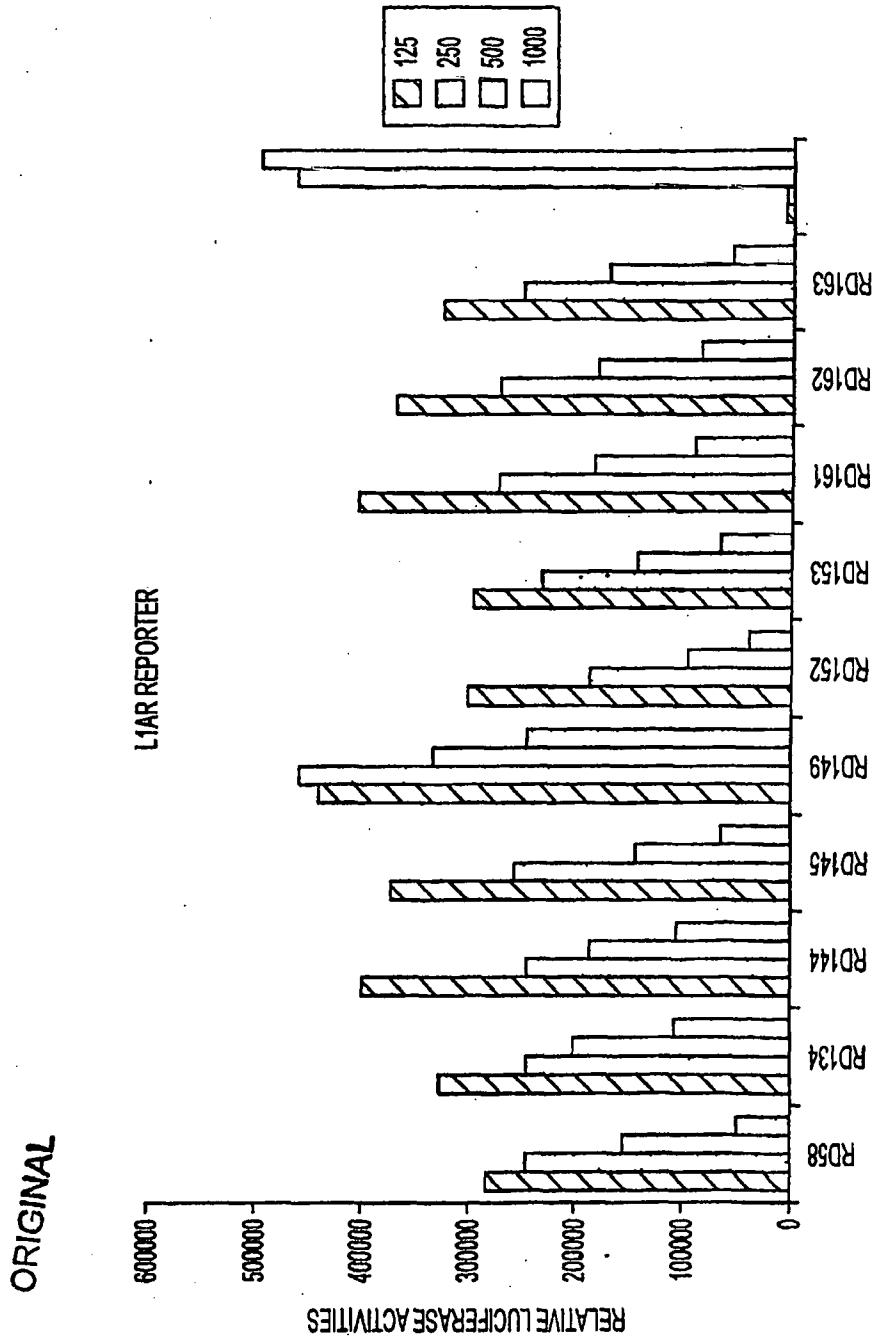


FIG. 24

