

April 12, 2022

THE CONTROLLER OF PATENT
THE PATENT OFFICE
BOUDHIK SAMPADA BHAWAN, PLOT NO. 32
SECTOR 14, DWARKA, NEW DELHI-110078

Re: REPRESENTATION U/S 25(1) OF THE PATENT ACT – BY SANKALP
REHABILITATION TRUST AGAINST INDIAN PATENT APPLICATION NO.
201717035303 FILED ON 05/10/2017
APPLICANT: ATEA PHARMACEUTICALS INC.
R&A Ref. No.: OPP0402

Respected Sir,

We are filing this representation by way of Pre-Grant Opposition along with annexures u/s 25 (1) of the Patents Act, 1970 and Rule 55 of the Patent Rules, 2003 in Form 7A.

The Learned Controller is requested to take said opposition along with annexures on record and proceed further in the matter and keep the Opponent advised of each and every step taken in the matter.

We crave the leave of the Learned Controller to submit additional documents and/or evidence to support any of the averments in the representation as may be necessitated during the future proceeding.

Lastly, we request the Learned Controller to grant an opportunity of being heard before the present Opposition is finally decided.

Thanking you,

Yours faithfully,



RAJESHWARI H. IN/PA - 0358
OF RAJESHWARI AND ASSOCIATES
AGENT FOR THE OPPONENT

Encl: As stated

C.C.: LAKSHMI KUMARAN & SRIDHARAN
Email: iprdel@lakshmisri.com;

Also at: A - 202, First Floor, Shivalik Enclave, Malviya Nagar, New Delhi-110017

BEFORE THE CONTROLLER OF PATENTS, NEW DELHI

IN THE MATTER OF:

The Patents Act, 1970 as amended by the Patents (Amendment) Act 2005, and The Patents Rules, 2003, as amended by The Patents (Amendment) Rules, 2006

AND

IN THE MATTER of Pre-grant opposition under Section 25(1)

AND

IN THE MATTER of Indian Patent Application No. 201717035303

IN THE MATTER OF:

SANKALP REHABILITATION TRUST

.....OPPONENT

VS.

ATEA PHARMACEUTICALS INC.

.....APPLICANT

PRE-GRANT OPPOSITION BY SANKALP REHABILITATION TRUST

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7.	Annexure – 5: Copy of article P.A. Furman et al. Antiviral Research 91 (2011) 120–132	157-169
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Dated this day 12th of April, 2022



RAJESHWARI H. IN/PA – 0358
OF RAJESHWARI AND ASSOCIATES
AGENT FOR THE OPPONENT

TO,
THE CONTROLLER OF PATENTS
THE PATENT OFFICE, NEW DELHI

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, **SANKALP REHABILITATION TRUST**, having its registered office at SS Bengali Municipal School, First Floor, Thakurdwar Road, Charni Road East, Mumbai – 400002, India, hereby give Notice of opposition to the grant of patent in respect of Indian Patent Application No. 201717035303 filed on 05/10/2017 made by ATEA PHARMACEUTICALS INC. on the grounds.

- (a) Section 25(1)(b): Lack of novelty
- (b) Section 25(1)(e): Lack of inventive step
- (c) Section 25(1)(f): Invention is not patentable under section 3(d) and 3(e)
- (d) Section 25(1)(g): The complete specification does not sufficiently and clearly describe the invention or the method by which it is to be performed.
- (e) Section 25(1)(h): Failed to disclose to the Controller the information required by section 8.

(Detailed grounds are set out in the Opposition)

Our address for service in India is:

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Dated this 12th day of April, 2022



RAJESHWARI H. IN/PA – 0358
OF RAJESHWARI AND ASSOCIATES
AGENT FOR THE OPPONENT

TO
THE CONTROLLER OF PATENTS
PATENT OFFICE, NEW DELHI

BEFORE THE CONTROLLER OF PATENTS, THE PATENT OFFICE,
NEW DELHI

In the matter of Section 25(1) of The Patents Act, 1970 as amended by The Patents (Amendment) Act 2005;

And

In the matter of Rule 55 of The Patents Rules 2003 as amended by the Patent (Amendment) Rules, 2006

And

IN THE MATTER of Indian Patent Application 201717035303 dated 05/10/2017 in the name of **ATEA PHARMACEUTICALS INC.**

REPRESENTATION BY:

SANKALP REHABILITATION TRUST

..... OPPONENT

VS.

ATEA PHARMACEUTICALS INC.

.....APPLICANT

REPRESENTATION BY WAY OF PRE-GRANT OPPOSITION UNDER
SECTION 25(1) OF THE PATENTS ACT, 1970

We, **SANKALP REHABILITATION TRUST** an Indian organization, hereby submit our representation by way of opposition to the grant of patent in respect of Indian Patent Application 201717035303 dated 05/10/2017 in the name of **ATEA PHARMACEUTICALS INC.** entitled “ β D 2 DEOXY 2 A FLUORO 2 β C SUBSTITUTED 2 MODIFIED N6 SUBSTITUTED PURINE NUCLEOTIDES FOR HCV TREATMENT”.

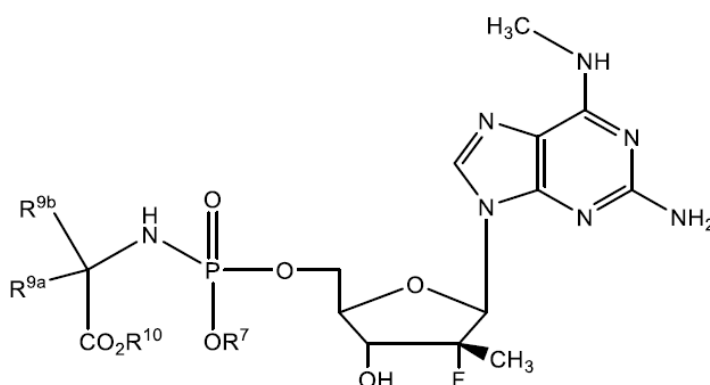
STATEMENT OF CASE OF OPPONENT

1. The Opponent has learnt that the Applicant has filed an Indian Patent Application No. 201717035303 (hereinafter “the Impugned Patent Application”) on 05/10/2017. The impugned patent application was published in the official journal of the patent office

on 01/12/2017, which is currently pending before the Patent Office. The Impugned Patent Application has a priority date of 06/03/2015.

2. The Impugned Patent Application is entitled “β D 2 DEOXY 2 A FLUORO 2 β C SUBSTITUTED 2 MODIFIED N6 SUBSTITUTED PURINE NUCLEOTIDES FOR HCV TREATMENT”.
3. The Opponent by way of this present pre-grant opposition submits that the claims currently pending on record are not patentable under the provisions provided in this Act. The claims as filed and currently on record are annexed herewith as **ANNEXURE-1** and reproduced herein below for ready reference:

1. A compound of the formula:



wherein:

R7 is C1-6alkyl, C3-7cycloalkyl, heteroaryl, heterocyclic, or aryl;

R9a and R9b are

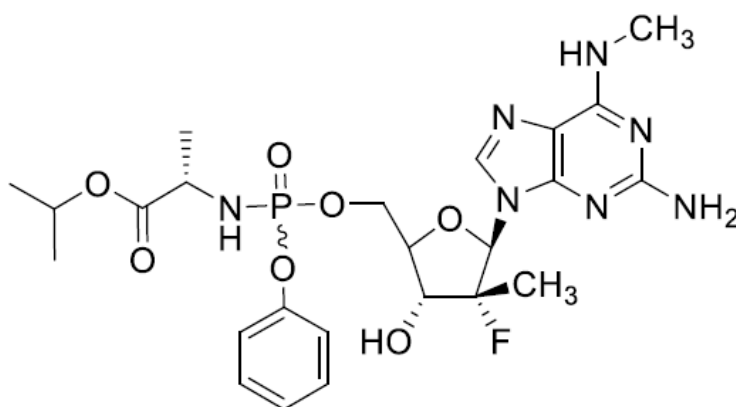
independently selected from hydrogen and C1-6alkyl; and

R10 is C1-6alkyl, C3-7cycloalkyl, heterocycloalkyl, aryl or heteroaryl;

or a pharmaceutically acceptable salt thereof.

2. The compound of claim 1, wherein R7 is heteroaryl, heterocyclic, or aryl.
3. The compound of claim 1, wherein:
R7 is phenyl, R9a is hydrogen, R9b is methyl, and R10 is isopropyl.
4. The compound of claim 1, wherein: R7 is aryl.
5. A pharmaceutical composition comprising an effective amount of the compound of claim 1 in a pharmaceutically acceptable carrier.

6. A pharmaceutical composition comprising an effective amount of the compound of claim 4 in a pharmaceutically acceptable carrier.
7. A pharmaceutical composition comprising an effective amount of the compound of claim 3 in a pharmaceutically acceptable carrier.
8. The pharmaceutical composition of claim 5, wherein the composition is suitable for oral delivery.
9. The pharmaceutical composition of claim 6, wherein the composition is suitable for oral delivery.
10. The pharmaceutical composition of claim 7, wherein the composition is suitable for oral delivery.
11. A compound of the formula:

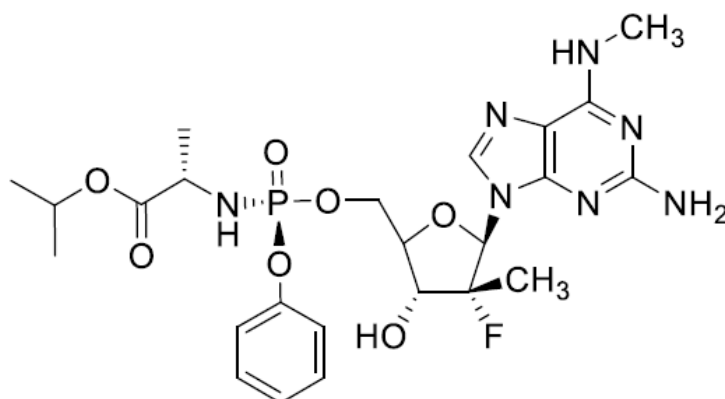


or a pharmaceutically acceptable salt thereof.

12. A pharmaceutical composition comprising the compound of claim 11; or a pharmaceutically acceptable salt thereof in a pharmaceutically acceptable carrier.
13. The compound of claim 1,
wherein
R7 is aryl; and
R10 is C1-6alkyl
and wherein the compound is in the form of an isolated phosphorus S-enantiomer and is at least 90% free of the opposite phosphorus R-enantiomer.
14. A pharmaceutical composition comprising the compound of claim 13 in a pharmaceutically acceptable carrier.
15. The compound of claim 1,
R7 is aryl; and
R10 is C1-6alkyl;

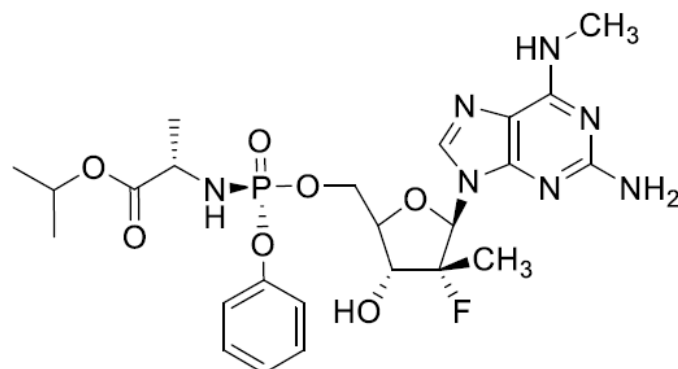
and wherein the compound is in the form of an isolated phosphorus R-enantiomer and is at least 90% free of the opposite phosphorus S-enantiomer.

16. A pharmaceutical composition comprising the compound of claim 15 in a pharmaceutically acceptable carrier.
17. A compound of claim 1, wherein R7 is phenyl.
18. The compound of claim 11, wherein the compound is at least 90% free of the opposite phosphorus R-enantiomer.
19. The compound of claim 11, wherein the compound is at least 90% free of the opposite phosphorus S-enantiomer.
20. The compound of claim 18, wherein the compound is at least 98% free of the opposite phosphorus R-enantiomer.
21. The compound of claim 19, wherein the compound is at least 98% free of the opposite phosphorus S-enantiomer.
22. The compound of claim 11 of the formula:



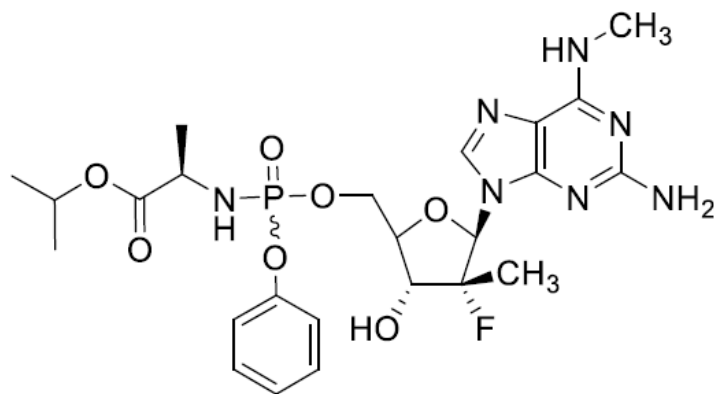
or a pharmaceutically acceptable salt thereof.

23. The compound of claim 11 of the formula:



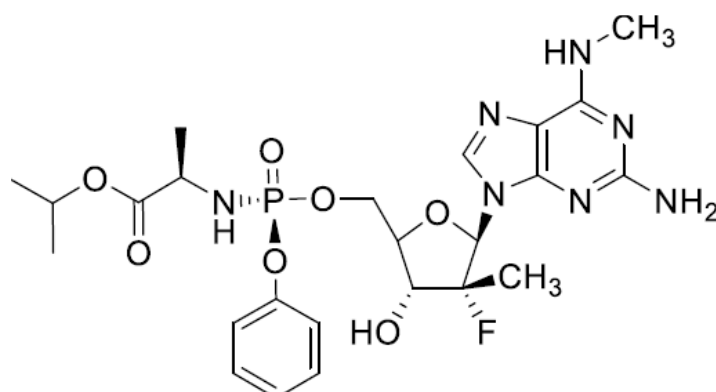
or a pharmaceutically acceptable salt thereof.

24. A compound of the formula:



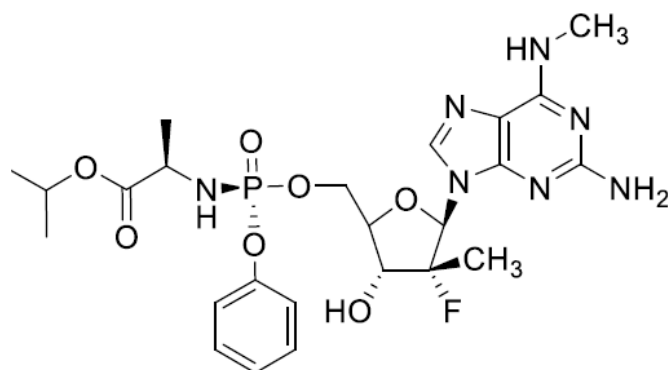
or a pharmaceutically acceptable salt thereof.