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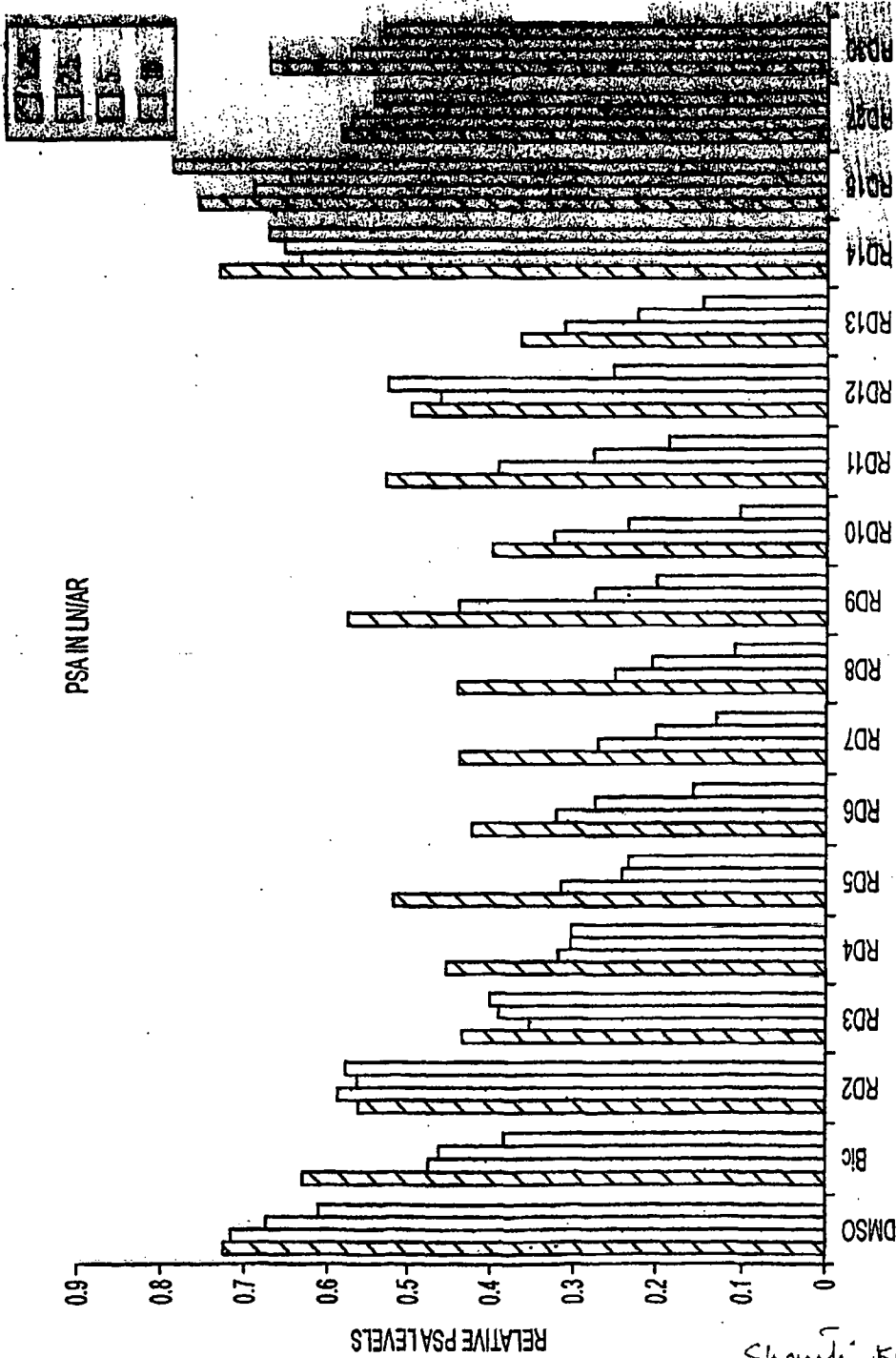


FIG. 25

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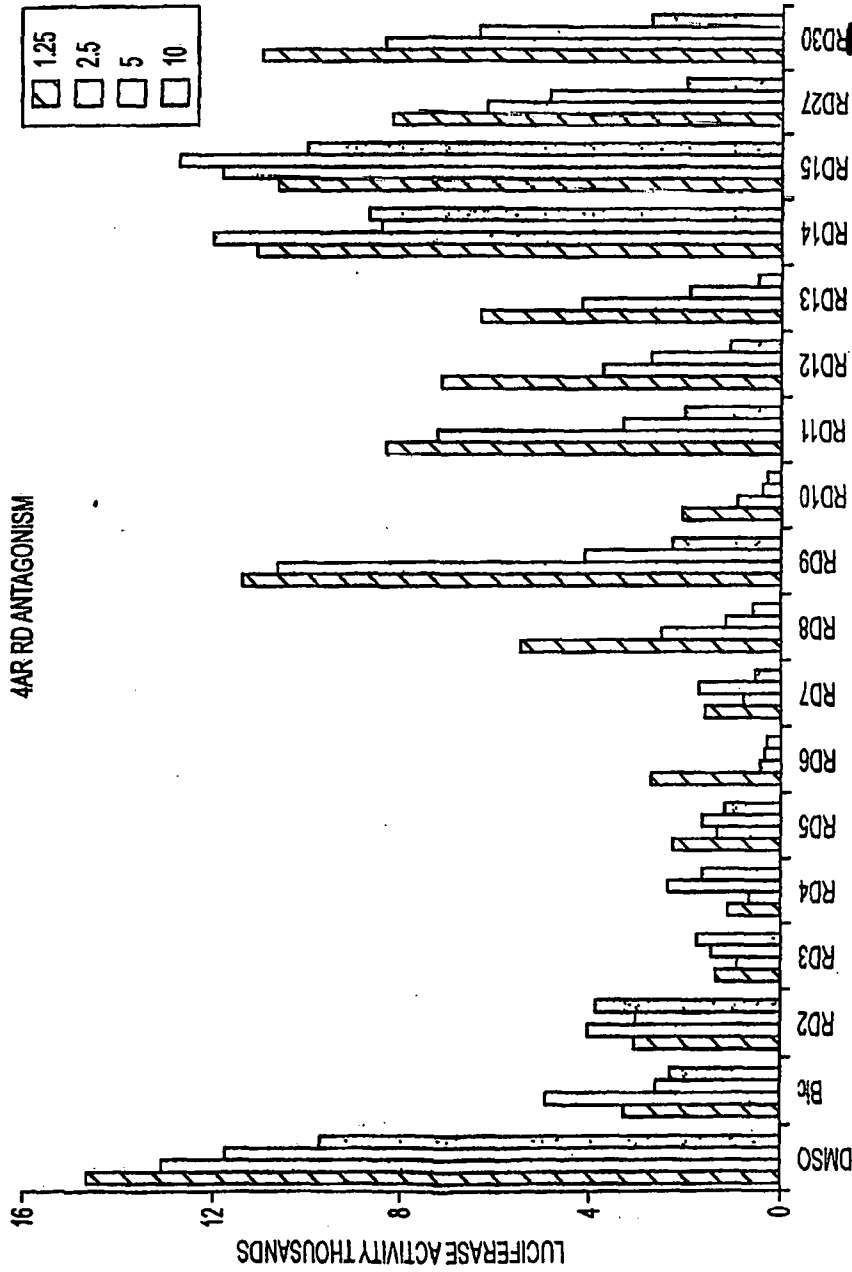


FIG. 26

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1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30

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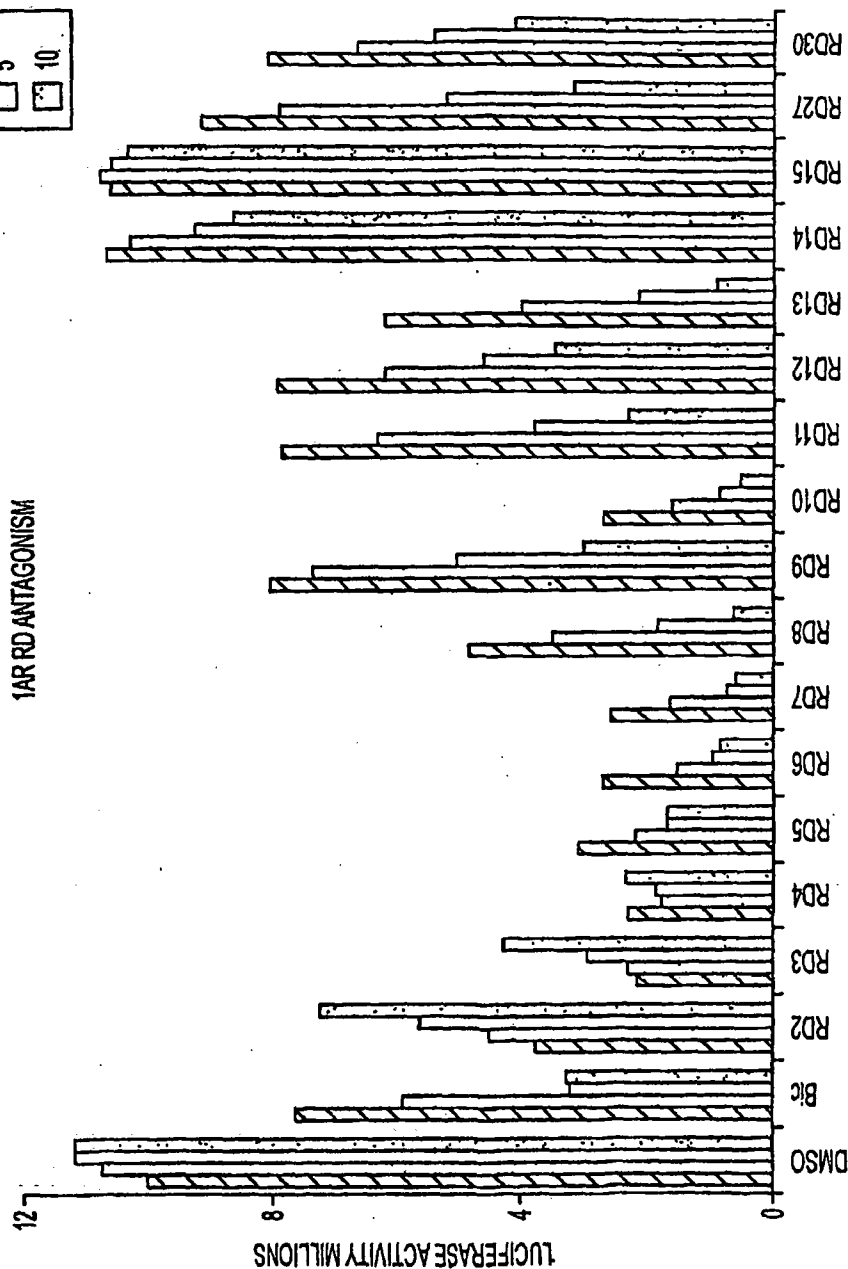
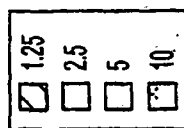