25. The compound of claim 24 of the formula:



or a pharmaceutically acceptable salt thereof.

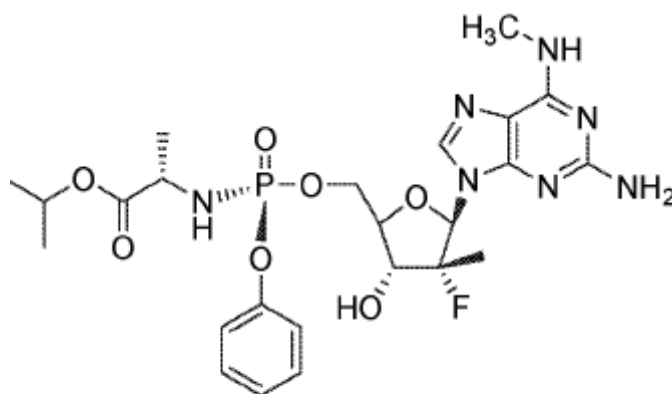
26. The compound of claim 24 of the formula:



or a pharmaceutically acceptable salt thereof.

27. A pharmaceutical composition comprising the compound of any one of claims 22-26 in a pharmaceutically acceptable carrier.
28. A pharmaceutical composition comprising the compound of claim 22 in a pharmaceutically acceptable carrier.
29. A pharmaceutical composition comprising the compound of claim 23 in a pharmaceutically acceptable carrier.

30. The compound of claim 1, wherein R7 is naphthyl.
 31. The compound of claim 1, wherein R10 is C1-6alkyl or C3-7cycloalkyl.
 32. The compound of claim 1, wherein R10 is C1-6alkyl.
 33. The compound of claim 32, wherein R10 is C1-4alkyl.
 34. The compound of claim 33, wherein R10 is selected from methyl, ethyl, n-propyl, and isopropyl.
 35. The compound of claim 33, wherein R10 is iso-propyl.
 36. The compound of claim 1, wherein R10 is aryl or heteroaryl.
 37. The compound of claim 36, wherein R10 is phenyl.
 38. The compound of claim 36, wherein R10 is naphthyl.
4. **Impugned Patent Application:** The present pre-grant opposition is against Indian Patent Application 201717035303 dated 05/10/2017 in the name of ATEA PHARMACEUTICALS INC. entitled “ β D 2 DEOXY 2 A FLUORO 2 β C SUBSTITUTED 2 MODIFIED N6 SUBSTITUTED PURINE NUCLEOTIDES FOR HCV TREATMENT” and is drawn towards nucleotide compounds and compositions and uses thereof to treat the Hepatitis C virus ("HCV").
 5. **Disclosure in the impugned patent application:** As per the Applicant, the present invention relates to the compounds of Formula I, Formula II, Formula III, Formula IV, Formula V, Formula VI, Formula VII and including P-D-2'-deoxy-2'-a-fluoro-2'-PC-substituted-N 6-(mono- or di-methyl) purine nucleotides, are highly active against the HCV virus when administered in an effective amount to a host in need thereof. The host can be a human or any animal that carries the viral infection. It further discloses a specific compound which is represented as 5-2 and the structure is given below.



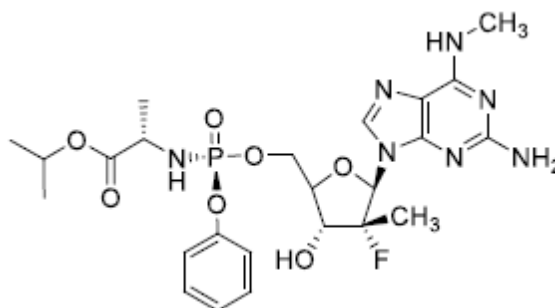
Compound 5-2 (Table 7)

Fig 1: Compound 5-2

6. **PRIOR ARTS:** The Opponent wishes to rely on the following prior arts as evidence in support of the grounds of opposition.
 - i. **D1-** US 2010/0016251 (US'251) published on 21 Jan 2010 (annexed herewith as **Annexure – 2**).
 - ii. **D2-** US 2011/0257121 (US'121) published on 20 Oct. 2011 (annexed herewith as **Annexure – 3**).
 - iii. **D3-** Priyanka L. Gaikwad et al; The Use of Bioisosterism in Drug Design and Molecular Modification; Am. J. PharmTech Res. 2012; 2(4), 1-23 (annexed herewith as **Annexure – 4**).
 - iv. **D4-** P.A. Furman et al. Antiviral Research 91 (2011) 120–132 (annexed herewith as **Annexure – 5**).
7. It is submitted that all claims of the impugned patent application are liable to be refused on following grounds as below:
 - (a) Section 25(1)(b): Lack of novelty
 - (b) Section 25(1)(e): Lack of inventive step
 - (c) Section 25(1)(f): Invention is not patentable under section 3(d) and 3(e)
 - (d) Section 25(1)(g): The complete specification does not sufficiently and clearly describe the invention or the method by which it is to be performed.
 - (e) Section 25(1)(h): Failed to disclose to the Controller the information required by section 8.

(a) GROUND 1: LACK OF NOVELTY: Section 25(1)(b)

8. It is submitted that claims of the impugned application are not novel, and therefore have to be rejected under section 25(1)(b) of the Act.
9. The impugned patent application lacks novelty in view of US 2010/0016251(US'251). This document was published on 21 January 2010 which is prior to priority date of impugned patent application.
10. The impugned patent application claims compounds including the compound, denoted as "Compound 5-2" in the specification and pharmaceutical composition thereof. The compound named 5-2 in the impugned specification has the following structure:



Compound 5-2

11. It is submitted that US'251 discloses phosphoramidate prodrugs of nucleoside derivatives for the treatment of viral infections in mammals, which is a compound, its stereoisomer, salt (acid or basic addition salt), hydrate, solvate, or crystalline form. It is further submitted that the compound 5-2 of impugned patent application is also disclosed in US'251.
12. It is further submitted that US'251 also disclose a composition for the treatment of any of the viral agents and said composition comprising a pharmaceutically acceptable medium selected from among an excipient, diluent, and equivalent medium and a compound, that is intended to include its salts (acid or basic addition salts), hydrates, Solvates, and crystalline forms can be obtained, represented by formula I.
13. It is submitted that a compound of formula of claim 1 of impugned patent application including the compound 5-2 of impugned patent application is disclosed in prior art US'251. The table given below depicts that the compound 5-2 is covered in WO251 and can be obtained by making appropriate substitutions, as per the teaching of US'251, in the Markush of US'251:

Table 1:

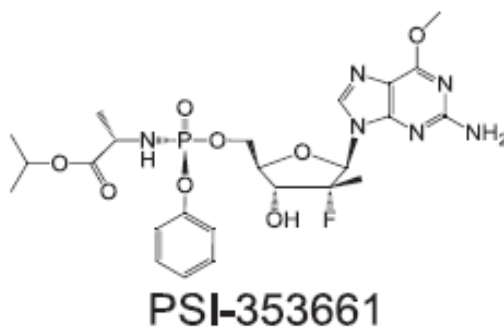
S. No.	Markush Structure	Resultant structure
1	<p>Para [0018]</p>	
2	R ₁ is phenyl (Para 0020)	
3.	R ₂ is H (Para 0021)	
4.	R _{3a} is H and R _{3b} is alkyl (methyl) (Para 0022)	
5.	R ₄ is alkyl (isopropyl) (Para 0023)	
6.	R ₅ is H (Para 0024)	

7.	Y is OH (Para 0027)	
8.	X is F (Para 0026)	
9.	R6 is CH ₃ (Para 0025)	
10.	Base is 	
11.	Z is N (Para 0030)	
12.	R10 is NHR' (Para 0031) And R' is alkyl (methyl) (Para 0032)	
13.	R11 is NH ₂ (Para 0031)	

14. The above table discloses that compound 5-2 is disclosed in US'251. Moreover, US'251 also disclose the composition of such compounds in pharmaceutically acceptable excipients and their use in therapy.
15. Therefore, the subject matter claimed in the impugned application is disclosed in the prior art. Hence, impugned application lacks novelty over US'251 and ought to be rejected on this ground alone.

(b) GROUND 3: LACK OF INVENTIVE STEP: Section 25(1)(e)

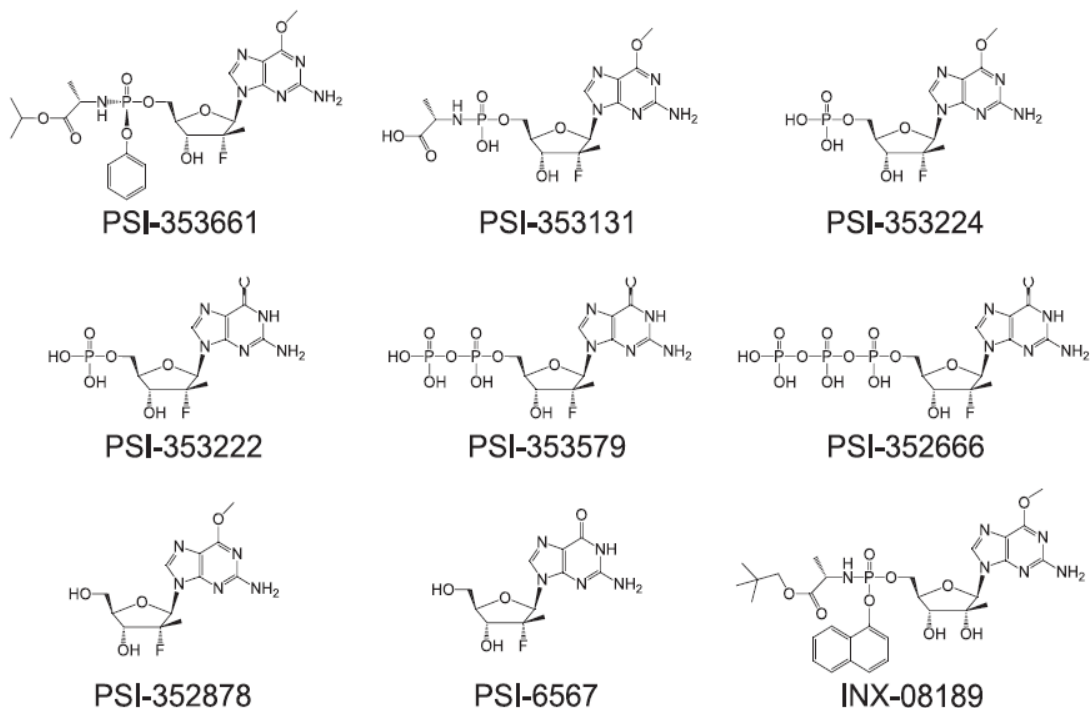
16. It is submitted that the invention as claimed is obvious and does not involve any inventive step in view of whatever was known and published in India or elsewhere prior to the priority date of impugned patent application i.e. prior to 06/03/2015 the earliest claimed priority.
17. It is submitted that the claimed subject matter of the impugned patent application are not inventive and are obvious in view of common general knowledge in art and combined with teachings of above-mentioned prior arts.
18. Furman et al. discloses the compound denoted as PSI-353661 therein which is stated to be an effective HCV antiviral since it is disclosed that PSI-353661 clears HCV replicon RNA and prevents viral rebound. The structure of PSI-353661 is as shown below:



19. As per the disclosure of Furman et al PSI-353661 exhibits inhibition of HCV replicon RNA and infectious viral replication and the EC50 and EC90 values for PSI-353661 were 0.0030 ± 0.0014 IM and 0.0085 ± 0.0007 IM, respectively,
20. In vitro safety profile of PSI-353661 was evaluated for cytotoxicity in an 8-day assay using four different human derived cell lines: Huh7 (human hepatoma), HepG2 (human hepatoma), CEM (human T lymphocyte), and BxPC3 (human pancreatic) cells. The

results showed no measurable cytotoxicity for PSI-353661 toward HepG2, BxPC-3, or CEM cells ($CC_{50} > 100$ IM). The CC_{50} value determined for PSI-353661 with Huh 7 cells was 80.0 ± 6.0 IM.

21. Mitochondrial toxicity has been associated with long-term use of some nucleoside analogs - As a measure of mitochondrial toxicity the effect of PSI-353661 on mitochondrial DNA content and lactic acid production was assessed. Exposing CEM and HepG2 cells to PSI-353661 did not affect mitochondrial DNA content and there was no increase in lactic acid.
22. The effect of PSI-353661 on the proliferation of human erythroid and myeloid progenitor cells was evaluated using a 14-day colony formation assay - PSI-353661 was not significantly toxic toward bone marrow progenitor cells.
23. PSI-353661 was assessed for activity against replicons harbouring the NS5B S282T or S96T/N142T amino acid alterations that confer decreased susceptibility to nucleoside/tide analogs. Neither the S96T/N142T amino acid alterations nor the S282T amino acid alteration conferred resistance to PSI-353661. The EC_{90} value for the wild-type replicon and the S96T/N142T replicon were 0.012 ± 0.004 and 0.009 ± 0.004 IM, respectively.
24. PSI-353661 was tested against replicons containing amino acid alterations that confer resistance to non-nucleoside inhibitors of NS5B. Results showed that PSI-353661 remained fully active against HCV replicons harbouring any one of these resistant mutations, which conferred various levels of resistance towards each of the corresponding reference compounds.
25. Furman also discloses that the compound PSI-353661 metabolizes into various metabolites including an active metabolite wherein the phosphoramidate derivative is converted to triphosphate and methoxy group is metabolized to oxo group, however, rest of the molecule remains intact.



26. It is submitted that US'121 discloses compounds, pharmaceutically acceptable salt and their compositions that are inhibitors of RNA-dependent RNA viral replication and are useful as inhibitors of HCV NS5B polymerase, as inhibitors of HCV replication and for treatment of hepatitis C infection in mammals.
27. It is submitted that US'121 further discloses the activity data i.e. EC₉₀ (0.02uM) in replicon assay for a compound which is given below. The data reflects that this compound possesses promising antiviral activity.