FIG. 27

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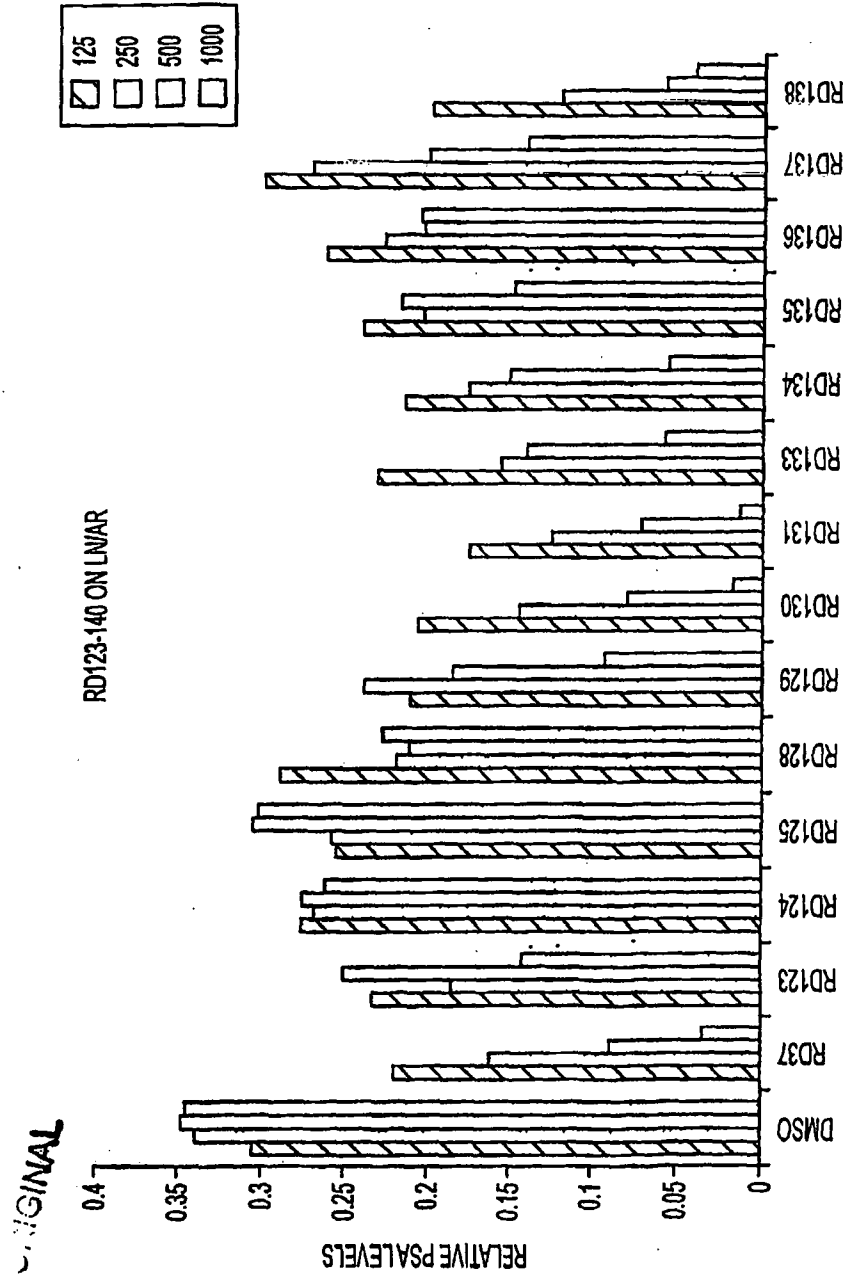