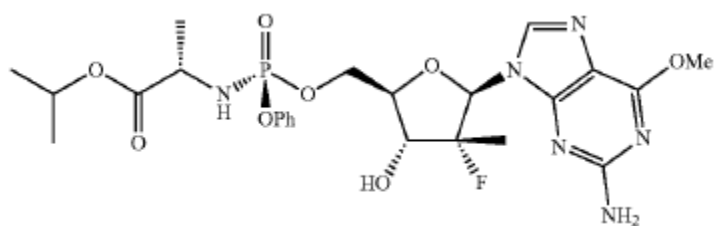


Fig. 2: Compound 1 of US'121

28. To express the antiviral effectiveness of a compound, the EC₅₀ for reduction of HCV RNA to 90% was found to be 0.02μM.
29. It is submitted that Priyanka et al discloses that in drug design the purpose of exchanging one bioisostere for another is :

1. To enhance the desired biological or physical properties of a compound without making significant changes in chemical structure.
 2. To attenuate toxicity.
 3. To modify the activity of the lead compound.
 4. To alter the metabolism of the lead.
30. Furthermore, Priyanka et al further discloses that –O- group can be interchanged with –NH-, amino group.
31. Thus, in light of the disclosure of Furman et al, US'121 and Priyanka et al a person skilled in the art engaged in the pursuit of designing an alternative anti-viral molecule of PSI-353661 gets a teaching to change the oxo group in the methoxy group since the phosphoramidate group is important for change into triphosphate group which in turn is important for pharmacological activity of the compound.
32. Therefore, the most viable substituent to replace in order to achieve an alternative molecule is oxo group of methoxy which may be replaced with amino group as per Priyanka et al.
33. Thus, the claimed compound 5-2 is obvious in light of combined teachings of prior art documents discussed above.
34. Further, the Applicant has failed to provide any data in the specification to establish technical advancement in the field of invention. The Applicant has provided data in the specification as compared to the parent nucleoside rather than providing comparative data as compared to the molecule known in the art at the time of the invention such as the compound PSI-353661.
35. Furthermore, the biological activity data provided in the specification does not pertain to the claimed compounds.
36. Therefore, Applicant has failed to establish any technical advancement of the claimed subject matter over what was already known in the art at the time of the invention. Therefore, the impugned patent application lack inventive step.

37. Considering above, the present impugned patent application lack inventive step and therefore, this application should be rejected on this ground alone.

(c) GROUND 4: Claims not patentable under Section 25(1)(f)

38. It is submitted that the impugned patent application should not be allowed under Section 3(d) which states that “*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.”

39. The Opponent submits that the applicant has failed to demonstrate any enhancement in the therapeutic efficacy with respect to the known in prior art. The Opponent states that the Applicant miserably failed to provide data demonstrating enhanced 'therapeutic' over known substance disclosed art.
40. It is further submitted that PSI-353661(compound 1 of US'121) was a known molecule and Applicant has failed to establish enhanced therapeutic efficacy of the claimed compound with respect to PSI-353661(compound 1 of US'121). PSI-353661(compound 1 of US'121) was the closest compound which was known in art before the priority date of impugned patent application. There was only one difference and i.e. presence of -NH (impugned molecule) in place of O (prior art molecule) at bases of the molecules. Thus, compound 5-2 of impugned application can be considered as new form of the known compound PSI-353661(compound 1 of US'121) and it was incumbent upon the Applicant to demonstrate enhancement in the therapeutic efficacy with respect to PSI-353661(compound 1 of US'121), which the Applicant has failed to do.

41. Therefore, the claimed subject matter of the impugned patent application falls under barr of Section 3(d) of the Act and is thus not patentable. Hence the impugned patent application should be rejected under this ground alone.

Claims of impugned application is not patentable as per section 3(e)

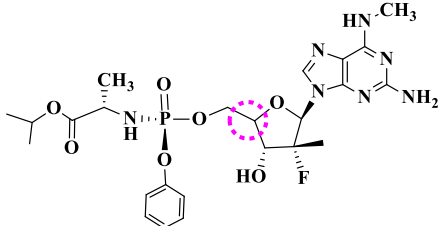
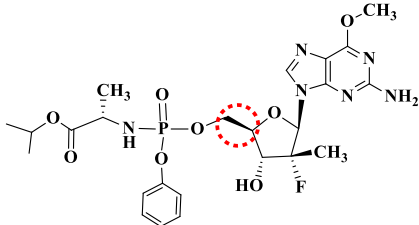
42. The Opponent states that the claimed invention clearly falls under the section 3(e) which clearly states that a, substance obtained by a mere admixture resulting only in the aggregation of the properties of the components thereof or a process for producing such substance is not considered as an invention and not patentable.
43. The Opponent submits that the subject matter of Claims 27 to 29 is drawn to compositions. The Applicant has failed to provide any data demonstrating any unexpected effect of the claimed composition.
44. In absence of any comparative data highlighting the synergistic effect of the components of the claimed composition of the impugned patent over its individual components, the claims of the impugned patent application fall under the purview of Section 3(e) of the Act.
45. Further, the claimed composition in claim 27 to 29 is not defined in terms of its constituent's ratio/percentage which are essential features of a composition claim in order to establish the precise range of ratio/percentage of the constituents in which the composition exhibits synergistic effect.
46. Thus, impugned application is liable to be rejected on this ground alone.

(d) GROUND 5: INSUFFICIENCY OF DISCLOSURE

47. The complete specification does not sufficiently and clearly describe the invention or the method by which it is to be performed.
48. The Opponent states that the specification should clearly and fairly describe the invention and disclose the best mode of working the invention so that the person skilled in the art could perform the invention without any undue efforts and it is hereby stated that the applicant has failed to do so.

49. It is submitted that claims are not fairly based on the specification and complete specification does not describe the invention and the method of performing the invention.
50. It is submitted that Applicant has mentioned the activity of the compound 5-2 i.e. 4 nM, which is at least a 3900 fold increase in activity. It is further submitted that compound 5-2 is a mixture of isomers. The basic skeleton of impugned molecule is same with respect to prior art molecule and prior art molecule shows the possibility of isomers at 4' position of pentose sugar. The relevant figure is given below.

Table 2:

Impugned molecule (5-2)	
Compound 1 of US'121	

51. It is submitted that the above mentioned table 2 clearly shows that compound 5-2 is mixture of isomers and the data provided by applicant does not pertain to claimed compound. Therefore, impugned application does not sufficiently and clearly describe the invention.
52. It is further submitted that the entire data provided in table 7 is for mixture of isomers and not for a specific compound. As mentioned above, 4' position of pentose sugar will contain isomers. The said observation is also supported by scheme 1, 2 and 3 of impugned application (Page 37-39). The relevant figure is given below.

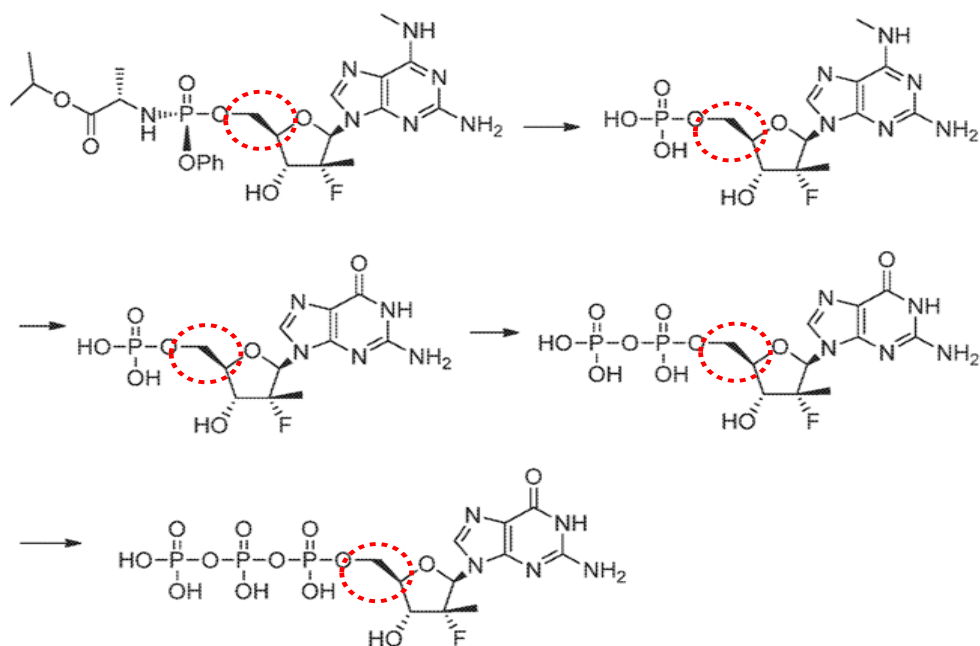


Figure: Scheme 1

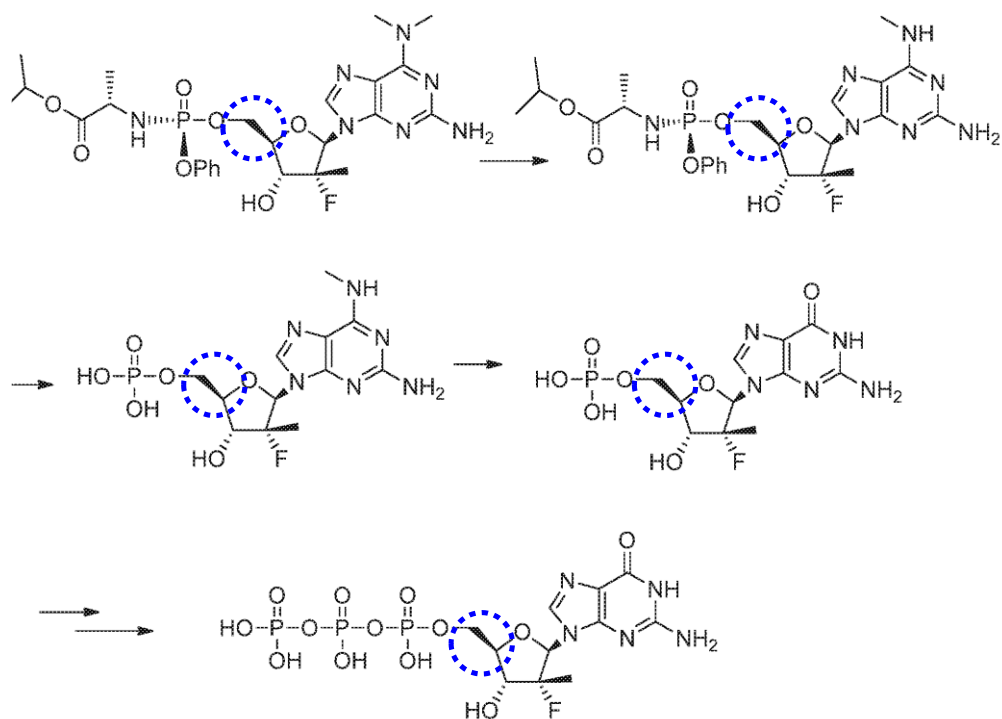


Figure: Scheme 2

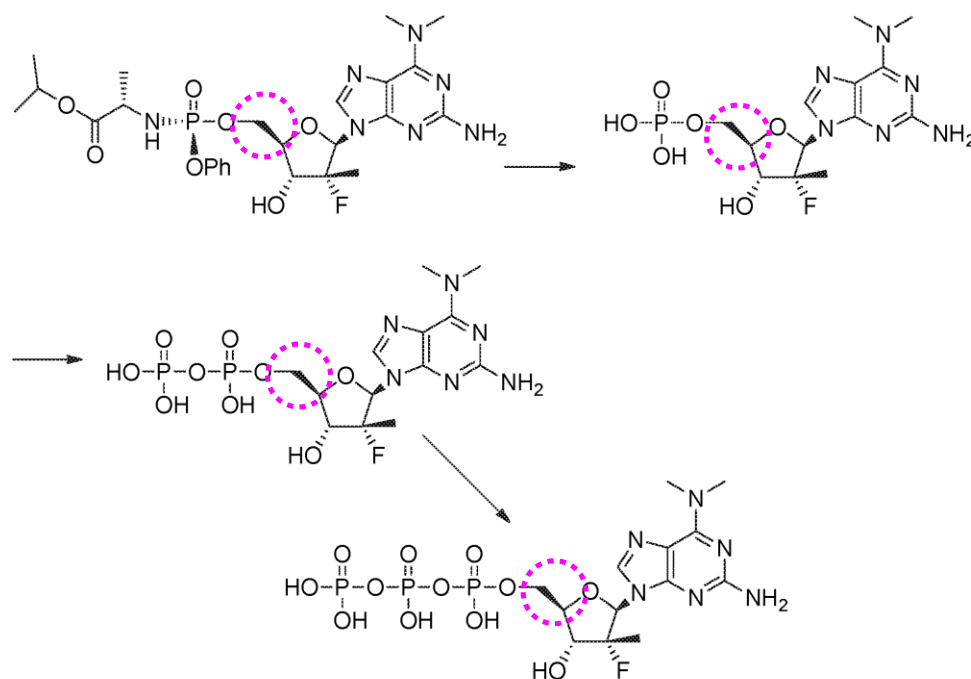


Figure: Scheme 3

53. It is further submitted that the data provided in other tables also for the mixture of isomers. Therefore, these data does not pertain to a specific compound.
54. It is further submitted that Applicant has mentioned in the impugned patent application that the invention may be useful to administer the compound to a host in combination with, for example a :
 - (1) Protease inhibitor, such as an NS3/4A protease inhibitor;
 - (2) NS5A inhibitor;
 - (3) Another NS5B polymerase inhibitor;
 - (4) NS5B non-substrate inhibitor;
 - (5) Interferon alfa-2a, which may be pegylated or otherwise modified, and/or ribavirin;
 - (6) Non-substrate-based inhibitor;
 - (7) Helicase inhibitor;
 - (8) Antisense oligodeoxynucleotide (S-ODN);
 - (9) Aptamer;
 - (10) Nuclease-resistant ribozyme;
 - (11) iRNA, including microRNA and SiRNA;
 - (12) Antibody, partial antibody or domain antibody to the virus, or
 - (13) Viral antigen or partial antigen that induces a host antibody response.
 It is submitted that the Applicant has not provided a single example in respect to the said combination therapy that provided better efficacy.
55. Furthermore, the claims of the impugned invention are drawn to compositions; however, the specification is silent about any composition. The breadth of the claims is too wide which is not supported by the disclosures. The claims do not define the constituents of the compositions.

56. The above clearly discloses that impugned patent application does not sufficiently and clearly describe the invention. Therefore, impugned patent application should be rejected.
57. The impugned patent application does not provide adequate teaching to a person skilled in the art to practice the invention. Considering above, impugned patent application does not sufficiently and clearly describe the invention. Therefore, the impugned patent application should be refused on this ground alone.

(e) GROUND 5 -Section 25(1)(h)

58. The Applicant has failed to disclose to the Controller the information required under Section 8. The applicant is required to provide all the information regarding the prosecution of the equivalent applications till the grant of the Indian application to the Controller in writing from time to time and also within the prescribed time. It is observed that applicant has not updated the status of corresponding application in the Form-3 which information has not been provided to the learned Controller.
59. Therefore, the applicant has failed to comply with the requirements of the section 8 of the act and the Opponent demands rejection on this ground also. It is submitted that the Applicant has failed to disclose the details of corresponding foreign applications and impugned patent application to be refused.

CONCLUSION

60. In view of the above, the claims are not novel, inventive and not patentable and insufficient. The pre-grant opposition as filed may be allowed and the subject patent application may be refused.

HEARING REQUESTED

61. The Opponent hereby requests a hearing under section 25(1) of the Patents Act, 1970 (hereinafter referred to as “the Patents Act”) and Rule 55 of the Patents Rules (hereinafter referred to as “the Rules”).

P R A Y E R

In the fact and circumstances of the case, the Opponent prays as follows:

- i. that the Controller take the present Opposition on record;
- ii. that the Indian application 201717035303, be rejected under Section 25(1) of the Patents (Amendment) Act, 2005;
- iii. that the Opponent may be allowed to file further documents and evidence if necessary to support their averments;
- iv. that the Opponent may be allowed to file rejoinder and affidavit if necessary to support their averments;
- v. that the Opponent may be granted an opportunity of being heard in the matter before any final orders are passed;
- vi. that the Opponent may be allowed to make further submissions in case the Patentee makes any amendments in the claims;
- vii. any other reliefs considering the facts and circumstances may be granted in favour of the Opponent in the interest of justice.

Dated this 12th day of April, 2022



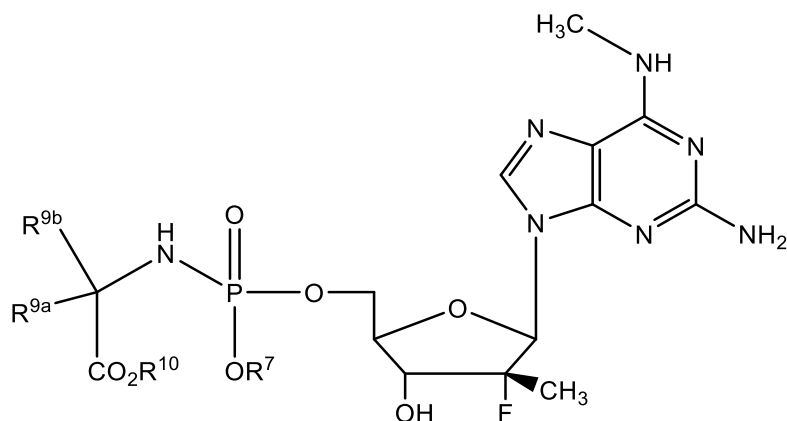
RAJESHWARI H. IN/PA - 0358
OF RAJESHWARI AND ASSOCIATES
AGENT FOR THE OPPONENT

TO,
THE CONTROLLER OF PATENTS
THE PATENT OFFICE, NEW DELHI

Annexure - 1

I/We claim:

1. A compound of the formula:



wherein:

R^7 is C_{1-6} alkyl, C_{3-7} cycloalkyl, heteroaryl, heterocyclic, or aryl;

R^{9a} and R^{9b} are

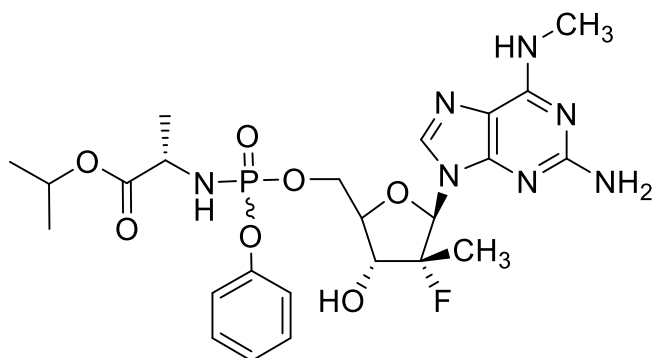
independently selected from hydrogen and C_{1-6} alkyl; and

R^{10} is C_{1-6} alkyl, C_{3-7} cycloalkyl, heterocycloalkyl, aryl or heteroaryl;

or a pharmaceutically acceptable salt thereof.

2. The compound of claim 1, wherein R^7 is heteroaryl, heterocyclic, or aryl.
3. The compound of claim 1, wherein:
 R^7 is phenyl, R^{9a} is hydrogen, R^{9b} is methyl, and R^{10} is isopropyl.
4. The compound of claim 1, wherein: R^7 is aryl.
5. A pharmaceutical composition comprising an effective amount of the compound of claim 1 in a pharmaceutically acceptable carrier.

6. A pharmaceutical composition comprising an effective amount of the compound of claim 4 in a pharmaceutically acceptable carrier.
7. A pharmaceutical composition comprising an effective amount of the compound of claim 3 in a pharmaceutically acceptable carrier.
8. The pharmaceutical composition of claim 5, wherein the composition is suitable for oral delivery.
9. The pharmaceutical composition of claim 6, wherein the composition is suitable for oral delivery.
10. The pharmaceutical composition of claim 7, wherein the composition is suitable for oral delivery.
11. A compound of the formula:



or a pharmaceutically acceptable salt thereof.

12. A pharmaceutical composition comprising the compound of claim 11; or a pharmaceutically acceptable salt thereof in a pharmaceutically acceptable carrier.
13. The compound of claim 1,

wherein

R^7 is aryl; and

R^{10} is C_{1-6} alkyl

and wherein the compound is in the form of an isolated phosphorus S-enantiomer and is at least 90% free of the opposite phosphorus R-enantiomer.

14. A pharmaceutical composition comprising the compound of claim 13 in a pharmaceutically acceptable carrier.

15. The compound of claim 1,

R^7 is aryl; and

R^{10} is C_{1-6} alkyl;

and wherein the compound is in the form of an isolated phosphorus R-enantiomer and is at least 90% free of the opposite phosphorus S-enantiomer.

16. A pharmaceutical composition comprising the compound of claim 15 in a pharmaceutically acceptable carrier.

17. A compound of claim 1, wherein R^7 is phenyl.

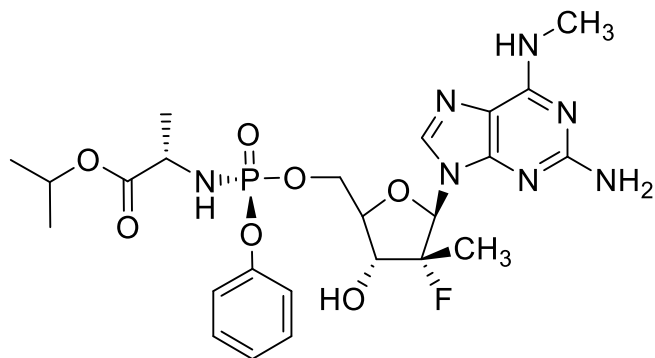
18. The compound of claim 11, wherein the compound is at least 90% free of the opposite phosphorus R-enantiomer.

19. The compound of claim 11, wherein the compound is at least 90% free of the opposite phosphorus S-enantiomer.

20. The compound of claim 18, wherein the compound is at least 98% free of the opposite phosphorus R-enantiomer.

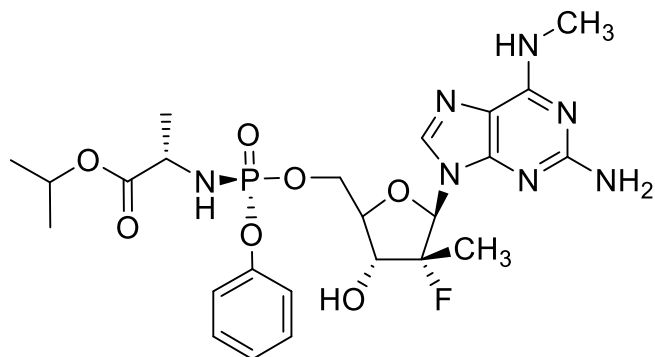
21. The compound of claim 19, wherein the compound is at least 98% free of the opposite phosphorus S-enantiomer.

22. The compound of claim 11 of the formula:



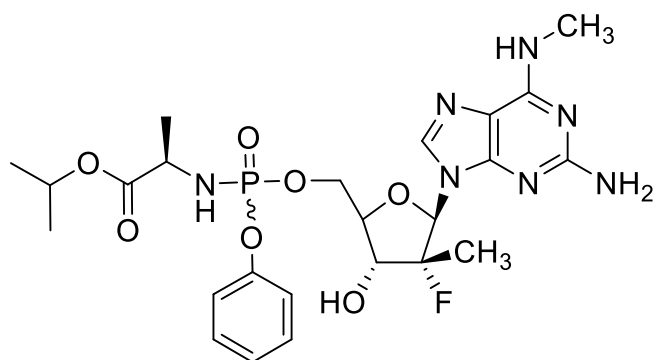
or a pharmaceutically acceptable salt thereof.

23. The compound of claim 11 of the formula:



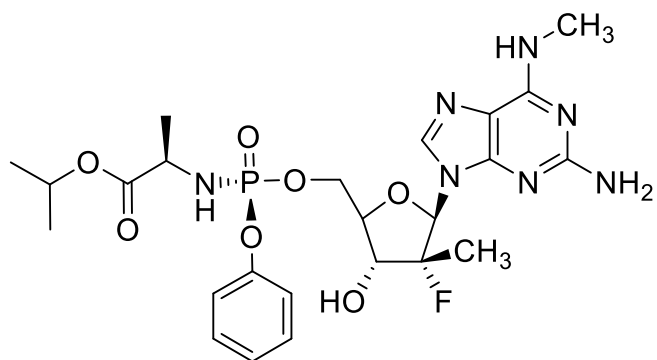
or a pharmaceutically acceptable salt thereof.

24. A compound of the formula:



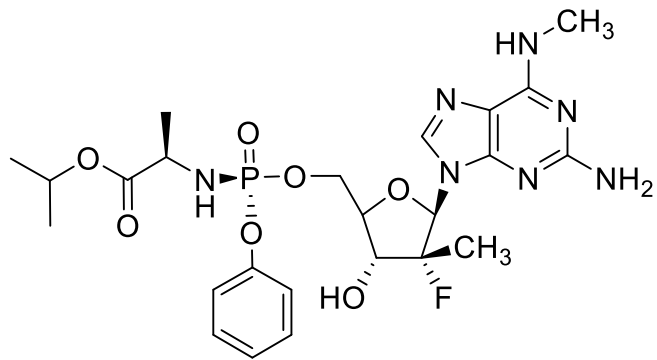
or a pharmaceutically acceptable salt thereof.

25. The compound of claim 24 of the formula:



or a pharmaceutically acceptable salt thereof.

26. The compound of claim 24 of the formula:



or a pharmaceutically acceptable salt thereof.

27. A pharmaceutical composition comprising the compound of any one of claims 22-26 in a pharmaceutically acceptable carrier.
28. A pharmaceutical composition comprising the compound of claim 22 in a pharmaceutically acceptable carrier.
29. A pharmaceutical composition comprising the compound of claim 23 in a pharmaceutically acceptable carrier.
30. The compound of claim 1, wherein R^7 is naphthyl.
31. The compound of claim 1, wherein R^{10} is C_{1-6} alkyl or C_{3-7} cycloalkyl.
32. The compound of claim 1, wherein R^{10} is C_{1-6} alkyl.
33. The compound of claim 32, wherein R^{10} is C_{1-4} alkyl.
34. The compound of claim 33, wherein R^{10} is selected from methyl, ethyl, n-propyl, and isopropyl.
35. The compound of claim 33, wherein R^{10} is iso-propyl.
36. The compound of claim 1, wherein R^{10} is aryl or heteroaryl.
37. The compound of claim 36, wherein R^{10} is phenyl.
38. The compound of claim 36, wherein R^{10} is naphthyl.

Dated 05 October 2017

MALATHI LAKSHMIKUMARAN
IN/PA-1433

Agent for the Applicant

To,
The Controller of Patents
The Patent Office at **New Delhi**

(19) **United States**(12) **Patent Application Publication**
SOFIA et al.(10) **Pub. No.: US 2010/0016251 A1**(43) **Pub. Date: Jan. 21, 2010**(54) **NUCLEOSIDE PHOSPHORAMIDATE
PRODRUGS**(75) Inventors: **MICHAEL JOSEPH SOFIA**,
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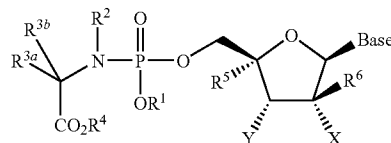
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MERCHANT & GOULD PC**P.O. BOX 2903****MINNEAPOLIS, MN 55402-0903 (US)**(73) Assignee: **PHARMASSET, INC.**,
PRINCETON, NJ (US)(21) Appl. No.: **12/053,015**(22) Filed: **Mar. 21, 2008****Related U.S. Application Data**(60) Provisional application No. 60/909,315, filed on Mar.
30, 2007, provisional application No. 60/982,309,
filed on Oct. 24, 2007.**Publication Classification**(51) **Int. Cl.****A61K 31/7076** (2006.01)**C07H 19/073** (2006.01)**A61K 31/706** (2006.01)**A61P 31/12** (2006.01)**A61K 31/702** (2006.01)**C07H 19/173** (2006.01)(52) **U.S. Cl. 514/48; 536/26.8; 536/26.7; 514/49;**
514/51

(57)

ABSTRACT

Disclosed herein are phosphoramidate prodrugs of nucleoside derivatives for the treatment of viral infections in mammals, which is a compound, its stereoisomer, salt (acid or basic addition salt), hydrate, solvate, or crystalline form thereof, represented by the following structure:



Also disclosed are methods of treatment, uses, and processes for preparing each of which utilize the compound represented by formula I.

NUCLEOSIDE PHOSPHORAMIDATE PRODRUGS

FIELD OF INVENTION

[0001] The present invention pertains to nucleoside phosphoramidates and their use as agents for treating viral diseases. These compounds are inhibitors of RNA-dependent RNA viral replication and are useful as inhibitors of HCV NS5B polymerase, as inhibitors of HCV replication and for treatment of hepatitis C infection in mammals. The invention provides novel chemical compounds, and the use of these compounds alone or in combination with other antiviral agents for treating HCV infection.

BACKGROUND

[0002] Hepatitis C virus (HCV) infection is a major health problem that leads to chronic liver disease, such as cirrhosis and hepatocellular carcinoma, in a substantial number of infected individuals, estimated to be 2-15% of the world's population. There are an estimated 4.5 million infected people in the United States alone, according to the U.S. Center for Disease Control. According to the World Health Organization, there are more than 200 million infected individuals worldwide, with at least 3 to 4 million people being infected each year. Once infected, about 20% of people clear the virus, but the rest can harbor HCV the rest of their lives. Ten to twenty percent of chronically infected individuals eventually develop liver-destroying cirrhosis or cancer. The viral disease is transmitted parenterally by contaminated blood and blood products, contaminated needles, or sexually and vertically from infected mothers or carrier mothers to their offspring. Current treatments for HCV infection, which are restricted to immunotherapy with recombinant interferon- α alone or in combination with the nucleoside analog ribavirin, are of limited clinical benefit. Moreover, there is no established vaccine for HCV. Consequently, there is an urgent need for improved therapeutic agents that effectively combat chronic HCV infection.

[0003] The HCV virion is an enveloped positive-strand RNA virus with a single oligoribonucleotide genomic sequence of about 9600 bases which encodes a polyprotein of about 3,010 amino acids. The protein products of the HCV gene consist of the structural proteins C, E1, and E2, and the non-structural proteins NS2, NS3, NS4A and NS4B, and NS5A and NS5B. The nonstructural (NS) proteins are believed to provide the catalytic machinery for viral replication. The NS3 protease releases NS5B, the RNA-dependent RNA polymerase from the polyprotein chain. HCV NS5B polymerase is required for the synthesis of a double-stranded RNA from a single-stranded viral RNA that serves as a template in the replication cycle of HCV. Therefore, NS5B polymerase is considered to be an essential component in the HCV replication complex (K. Ishi, et al. *Heptology*, 1999, 29: 1227-1235; V. Lohmann, et al., *Virology*, 1998, 249: 108-118). Inhibition of HCV NS5B polymerase prevents formation of the double-stranded HCV RNA and therefore constitutes an attractive approach to the development of HCV-specific antiviral therapies.