FIG. 28

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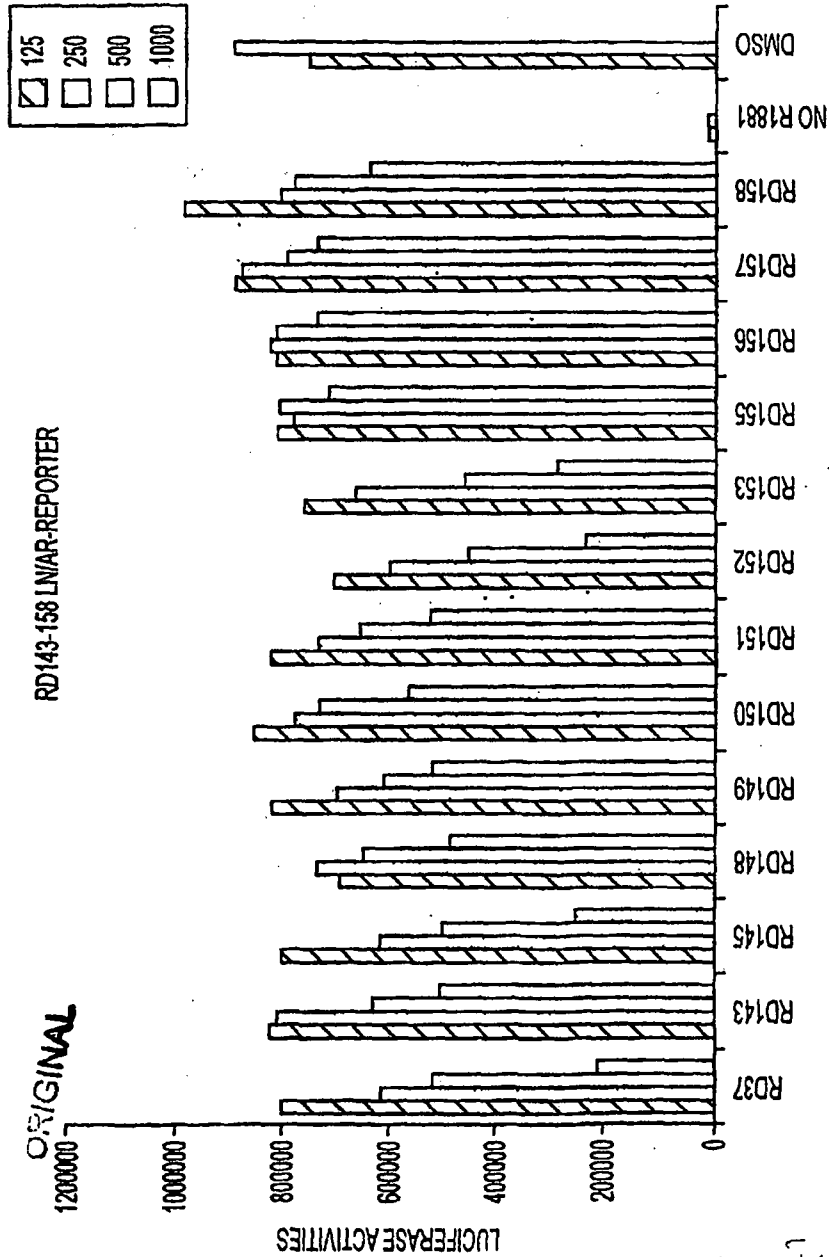


FIG. 29

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