[0004] HCV belongs to a much larger family of viruses that share many common features.

[0005] Flaviviridae Viruses

[0006] The Flaviviridae family of viruses comprises at least three distinct genera: pestiviruses, which cause disease in

cattle and pigs; flaviviruses, which are the primary cause of diseases such as dengue fever and yellow fever; and hepaciviruses, whose sole member is HCV. The flavivirus genus includes more than 68 members separated into groups on the basis of serological relatedness (Calisher et al., *J. Gen. Virol.*, 1993, 70, 37-43). Clinical symptoms vary and include fever, encephalitis and hemorrhagic fever (*Fields Virology*, Editors: Fields, B. N., Knipe, D. M., and Howley, P. M., Lippincott-Raven Publishers, Philadelphia, Pa., 1996, Chapter 31, 931-959). Flaviviruses of global concern that are associated with human disease include the Dengue Hemorrhagic Fever viruses (DHF), yellow fever virus, shock syndrome and Japanese encephalitis virus (Halstead, S. B., *Rev. Infect. Dis.*, 1984, 6, 251-264; Halstead, S. B., *Science*, 239:476-481, 1988; Monath, T. P., *New Eng. J. Med.*, 1988, 319, 641-643).

[0007] The pestivirus genus includes bovine viral diarrhea virus (BVDV), classical swine fever virus (CSFV, also called hog cholera virus) and border disease virus (BDV) of sheep (Moennig, V. et al. *Adv. Vir. Res.* 1992, 41, 53-98). Pestivirus infections of domesticated livestock (cattle, pigs and sheep) cause significant economic losses worldwide. BVDV causes mucosal disease in cattle and is of significant economic importance to the livestock industry (Meyers, G. and Thiel, H. J., *Advances in Virus Research*, 1996, 47, 53-118; Moennig V., et al. *Adv. Vir. Res.* 1992, 41, 53-98). Human pestiviruses have not been as extensively characterized as the animal pestiviruses. However, serological surveys indicate considerable pestivirus exposure in humans.

[0008] Pestiviruses and hepaciviruses are closely related virus groups within the Flaviviridae family. Other closely related viruses in this family include the GB virus A, GB virus A-like agents, GB virus-B and GB virus-C (also called hepatitis G virus, HGV). The hepacivirus group (hepatitis C virus; HCV) consists of a number of closely related but genotypically distinguishable viruses that infect humans. There are at least 6 HCV genotypes and more than 50 subtypes. Due to the similarities between pestiviruses and hepaciviruses, combined with the poor ability of hepaciviruses to grow efficiently in cell culture, bovine viral diarrhea virus (BVDV) is often used as a surrogate to study the HCV virus.

[0009] The genetic organization of pestiviruses and hepaciviruses is very similar. These positive stranded RNA viruses possess a single large open reading frame (ORF) encoding all the viral proteins necessary for virus replication. These proteins are expressed as a polyprotein that is co- and post-translationally processed by both cellular and virus-encoded proteinases to yield the mature viral proteins. The viral proteins responsible for the replication of the viral genome RNA are located within approximately the carboxy-terminal. Two-thirds of the ORF are termed nonstructural (NS) proteins. The genetic organization and polyprotein processing of the non-structural protein portion of the ORF for pestiviruses and hepaciviruses is very similar. For both the pestiviruses and hepaciviruses, the mature nonstructural (NS) proteins, in sequential order from the amino-terminus of the nonstructural protein coding region to the carboxy-terminus of the ORF, consist of p7, NS2, NS3, NS4A, NS4B, NS5A, and NS5B.

[0010] The NS proteins of pestiviruses and hepaciviruses share sequence domains that are characteristic of specific protein functions. For example, the NS3 proteins of viruses in both groups possess amino acid sequence motifs characteristic of serine proteinases and of helicases (Gorbalenya et al., *Nature*, 1988, 333, 22; Bazan and Fletterick *Virology*, 1989,

171,637-639; Gorbalenya et al., *Nucleic Acid Res.*, 1989, 17, 3889-3897). Similarly, the NS5B proteins of pestiviruses and hepaciviruses have the motifs characteristic of RNA-directed RNA polymerases (Koonin, E. V. and Dolja, V. V., *Crit. Rev. Biochem. Molec. Biol.* 1993, 28, 375-430).

[0011] The actual roles and functions of the NS proteins of pestiviruses and hepaciviruses in the lifecycle of the viruses are directly analogous. In both cases, the NS3 serine proteinase is responsible for all proteolytic processing of polyprotein precursors downstream of its position in the ORF (Wiskerchen and Collett, *Virology*, 1991, 184, 341-350; Bartenschlager et al., *J. Virol.* 1993, 67, 3835-3844; Eckart et al. *Biochem. Biophys. Res. Comm.* 1993, 192, 399406; Grakoui et al., *J. Virol.* 1993, 67, 2832-2843; Grakoui et al., *Proc. Natl. Acad. Sci. USA* 1993, 90, 10583-10587; Hijikata et al., *J. Virol.* 1993, 67, 4665-4675; Tome et al., *J. Virol.*, 1993, 67, 4017-4026). The NS4A protein, in both cases, acts as a cofactor with the NS3 serine protease (Bartenschlager et al., *J. Virol.* 1994, 68, 5045-5055; Failla et al., *J. Virol.* 1994, 68, 3753-3760; Xu et al., *J. Virol.*, 1997, 71:53 12-5322). The NS3 protein of both viruses also functions as a helicase (Kim et al., *Biochem. Biophys. Res Comm.*, 1995, 215, 160-166; Jin and Peterson, *Arch. Biochem. Biophys.*, 1995, 323, 47-53; Warrenner and Collett, *J. Virol.* 1995, 69, 1720-1726). Finally, the NS5B proteins of pestiviruses and hepaciviruses have the predicted RNA-directed RNA polymerases activity (Behrens et al., *EMBO*, 1996, 15, 12-22; Lechmann et al., *J. Virol.*, 1997, 71, 8416-8428; Yuan et al., *Biochem. Biophys. Res. Comm.* 1997, 232, 231-235; Hagedorn, PCT WO 97/12033; Zhong et al, *J. Virol.*, 1998, 72, 9365-9369).

[0012] Currently, there are limited treatment options for individuals infected with hepatitis C virus. The current approved therapeutic option is the use of immunotherapy with recombinant interferon- α alone or in combination with the nucleoside analog ribavirin. This therapy is limited in its clinical effectiveness and only 50% of treated patients respond to therapy. Therefore, there is significant need for more effective and novel therapies to address the unmet medical need posed by HCV infection.

[0013] A number of potential molecular targets for drug development of direct acting antivirals as anti-HCV therapeutics have now been identified including, but not limited to, the NS2-NS3 autoprotease, the N3 protease, the N3 helicase and the NS5B polymerase. The RNA-dependent RNA polymerase is absolutely essential for replication of the single-stranded, positive sense, RNA genome and this enzyme has elicited significant interest among medicinal chemists.

[0014] Inhibitors of HCV NS5B as potential therapies for HCV infection have been reviewed: Tan, S.-L., et al., *Nature. Rev. Drug Discov.*, 2002, 1, 867-881; Walker, M. P. et al., *Exp. Opin. Investigational Drugs* 2003, 12, 1269-12807 Ni, Z.-J., et al., *Current Opinion in Drug Discovery and Development*, 2004, 7, 446-459; Beaulieu, P. L., et al., *Current Opinion in Investigational Drugs*, 2004, 5, 838-850; Wu, J., et al., *Current Drug Targets-Infectious Disorders*, 2003, 3, 207-219; Griffith, R. C., et al, *Annual Reports in Medicinal Chemistry*, 2004, 39, 223-237; Carrol, S., et al., *Infectious Disorders-Drug Targets*, 2006, 6, 17-29. The potential for the emergence of resistant HCV strains and the need to identify agents with broad genotype coverage supports the need for continuing efforts to identify novel and more effective nucleosides as HCV NS5B inhibitors.

[0015] Nucleoside inhibitors of NS5B polymerase can act either as a non-natural substrate that results in chain termina-

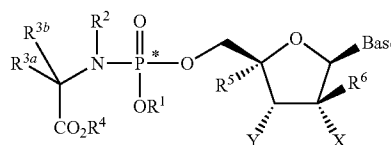
tion or as a competitive inhibitor which competes with nucleotide binding to the polymerase. To function as a chain terminator the nucleoside analog must be taken up by the cell and converted in vivo to a triphosphate to compete for the polymerase nucleotide binding site. This conversion to the triphosphate is commonly mediated by cellular kinases which imparts additional structural requirements on a potential nucleoside polymerase inhibitor. Unfortunately, this limits the direct evaluation of nucleosides as inhibitors of HCV replication to cell-based assays capable of in situ phosphorylation.

[0016] In some cases, the biological activity of a nucleoside is hampered by its poor substrate characteristics for one or more of the kinases needed to convert it to the active triphosphate form. Formation of the monophosphate by a nucleoside kinase is generally viewed as the rate limiting step of the three phosphorylation events. To circumvent the need for the initial phosphorylation step in the metabolism of a nucleoside to the active triphosphate analog, the preparation of stable phosphate prodrugs has been reported. Nucleoside phosphoramidate prodrugs have been shown to be precursors of the active nucleoside triphosphate and to inhibit viral replication when administered to viral infected whole cells (McGuigan, C., et al., *J. Med. Chem.* 1996, 39, 1748-1753; Valette, G., et al., *J. Med. Chem.*, 1996, 39, 1981-1990; Balzarini, J., et al., *Proc. National Acad Sci USA*, 1996, 93, 7295-7299; Siddiqui, A. Q., et al., *J Med. Chem.*, 1999, 42, 4122-4128; Eisenberg, E. J., et al., *Nucleosides, Nucleotides and Nucleic Acids*, 2001, 20, 1091-1098; Lee, W. A., et al, *Antimicrobial Agents and Chemotherapy*, 2005, 49, 1898); US 2006/0241064; and WO 2007/095269.

[0017] Also limiting the utility of nucleosides as viable therapeutic agents is their sometimes poor physicochemical and pharmacokinetic properties. These poor properties can limit the intestinal absorption of an agent and limit uptake into the target tissue or cell. To improve on their properties prodrugs of nucleosides have been employed. It has been demonstrated that preparation of nucleoside phosphoramidates improves the systemic absorption of a nucleoside and furthermore, the phosphoramidate moiety of these "pronucleotides" is masked with neutral lipophilic groups to obtain a suitable partition coefficient to optimize uptake and transport into the cell dramatically enhancing the intracellular concentration of the nucleoside monophosphate analog relative to administering the parent nucleoside alone. Enzyme-mediated hydrolysis of the phosphate ester moiety produces a nucleoside monophosphate wherein the rate limiting initial phosphorylation is unnecessary.

SUMMARY OF THE INVENTION

[0018] The present invention is directed toward novel phosphoramidate prodrugs of nucleoside derivatives for the treatment of viral infections in mammals, which is a compound, its stereoisomers, salts (acid or basic addition salts), hydrates, solvates, or crystalline forms thereof, represented by the following structure:



[0019] wherein

[0020] (a) R^1 is hydrogen, n-alkyl, branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-6} alkoxy, F, Cl, Br, I, nitro, cyano, C_{1-6} haloalkyl, $-N(R^1)_2$, C_{1-6} acylamino, $-NHSO_2C_{1-6}$ alkyl, $-SO_2N(R^1)_2$, COR^1 , and $-SO_2C_{1-6}$ alkyl; (R^1 is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, R^1 is $-OR^1$ or $-N(R^1)_2$);

[0021] (b) R^2 is hydrogen, C_{1-10} alkyl, R^{3a} or R^{3b} and R^2 together are $(CH_2)_n$, so as to form a cyclic ring that includes the adjoining N and C atoms, $C(O)CR^{3a}R^{3b}NHR^1$, where n is 2 to 4 and R^1 , R^{3a} , and R^{3b} ;

[0022] (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C_{1-10} alkyl, cycloalkyl, $-(CH_2)_c(NR^3)_2$, C_{1-6} hydroxyalkyl, $-CH_2SH$, $-(CH_2)_2S(O)(Me)$, $-(CH_2)_3NHC(=NH)NH_2$, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, $-(CH_2)_cCOR^3$, aryl and aryl C_{1-3} alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C_{1-10} alkyl, C_{1-6} alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C_{1-6} alkyl; (iii) R^{3a} and R^{3b} together are $(CH_2)_n$, so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R^2 together are $(CH_2)_n$, so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R^2 together are $(CH_2)_n$, so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where R^3 is independently hydrogen or C_{1-6} alkyl and R^3 is $-OR'$ or $-N(R^3)_2$; (vi) R^{3a} is H and R^{3b} is H, CH_3 , CH_2CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-C_2H_4CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl; or (viii) R^{3a} is CH_3 , $-CH_2CH_3$, $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl and R^{3b} is H, where R^3 is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, R^3 is $-OR'$ or $-N(R^3)_2$;

[0023] (d) R^4 is hydrogen, C_{1-10} alkyl, C_{1-10} alkyl optionally substituted with a lower alkyl, alkoxy, di(lower alkyl)-amino, or halogen, C_{1-10} haloalkyl, C_{3-10} cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;

[0024] (e) R^5 is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., $-(CH_2)_pOH$, where p is 1-6, including hydroxyl methyl (CH_2OH), CH_2F , N_3 , CH_2CN , CH_2NH_2 , CH_2NHCH_3 , $CH_2N(CH_3)_2$, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R^6 is H, R^5 cannot be N_3 and when X is OH, R^6 is CH_3 or CH_2F and B is a purine base, R^5 cannot be H;

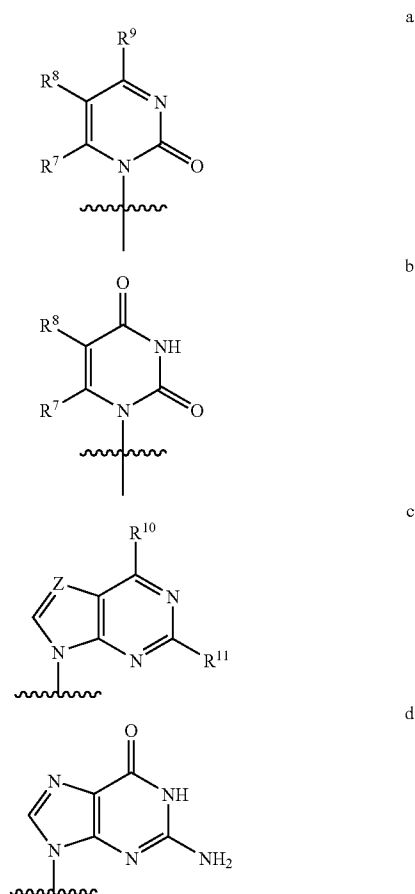
[0025] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

[0026] (g) X is H, OH, F, OMe, halogen, NH_2 , or N_3 ;

[0027] (h) Y is OH, H, C_{1-4} alkyl, C_{2-4} alkenyl, C_{2-4} alkynyl, vinyl, N_3 , CN, Cl, Br, F, I, NO_2 , $OC(O)O(CO_{1-4}alkyl)$, $OC(O)O(C_{1-4}alkyl)$, $OC(O)O(C_{2-4}alkynyl)$, $OC(O)O(C_{2-4}$

alkenyl), OC_{1-10} haloalkyl, O(aminoacyl), $O(C_{1-10}alkyl)$, $O(C_{1-4}alkyl)$, $O(C_{2-4}alkenyl)$, $S(C_{1-4}alkyl)$, $S(C_{1-4}alkyl)$, $S(C_{2-4}alkynyl)$, $S(C_{2-4}alkenyl)$, $SO(C_{1-4}alkyl)$, $SO(C_{1-4}alkyl)$, $SO(C_{2-4}alkynyl)$, $SO(C_{2-4}alkenyl)$, $SO_2(C_{1-4}alkyl)$, $SO_2(C_{2-4}alkynyl)$, $SO_2(C_{2-4}alkenyl)$, $OS(O)_2(C_{1-4}alkyl)$, $OS(O)_2(C_{1-4}alkyl)$, $OS(O)_2(C_{2-4}alkenyl)$, NH_2 , $NH(C_{1-4}alkyl)$, $NH(C_{2-4}alkenyl)$, $NH(C_{2-4}alkynyl)$, $NH(C_{1-4}alkyl)$, $N(C_{1-4}alkyl)_2$, $N(C_{1-18}alkyl)_2$, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N_3 , CN, one to three halogen (Cl, Br, F, I), NO_2 , $C(O)O(C_{1-4}alkyl)$, $C(O)O(C_{1-4}alkyl)$, $C(O)O(C_{2-4}alkynyl)$, $C(O)O(C_{2-4}alkenyl)$, $O(C_{1-4}alkyl)$, $O(C_{1-4}alkyl)$, $O(C_{2-4}alkenyl)$, $S(C_{1-4}alkyl)$, $S(C_{1-4}alkyl)$, $S(C_{2-4}alkynyl)$, $S(C_{2-4}alkenyl)$, $SO(C_{1-4}alkyl)$, $SO(C_{1-4}alkyl)$, $SO(C_{2-4}alkynyl)$, $SO(C_{2-4}alkenyl)$, $SO_2(C_{1-4}alkyl)$, $SO_2(C_{1-4}alkyl)$, $SO_2(C_{2-4}alkynyl)$, $SO_2(C_{2-4}alkenyl)$, $OS(O)_2(C_{1-4}alkyl)$, $OS(O)_2(C_{1-4}alkyl)$, $OS(O)_2(C_{2-4}alkenyl)$, NH_2 , $NH(C_{1-4}alkyl)$, $NH(C_{2-4}alkenyl)$, $NH(C_{2-4}alkynyl)$, $NH(C_{1-4}alkyl)$, $N(C_{1-4}alkyl)_2$, $N(C_{1-4}alkyl)_2$;

[0028] the base is a naturally occurring or modified purine or pyrimidine base represented by the following structures:



[0029] wherein

[0030] Z is N or CR^{12} ;

[0031] R^7 , R^8 , R^9 , R^{10} , and R^{11} are independently H, F, Cl, Br, I, OH, OR' , SH, SR' , NH_2 , NHR' , NR'_2 , lower alkyl of C_{1-6} halogenated (F, Cl, Br, I) lower alkyl of C_{1-6} , lower alkenyl of C_{2-6} , halogenated (F, Cl, Br, I) lower alkenyl of

C₂-C₆, lower alkynyl of C₂-C₆ such as C≡CH, halogenated (F, Cl, Br, I) lower alkynyl of C₂-C₆, lower alkoxy of C₁-C₆, halogenated (F, Cl, Br, I) lower alkoxy of C₁-C₆, CO₂H, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R',

[0032] wherein R' is an optionally substituted alkyl, which includes, but is not limited to, an optionally substituted C₁₋₂₀ alkyl, an optionally substituted C₁₋₁₀ alkyl, an optionally substituted lower alkyl; an optionally substituted cycloalkyl; an optionally substituted alkynyl of C₂-C₆, an optionally substituted lower alkenyl of C₂-C₆, or optionally substituted acyl, which includes but is not limited to C(O) alkyl, C(O)(C₁₋₂₀ alkyl), C(O)(C₁₋₁₀ alkyl), or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0033] R¹² is H, halogen (including F, Cl, Br, I), OH, OR', SH, SR', NH₂, NHR', NR'₂, NO₂ lower alkyl of C₁-C₆, halogenated (F, Cl, Br, I) lower alkyl of C₁-C₆, lower alkenyl of C₂-C₆, halogenated (F, Cl, Br, I) lower alkenyl of C₂-C₆, lower alkynyl of C₂-C₆, halogenated (F, Cl, Br, I) lower alkynyl of C₂-C₆, lower alkoxy of C₁-C₆, halogenated (F, Cl, Br, I) lower alkoxy of C₁-C₆, CO₂H, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'; with the proviso that when base is represented by the structure c with R¹¹ being hydrogen, R¹² is not a: (i) —C≡C—H, (ii) —C=CH₂, or (iii) —NO₂.

DEFINITIONS

[0034] The phrase “a” or “an” entity as used herein refers to one or more of that entity; for example, a compound refers to one or more compounds or at least one compound. As such, the terms “a” (or “an”), “one or more”, and “at least one” can be used interchangeably herein.

[0035] The phrase “as defined herein above” refers to the first definition provided in the Summary of the Invention.

[0036] The terms “optional” or “optionally” as used herein means that a subsequently described event or circumstance may but need not occur, and that the description includes instances where the event or circumstance occurs and instances in which it does not. For example, “optional bond” means that the bond may or may not be present, and that the description includes single, double, or triple bonds.

[0037] The term “independently” is used herein to indicate that a variable is applied in any one instance without regard to the presence or absence of a variable having that same or a different definition within the same compound. Thus, in a compound in which R appears twice and is defined as “independently carbon or nitrogen”, both R's can be carbon, both R's can be nitrogen, or one R' can be carbon and the other nitrogen.

[0038] The term “alkenyl” refers to an unsubstituted hydrocarbon chain radical having from 2 to 10 carbon atoms having one or two olefinic double bonds, preferably one olefinic double bond. The term “C_{2-N} alkenyl” refers to an alkenyl comprising 2 to N carbon atoms, where N is an integer having the following values: 3, 4, 5, 6, 7, 8, 9, or 10. The term “C₂₋₁₀ alkenyl” refers to an alkenyl comprising 2 to 10 carbon atoms. The term “C₂₋₄ alkenyl” refers to an alkenyl comprising 2 to 4 carbon atoms. Examples include, but are not limited to, vinyl, 1-propenyl, 2-propenyl(allyl) or 2-butenyl(crotyl).

[0039] The term “halogenated alkenyl” refers to an alkenyl comprising at least one of F, Cl, Br, and I.

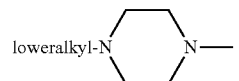
[0040] The term “alkyl” refers to an unbranched or branched chain, saturated, monovalent hydrocarbon residue containing 1 to 30 carbon atoms. The term “C_{1-M} alkyl” refers to an alkyl comprising 1 to M carbon atoms, where M is an integer having the following values, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, or 30. The term “C₁₋₄ alkyl” refers to an alkyl containing 1 to 4 carbon atoms. The term “lower alkyl” denotes a straight or branched chain hydrocarbon residue comprising 1 to 6 carbon atoms. “C₁₋₂₀ alkyl” as used herein refers to an alkyl comprising 1 to 20 carbon atoms. “C₁₋₁₀ alkyl” as used herein refers to an alkyl comprising 1 to 10 carbons. Examples of alkyl groups include, but are not limited to, lower alkyl groups include methyl, ethyl, propyl, i-propyl, n-butyl, i-butyl, t-butyl or pentyl, isopentyl, neopentyl, hexyl, heptyl, and octyl. The term (ar)alkyl or (heteroaryl)alkyl indicate the alkyl group is optionally substituted by an aryl or a heteroaryl group respectively.

[0041] The term “cycloalkyl” refers to an unsubstituted or substituted carbocycle, in which the carbocycle contains 3 to 10 carbon atoms; preferably 3 to 8 carbon atoms; more preferably 3 to 6 carbon atoms (i.e., lower cycloalkyls). Examples of cycloalkyl groups include, but are not limited to, cyclopropyl, 2-methyl-cyclopropyl, cyclobutyl, cyclopentyl, and cyclohexyl.

[0042] The term “cycloalkyl alkyl” refers to an additionally unsubstituted or substituted alkyl substituted by a lower cycloalkyl. Examples of cycloalkyl alkyls include, but are not limited to, any one of methyl, ethyl, propyl, i-propyl, n-butyl, i-butyl, t-butyl or pentyl, isopentyl, neopentyl, hexyl, heptyl, and octyl that is substituted with cyclopropyl, 2-methyl-cyclopropyl, cyclobutyl, cyclopentyl, and cyclohexyl.

[0043] The term “cycloheteroalkyl” refers to an unsubstituted or substituted heterocycle, in which the heterocycle contains 2 to 9 carbon atoms; preferably 2 to 7 carbon atoms; more preferably 2 to 5 carbon atoms. Examples of cycloheteroalkyls include, but are not limited to, aziridin-2-yl, N—C₁₋₃-alkyl-aziridin-2-yl, azetidiny, N—C₁₋₃-alkyl-azetidin-m'-yl, pyrrolidin-m'-yl, N—C₁₋₃-alkyl-pyrrolidin-m'-yl, piperidin-m'-yl, and N—C₁₋₃-alkyl-piperidin-m'-yl, where m' is 2, 3, or 4 depending on the cycloheteroalkyl. Specific examples of N—C₁₋₃-alkyl-cycloheteroalkyls include, but are not limited to, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-2-yl, N-methyl-piperidin-3-yl, and N-methyl-piperidin-4-yl. In the instance of R⁴, the point of attachment between the cycloheteroalkyl ring carbon and the oxygen occurs at any one of m'

[0044] The term “heterocycle” refers to an unsubstituted or substituted heterocycle containing carbon, hydrogen, and at least one of N, O, and S, where the C and N can be trivalent or tetravalent, i.e., sp²- or sp³-hybridized. Examples of heterocycles include, but are not limited to, aziridine, azetidine, pyrrolidine, piperidine, imidazole, oxazole, piperazine, etc. In the instance of piperazine, as related to R¹⁰ for NR'₂, the corresponding opposite nitrogen atom of the piperazinyl is substituted by a lower alkyl represented by the following structure:



[0045] Preferably, the opposite nitrogen of the piperazinyl is substituted by a methyl group.

[0046] The term “halogenated alkyl” (or “haloalkyl”) refers to an unbranched or branched chain alkyl comprising at least one of F, Cl, Br, and I. The term “C_{1-M} haloalkyl” refers to an alkyl comprising 1 to M carbon atoms that comprises at least one of F, Cl, Br, and I, where M is an integer having the following values: 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, or 30. “C₁₋₃ haloalkyl” refers to a haloalkyl comprising 1 to 3 carbons and at least one of F, Cl, Br, and I. The term “halogenated lower alkyl” (or “lower haloalkyl”) refers to a haloalkyl comprising 1 to 6 carbon atoms and at least one of F, Cl, Br, and I. Examples include, but are not limited to, fluoromethyl, chloromethyl, bromomethyl, iodomethyl, difluoromethyl, dichloromethyl, dibromomethyl, diiodomethyl, trifluoromethyl, trichloromethyl, tribromomethyl, triiodomethyl, 1-fluoroethyl, 1-chloroethyl, 1-bromoethyl, 1-iodoethyl, 2-fluoroethyl, 2-chloroethyl, 2-bromoethyl, 2-iodoethyl, 2,2-difluoroethyl, 2,2-dichloroethyl, 2,2-dibromomethyl, 2,2-diiodomethyl, 3-fluoropropyl, 3-chloropropyl, 3-bromopropyl, 2,2,2-trifluoroethyl or 1,1,2,2,2-pentafluoroethyl.

[0047] The term “alkynyl” refers to an unbranched or branched hydrocarbon chain radical having from 2 to 10 carbon atoms, preferably 2 to 5 carbon atoms, and having one triple bond. The term “C_{2-N} alkynyl” refers to an alkynyl comprising 2 to N carbon atoms, where N is an integer having the following values: 3, 4, 5, 6, 7, 8, 9, or 10. The term “C₂₋₄ alkynyl” refers to an alkynyl comprising 2 to 4 carbon atoms. The term “C₂₋₁₀ alkynyl” refers to an alkynyl comprising 2 to 10 carbons. Examples include, but are limited to, ethynyl, 1-propynyl, 2-propynyl, 1-butylnyl, 2-butylnyl or 3-butylnyl.

[0048] The term “halogenated alkynyl” refers to an unbranched or branched hydrocarbon chain radical having from 2 to 10 carbon atoms, preferably 2 to 5 carbon atoms, and having one triple bond and at least one of F, Cl, Br, and I.

[0049] The term “cycloalkyl” refers to a saturated carbocyclic ring comprising 3 to 8 carbon atoms, i.e. cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl or cyclooctyl. The term “C₃₋₇ cycloalkyl” as used herein refers to a cycloalkyl comprising 3 to 7 carbons in the carbocyclic ring.

[0050] The term “alkoxy” refers to an —O-alkyl group or an —O-cycloalkyl group, wherein alkyl and cycloalkyl are as defined above. Examples of —O-alkyl groups include, but are not limited to, methoxy, ethoxy, n-propyloxy, i-propyloxy, n-butyloxy, i-butyloxy, t-butyloxy. “Lower alkoxy” as used herein denotes an alkoxy group with a “lower alkyl” group as previously defined. “C₁₋₁₀ alkoxy” refers to an —O-alkyl wherein alkyl is C₁₋₁₀. Examples of —O-cycloalkyl groups include, but are not limited to, —O-c-propyl, —O-c-butyl, —O-c-pentyl, and —O-c-hexyl.

[0051] The term “halogenated alkoxy” refers to an —O-alkyl group in which the alkyl group comprises at least one of F, Cl, Br, and I.

[0052] The term “halogenated lower alkoxy” refers to an —O-(lower alkyl) group in which the lower alkyl group comprises at least one of F, Cl, Br, and I.

[0053] The term “amino acid” includes naturally occurring and synthetic α , β γ or δ amino acids, and includes but is not limited to, amino acids found in proteins, i.e. glycine, alanine, valine, leucine, isoleucine, methionine, phenylalanine, tryptophan, proline, serine, threonine, cysteine, tyrosine, asparagine, glutamine, aspartate, glutamate, lysine, arginine and histidine. In a preferred embodiment, the amino acid is in the L-configuration. Alternatively, the amino acid can be a derivative of alanyl, valinyl, leucinyl, isoleucinyl, prolinyl

phenylalaninyl, tryptophanyl, methioninyl, glyciny, serinyl, threoninyl, cysteinyl, tyrosinyl, asparaginyl, glutaminyl, aspartoyl, glutaroyl, lysinyl, argininyl, histidinyl, β -alanyl, β -valinyl, β -leucinyl, β -isoleucinyl β -prolinyl, β -phenylalaninyl, β -tryptophanyl, β -methioninyl, β -glycinyl, β -serinyl, β -threoninyl, β -cysteinyl, β -tyrosinyl, β -asparaginyl, β -glutaminyl, β -aspartoyl, β -glutaroyl, β -lysinyl, β -argininyl or β -histidinyl. When the term amino acid is used, it is considered to be a specific and independent disclosure of each of the esters of α , β γ or δ glycine, alanine, valine, leucine, isoleucine, methionine, phenylalanine, tryptophan, proline, serine, threonine, cysteine, tyrosine, asparagine, glutamine, aspartate, glutamate, lysine, arginine and histidine in the D and L-configurations

[0054] The term “aminoacyl” includes N,N-unsubstituted, N,N-monosubstituted, and N,N-disubstituted derivatives of naturally occurring and synthetic α , β γ or δ amino acyls, where the amino acyls are derived from amino acids. The amino-nitrogen can be substituted or unsubstituted. When the amino-nitrogen is substituted, the nitrogen is either mono- or di-substituted, where the substituent bound to the amino-nitrogen is a lower alkyl or an alkaryl. In the instance of its use for Y, the expression “O(aminoacyl)” is used. It is understood that the C3' carbon of the ribose is bound to the oxygen “O”, which is then bound to the carbonyl carbon of the aminoacyl.

[0055] The terms “alkylamino” or “arylamino” refer to an amino group that has one or two alkyl or aryl substituents, respectively.

[0056] The term “protected,” as used herein and unless otherwise defined, refers to a group that is added to an oxygen, nitrogen, or phosphorus atom to prevent its further reaction or for other purposes. A wide variety of oxygen and nitrogen protecting groups are known to those skilled in the art of organic synthesis. Non-limiting examples include: C(O)-alkyl, C(O)Ph, C(O)aryl, CH₃, CH₂-alkyl, CH₂-alkenyl, CH₂Ph, CH₂-aryl, CH₂O-alkyl, CH₂O-aryl, SO₂-alkyl, SO₂-aryl, tert-butyldimethylsilyl, tert-butyldiphenylsilyl, and 1,3-(1,1,3,3-tetraisopropylidisiloxanylidene).

[0057] The term “aryl,” as used herein, and unless otherwise specified, refers to substituted or unsubstituted phenyl (Ph), biphenyl, or naphthyl, preferably the term aryl refers to substituted or unsubstituted phenyl. The aryl group can be substituted with one or more moieties selected from among hydroxyl, F, Cl, Br, I, amino, alkylamino, arylamino, alkoxy, aryloxy, nitro, cyano, sulfonic acid, sulfate, phosphonic acid, phosphate, and phosphonate, either unprotected, or protected as necessary, as known to those skilled in the art, for example, as taught in T. W. Greene and P. G. M. Wuts, “Protective Groups in Organic Synthesis,” 3rd ed., John Wiley & Sons, 1999.

[0058] The terms “alkaryl” or “alkylaryl” refer to an alkyl group with an aryl substituent, such as benzyl. The terms “aralkyl” or “arylalkyl” refer to an aryl group with an alkyl substituent.

[0059] The term “di(lower alkyl)amino-lower alkyl” refers to a lower alkyl substituted by an amino group that is itself substituted by two lower alkyl groups. Examples include, but are not limited to, (CH₃)₂NCH₂, (CH₃)₂NCH₂CH₂, (CH₃)₂NCH₂CH₂CH₂, etc. The examples above show lower alkyls substituted at the terminus carbon atom with an N,N-dimethyl-amino substituent. These are intended as examples only and are not intended to limit the meaning of the term “di(lower alkyl)amino-lower alkyl” so as to require the same. It is contemplated that the lower alkyl chain can be substituted

with an N,N-di(lower alkyl)-amino at any point along the chain, e.g., $\text{CH}_3\text{CH}(\text{N}(\text{lower alkyl})_2)\text{CH}_2\text{CH}_2$.

[0060] The term “halo,” as used herein, includes chloro, bromo, iodo and fluoro.

[0061] The term “acyl” refers to a substituent containing a carbonyl moiety and a non-carbonyl moiety. The carbonyl moiety contains a double-bond between the carbonyl carbon and a heteroatom, where the heteroatom is selected from among O, N and S. When the heteroatom is N, the N is substituted by a lower alkyl. The non-carbonyl moiety is selected from straight, branched, and cyclic alkyl, which includes, but is not limited to, a straight, branched, or cyclic C_{1-20} alkyl, C_{1-10} alkyl, or lower alkyl; alkoxyalkyl, including methoxymethyl; aralkyl, including benzyl; aryloxyalkyl, such as phenoxymethyl; or aryl, including phenyl optionally substituted with halogen (F, Cl, Br, I), hydroxyl, C_1 to C_4 alkyl, or C_1 to C_4 alkoxy, sulfonate esters, such as alkyl or aralkyl sulphonyl, including methanesulfonyl, the mono, di or triphosphate ester, trityl or monomethoxytrityl, substituted benzyl, trialkylsilyl (e.g. dimethyl-t-butylsilyl) or diphenyl-methylsilyl. When at least one aryl group is present in the non-carbonyl moiety, it is preferred that the aryl group comprises a phenyl group.

[0062] The term “lower acyl” refers to an acyl group in which the non-carbonyl moiety is lower alkyl.

[0063] The term “purine” or “pyrimidine” base includes, but is not limited to, adenine, N^6 -alkylpurines, N^6 -acylpurines (wherein acyl is $\text{C}(\text{O})(\text{alkyl, aryl, alkylaryl, or arylalkyl})$), N^6 -benzylpurine, N^6 -halopurine, N^6 -vinylpurine, N^6 -acetylenic purine, N^6 -acyl purine, N^6 -hydroxyalkyl purine, N^6 -allylaminopurine, N^6 -thioalkyl purine, N^2 -alkylpurines, N^2 -alkyl-6-thiopurines, thymine, cytosine, 5-fluorocytosine, 5-methylcytosine, 6-azapyrimidine, including 6-azacytosine, 2- and/or 4-mercaptopyrimidine, uracil, 5-halouracil, including 5-fluorouracil, C^5 -alkylpyrimidines, C^5 -benzylpyrimidines, C^5 -halopyrimidines, C^5 -vinylpyrimidine, C^5 -acetylenic pyrimidine, C^5 -acyl pyrimidine, C^5 -hydroxyalkyl purine, C^5 -amidopyrimidine, C^5 -cyanopyrimidine, C^5 -iodopyrimidine, C^6 -iodo-pyrimidine, C^5 -Br-vinyl pyrimidine, C^6 -Br-vinyl pyrimidine, C^5 -nitropyrimidine, C^5 -amino-pyrimidine, N^2 -alkylpurines, N^2 -alkyl-6-thiopurines, 5-azacytidinyl, 5-azauracilyl, triazolopyridinyl, imidazolopyridinyl, pyrrolopyrimidinyl, and pyrazolopyrimidinyl. Purine bases include, but are not limited to, guanine, adenine, hypoxanthine, 2,6-diaminopurine, and 6-chloropurine. Functional oxygen and nitrogen groups on the base can be protected as necessary or desired. Suitable protecting groups are well known to those skilled in the art, and include trimethylsilyl, dimethylhexylsilyl, t-butyl dimethylsilyl, and 1-butyl-diphenylsilyl, trityl, alkyl groups, and acyl groups such as acetyl and propionyl, methanesulfonyl, and p-toluenesulfonyl.

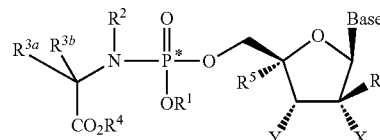
[0064] The term “tautomerism” and “tautomers” have their accepted plain meanings.

[0065] The term “P*” means that the phosphorous atom is chiral and that it has a corresponding Cahn-Ingold-Prelog designation of “R” or “S” which have their accepted plain meanings. It is contemplated that compounds of the formula I are racemic because the chirality at phosphorous. Applicants contemplate use of the racemate and/or the resolved enantiomers. In some instances, an asterisk does not appear next to the phosphoroamidate phosphorous atom. In these instances, it is understood that the phosphorous atom is chiral and that one of ordinary skill understands this to be so unless the

substituents bound to the phosphorous exclude the possibility of chirality at phosphorous, such as in $\text{P}(\text{O})\text{Cl}_3$.

DETAILED DESCRIPTION OF THE INVENTION

[0066] An aspect of the invention is directed to a compound, its salts, hydrates, solvates, crystalline forms, and the like represented by formula I:



[0067] wherein

[0068] (a) R^1 is hydrogen, n-alkyl; branched alkyl, cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-6} alkoxy, F, Cl, Br, I, nitro, cyano, C_{1-6} haloalkyl, $-\text{N}(\text{R}^1)_2$, C_{1-6} acylamino, $-\text{NHSO}_2\text{C}_{1-6}$ alkyl, $-\text{SO}_2\text{N}(\text{R}^1)_2$, COR^1 , and $-\text{SO}_2\text{C}_{1-6}$ alkyl; (R^1 is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, R^1 is $-\text{OR}^1$ or $-\text{N}(\text{R}^1)_2$);

[0069] (b) R^2 is hydrogen, C_{1-10} alkyl, R^{3a} or R^{3b} and R^2 together are $(\text{CH}_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms, $\text{C}(\text{O})\text{CR}^{3a}\text{R}^{3b}\text{NHR}^1$, where n is 2 to 4 and R^1 , R^{3a} , and R^{3b} ;

[0070] (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C_{1-10} alkyl, cycloalkyl, $-(\text{CH}_2)_c(\text{NR}^{3'})_2$, C_{1-6} hydroxyalkyl, $-\text{CH}_2\text{SH}$, $-(\text{CH}_2)_2\text{S}(\text{O})_d\text{Me}$, $-(\text{CH}_2)_3\text{NHC}(=\text{NH})\text{NH}_2$, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, $-(\text{CH}_2)_e\text{COR}^{3''}$, aryl and aryl C_{1-3} alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C_{1-10} alkyl, C_{1-6} alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C_{1-6} alkyl; (iii) R^{3a} and R^{3b} together are $(\text{CH}_2)_n$ so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R^2 together are $(\text{CH}_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R^2 together are $(\text{CH}_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where $\text{R}^{3'}$ is independently hydrogen or C_{1-6} alkyl and $\text{R}^{3''}$ is $-\text{OR}^1$ or $-\text{N}(\text{R}^{3'})_2$; (vi) R^{3a} is H and R^{3b} is H, CH_3 , CH_2CH_3 , $\text{CH}(\text{CH}_3)_2$, $\text{CH}_2\text{CH}(\text{CH}_3)_2$, $\text{CH}(\text{CH}_3)\text{CH}_2\text{CH}_3$, CH_2Ph , CH_2 -indol-3-yl, $-\text{CH}_2\text{CH}_2\text{SCH}_3$, $\text{CH}_2\text{CO}_2\text{H}$, $\text{CH}_2\text{C}(\text{O})\text{NH}_2$, $\text{CH}_2\text{CH}_2\text{COOH}$, $\text{CH}_2\text{CH}_2\text{C}(\text{O})\text{NH}_2$, $\text{CH}_2\text{CH}_2\text{CH}_2\text{CH}_2\text{NH}_2$, $-\text{CH}_2\text{CH}_2\text{CH}_2\text{NHC}(\text{NH})\text{NH}_2$, C_2 -imidazol-4-yl, CH_2OH , $\text{CH}(\text{OH})\text{CH}_3$, $\text{CH}_2((4'\text{-OH})\text{-Ph})$, CH_3SH , or lower cycloalkyl; or (viii) R^{3a} is CH_3 , $-\text{CH}_2\text{CH}_3$, $\text{CH}(\text{CH}_3)_2$, $\text{CH}_2\text{CH}(\text{CH}_3)_2$, $\text{CH}(\text{CH}_3)\text{CH}_2\text{CH}_3$, CH_2Ph , CH_2 -indol-3-yl, $-\text{CH}_2\text{CH}_2\text{SCH}_3$, $\text{CH}_2\text{CO}_2\text{H}$, $\text{CH}_2\text{C}(\text{O})\text{NH}_2$, $\text{CH}_2\text{CH}_2\text{COOH}$, $\text{CH}_2\text{CH}_2\text{C}(\text{O})\text{NH}_2$, $\text{CH}_2\text{CH}_2\text{CH}_2\text{CH}_2\text{NH}_2$, $-\text{CH}_2\text{CH}_2\text{CH}_2\text{NHC}(\text{NH})\text{NH}_2$, CH_2 -imidazol-4-yl, CH_2OH , $\text{CH}(\text{OH})\text{CH}_3$, $\text{CH}_2((4'\text{-OH})\text{-Ph})$, CH_3SH , or lower cycloalkyl and R^{3b} is H, where $\text{R}^{3'}$ is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, $\text{R}^{3''}$ is $-\text{OR}^1$ or $-\text{N}(\text{R}^{3'})_2$;

[0071] (d) R^4 is hydrogen, C_{1-10} alkyl, C_{1-10} alkyl optionally substituted with a lower alkyl, alkoxy, di(lower alkyl)-amino, or halogen, C_{1-10} haloalkyl, C_{3-10} cycloalkyl,

cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;

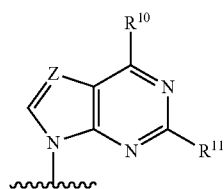
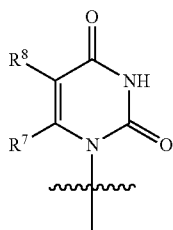
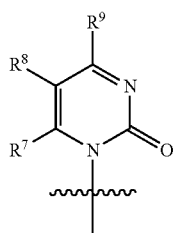
[0072] (e) R^5 is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., $-(CH_2)_pOH$, where p is 1-6, including hydroxyl methyl (CH_2OH), CH_2F , N_3 , CH_2CN , CH_2NH_2 , CH_2NHCH_3 , $CH_2N(CH_3)_2$, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R^6 is H, R^5 cannot be N_3 and when X is OH, R^6 is CH_3 or CH_2F and B is a purine base, R^5 cannot be H;

[0073] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

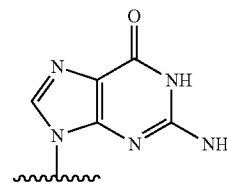
[0074] (g) X is H, OH, F, OMe, halogen, NH_2 , or N_3 ;

[0075] (h) Y is OH, H, C_{1-4} alkyl, C_{2-4} alkenyl, C_{2-4} alkynyl, vinyl, N_3 , CN, Cl, Br, F, I, NO_2 , $OC(O)O(C_{1-4}$ alkyl), $OC(O)O(C_{1-4}$ alkyl), $OC(O)O(C_{2-4}$ alkynyl), $OC(O)O(C_{2-4}$ alkenyl), OC_{1-10} haloalkyl, O(aminoacyl), $O(C_{1-10}$ acyl), $O(C_{1-4}$ alkyl), $O(C_{2-4}$ alkenyl), $S(C_{1-4}$ acyl), $S(C_{1-4}$ alkyl), $S(C_{2-4}$ alkynyl), $S(C_{2-4}$ alkenyl), $SO(C_{1-4}$ acyl), $SO(C_{1-4}$ alkyl), $SO(C_{2-4}$ alkynyl), $SO(C_{2-4}$ alkenyl), $SO_2(C_{1-4}$ acyl), $SO_2(C_{1-4}$ alkyl), $SO_2(C_{2-4}$ alkynyl), $SO_2(C_{2-4}$ alkenyl), $OS(O)_2(C_{1-4}$ acyl), $OS(O)_2(C_{1-4}$ alkyl), $OS(O)_2(C_{2-4}$ alkynyl), $OS(O)_2(C_{2-4}$ alkenyl), NH_2 , $NH(C_{1-4}$ alkyl), $NH(C_{2-4}$ alkenyl), $NH(C_{2-4}$ alkynyl), $NH(C_{1-4}$ acyl), $N(C_{1-4}$ alkyl), $N(C_{1-18}$ acyl), wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N_2 , CN, one to three halogen (Cl, Br, F, I), NO_2 , $C(O)O(C_{1-4}$ alkyl), $C(O)O(C_{1-4}$ alkyl), $C(O)O(C_{2-4}$ alkynyl), $C(O)O(C_{2-4}$ alkenyl), $O(C_{1-4}$ acyl), $O(C_{1-4}$ alkyl), $O(C_{2-4}$ alkenyl), $S(C_{1-4}$ acyl), $S(C_{1-4}$ alkyl), $S(C_{2-4}$ alkynyl), $S(C_{2-4}$ alkenyl), $SO(C_{1-4}$ acyl), $SO(C_{1-4}$ alkyl), $SO(C_{2-4}$ alkynyl), $SO(C_{2-4}$ alkenyl), $SO_2(C_{1-4}$ acyl), $SO_2(C_{1-4}$ alkyl), $SO_2(C_{2-4}$ alkynyl), $SO_2(C_{2-4}$ alkenyl), $OS(O)_2(C_{1-4}$ acyl), $OS(O)_2(C_{1-4}$ alkyl), $OS(O)_2(C_{2-4}$ alkynyl), $OS(O)_2(C_{2-4}$ alkenyl), NH_2 , $NH(C_{1-4}$ alkyl), $NH(C_{2-4}$ alkenyl), $NH(C_{2-4}$ alkynyl), $NH(C_{1-4}$ acyl), $N(C_{1-4}$ alkyl), $N(C_{1-4}$ acyl), $N(C_{1-4}$ alkyl), $N(C_{1-4}$ acyl);

[0076] the base is a naturally occurring or modified purine or pyrimidine base represented by the following structures:



-continued



[0077] wherein

[0078] Z is N or CR^{12} ;

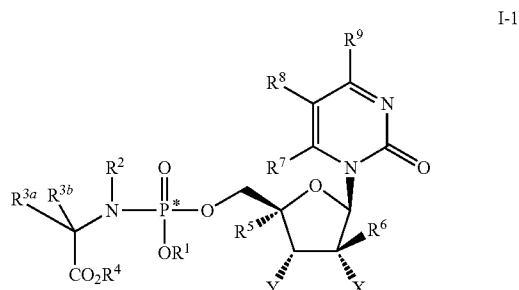
[0079] R^7 , R^8 , R^9 , R^{10} , and R^{11} are independently H, F, Cl, Br, I, OH, OR', SH, SR', NH_2 , NHR' , NR'_2 , lower alkyl of C_1 - C_6 halogenated (F, Cl, Br, I) lower alkyl of C_1 - C_6 , lower alkenyl of C_2 - C_6 , halogenated (F, Cl, Br, I) lower alkenyl of C_2 - C_6 , lower alkynyl of C_2 - C_6 such as $C\equiv CH$, halogenated (F, Cl, Br, I) lower alkynyl of C_2 - C_6 , lower alkoxy of C_1 - C_6 , halogenated (F, Cl, Br, I) lower alkoxy of C_1 - C_6 , CO_2H , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$,

[0080] wherein R' is an optionally substituted alkyl, which includes, but is not limited to, an optionally substituted C_{1-20} alkyl, an optionally substituted C_{1-10} alkyl, an optionally substituted lower alkyl; an optionally substituted cycloalkyl; an optionally substituted alkynyl of C_2 - C_6 , an optionally substituted lower alkenyl of C_2 - C_6 , or optionally substituted acyl, which includes but is not limited to $C(O)$ alkyl, $C(O)(C_{1-20}$ alkyl), $C(O)(C_{1-10}$ alkyl), or $C(O)$ (lower alkyl) or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0081] R^{12} is H, halogen (including F, Cl, Br, I), OH, OR', SH, SR', NH_2 , NHR' , NR'_2 , NO_2 lower alkyl of C_1 - C_6 , halogenated (F, Cl, Br, I) lower alkyl of C_1 - C_6 , lower alkenyl of C_2 - C_6 , halogenated (F, Cl, Br, I) lower alkenyl of C_2 - C_6 , lower alkynyl of C_2 - C_6 , halogenated (F, Cl, Br, I) lower alkynyl of C_2 - C_6 , lower alkoxy of C_1 - C_6 , halogenated (F, Cl, Br, I) lower alkoxy of C_1 - C_6 , CO_2H , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$; with the proviso that when base is represented by the structure c with R^{11} being hydrogen, R^{12} is not a: (i) $-C\equiv C-H$, (ii) $-C\equiv CH_2$, or (iii) $-NO_2$.

[0082] As can be appreciated from the structure represented by formula I above, there are myriad ways to express the several embodiments and aspects of each embodiment of the present invention. As seen below, the inventors have disclosed certain embodiments directed to the compound of formula I, each having several aspects, based on the identity of the modified purine or pyrimidine base. This is not intended to be an explicit or implicit admission that the three embodiments are independent or distinct nor should it be interpreted as such. Rather, it is intended to convey information so that the full breadth of the present invention can be understood. Furthermore, the following embodiments, and aspects thereof, are not meant to be limiting on the full breadth of the invention as recited by the structure of formula I.

[0083] A first embodiment of the invention is directed to a compound represented by formula I-1:



[0084] wherein

[0085] (a) R¹ is hydrogen, n-alkyl; branched alkyl, cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C₁₋₆ alkyl, C₂₋₆ alkenyl, C₂₋₆ alkynyl, C₁₋₆ alkoxy, F, Cl, Br, I, nitro, cyano, C₁₋₆ haloalkyl, —N(R^{1'})₂, C₁₋₆ acylamino, —NHSO₂C₁₋₆ alkyl, —SO₂N(R^{1'})₂, COR^{1''}, and —SO₂C₁₋₆ alkyl; (R^{1'} is independently hydrogen or alkyl, which includes, but is not limited to, C₁₋₂₀ alkyl, C₁₋₁₀ alkyl, or C₁₋₆ alkyl, R^{1''} is —OR' or —N(R^{1'})₂);

[0086] (b) R² is hydrogen, C₁₋₁₀ alkyl, R^{3a} or R^{3b} and R² together are (CH₂)_n, so as to form a cyclic ring that includes the adjoining N and C atoms, C(O)CR^{3a}R^{3b}NHR¹, where n is 2 to 4 and R¹, R^{3a}, and R^{3b};

[0087] (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C₁₋₁₀ alkyl, cycloalkyl, —(CH₂)_c(NR^{3'})₂, C₁₋₆ hydroxyalkyl, —CH₂SH, —(CH₂)₂S(O)_dMe, —(CH₂)₃NHC(=NH)NH₂, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, —(CH₂)_cCOR³, aryl and aryl C₁₋₃ alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C₁₋₁₀ alkyl, C₁₋₆ alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C₁₋₆ alkyl; (iii) R^{3a} and R^{3b} together are (CH₂)_n, so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R² together are (CH₂)_n, so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R² together are (CH₂)_n, so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where R^{3'} is independently hydrogen or C₁₋₆ alkyl and R^{3''} is —OR' or —N(R^{3'})₂; (vi) R^{3a} is H and R^{3b} is H, CH₃, CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl; or (viii) R^{3a} is CH₃, —CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl and R^{3b} is H, where R^{3'} is independently hydrogen or alkyl, which includes, but is not limited to, C₁₋₂₀ alkyl, C₁₋₁₀ alkyl, or C₁₋₆ alkyl, R^{3''} is —OR' or —N(R^{3'})₂);

[0088] (d) R⁴ is hydrogen, C₁₋₁₀ alkyl, C₁₋₁₀ alkyl optionally substituted with a lower alkyl, alkoxy, di(lower alkyl)-amino, or halogen, C₁₋₁₀ haloalkyl, C₃₋₁₀ cycloalkyl,

cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, di(lower alkyl)-amino, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;

[0089] (e) R⁵ is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., —(CH₂)_pOH, where p is 1-6, including hydroxyl methyl (CH₂OH), CH₂F, N₃, CH₂CN, CH₂NH₂, CH₂NHCH₃, CH₂N(CH₃)₂, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R⁶ is H, R⁵ cannot be N₃ and when X is OH, R⁶ is CH₃ or CH₂F and B is a purine base, R⁵ cannot be H;

[0090] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

[0091] (g) X is H, OH, F, OMe, Cl, Br, I, NH₂, or N₃;

[0092] (h) Y is OH, H, C₁₋₄ alkyl, C₂₋₄ alkenyl, C₂₋₄ alkynyl, vinyl, N₃, CN, Cl, Br, F, I, NO₂, OC(O)O(C₁₋₄ alkyl), OC(O)O(C₁₋₄ alkyl), OC(O)O(C₂₋₄ alkynyl), OC(O)O(C₂₋₄ alkenyl), OC₁₋₁₀ haloalkyl, O(aminoacyl), O(C₁₋₁₀ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₁₀ acyl), S(C₂₋₄ alkynyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl), SO(C₁₋₄ alkyl), SO(C₂₋₄ alkynyl), SO(C₂₋₄ alkenyl), SO₂(C₁₋₄ acyl), SO₂(C₁₋₄ alkyl), SO₂(C₂₋₄ alkynyl), SO₂(C₂₋₄ alkenyl), OS(O)₂(C₁₋₄ acyl), OS(O)₂(C₁₋₄ alkyl), OS(O)₂(C₂₋₄ alkenyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkynyl), NH(C₂₋₄ alkenyl), NH(C₁₋₄ acyl), N(C₁₋₄ alkyl)₂, N(C₁₋₁₈ acyl)₂, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N₃, CN, one to three halogen (Cl, Br, F, I), NO₂, C(O)O(C₁₋₄ alkyl), C(O)O(C₁₋₄ alkyl), C(O)O(C₂₋₄ alkynyl), C(O)O(C₂₋₄ alkenyl), O(C₁₋₄ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₄ alkyl), S(C₂₋₄ alkynyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl), SO(C₁₋₄ alkyl), SO(C₂₋₄ alkynyl), SO(C₂₋₄ alkenyl), SO₂(C₁₋₄ acyl), SO₂(C₁₋₄ alkyl), SO₂(C₂₋₄ alkynyl), SO₂(C₂₋₄ alkenyl), OS(O)₂(C₁₋₄ acyl), OS(O)₂(C₁₋₄ alkyl), OS(O)₂(C₂₋₄ alkenyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkynyl), NH(C₂₋₄ alkenyl), NH(C₁₋₄ acyl), N(C₁₋₄ alkyl)₂, N(C₁₋₄ alkyl)₂;

[0093] (i) R⁷, R⁸, R⁹ are independently H, F, Cl, Br, I, OH, OR', SH, SR', NH₂, NHR', NR'₂, lower alkyl, halogenated (F, Cl, Br, I) lower alkyl, lower alkenyl of C₂₋₆, CO₂H, CO₂R', CONH₂, CONHR', CONR'₂, wherein R' is a C₁₋₂₀ alkyl; a C₁₋₂₀ cycloalkyl; a C₂₋₆ alkenyl, a C₂₋₆ alkynyl.

[0094] A first aspect of the first embodiment is directed to a compound represented by formula I-1

[0095] wherein

[0096] (a) R¹ is hydrogen, n-alkyl or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C₁₋₆ alkyl, C₁₋₆ alkoxy, F, Cl, Br, I, nitro, cyano, C₁₋₆ haloalkyl;

[0097] (b) R² is hydrogen or CH₃;

[0098] (c) R^{3a} and R^{3b} are independently (i) R^{3a} is hydrogen and R^{3b} and R² together are (CH₂)_n, so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R² together are (CH₂)_n, so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 3 to 5, n is 2 to 4, and where R^{3'} is independently hydrogen or C₁₋₆ alkyl and R^{3''} is —OR' or —N(R^{3'})₂; (vi) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl; or (viii) R^{3a} is CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂,

CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂,
—CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl,
CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), or CH₂SH and R^{3b}
is H;

[0099] (d) R⁴ is hydrogen, C₁₋₁₀ alkyl, C₁₋₁₀ alkyl optionally substituted with a lower alkyl, alkoxy, di(lower alkyl)-amino, or halogen, C₁₋₁₀ haloalkyl, C₃₋₁₀ cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;

[0100] (e) R⁵ is H, CN, CH₃, vinyl, OCH₃, OCH₂CH₃, CH₂OH, CH₂(halo), such as CH₂F, N₃, CH₂CN, CH₂N₃, CH₂NH₂, CH₂NHCH₃, CH₂N(CH₃)₂, halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R⁶ is H, R⁵ cannot be N₃;

[0101] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

[0102] (g) X is H, OH, F, OMe, Cl, Br, I, NH₂, or N₃;

[0103] (h) Y is OH, H, C₁₋₄ alkyl, vinyl, N₃, CN, Cl, Br, F, I, O(C₁₋₆ acyl), O(C₁₋₄ alkyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkenyl), NH(C₂₋₄ alkynyl), NH(C₁₋₄ acyl), N(C₁₋₄ alkyl)₂, N(C₁₋₈ acyl)₂, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N₃, CN, one to three halogen (Cl, Br, F, I), NO₂, OC(O)O(C₁₋₄ alkyl), OC(O)O(C₁₋₄ alkyl), OC(O)O(C₂₋₄ alkynyl), OC(O)O(C₂₋₄ alkenyl), OC₁₋₄ haloalkyl, O(aminoacyl), O(C₁₋₄ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₄ alkyl), S(C₂₋₄ alkynyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl), SO(C₁₋₄ alkyl), SO(C₂₋₄ alkynyl), SO(C₁₋₄ alkenyl), SO₂(C₁₋₄ acyl), SO₂(C₁₋₄ alkyl), SO₂(C₂₋₄ alkynyl), SO₂(C₁₋₄ alkenyl), OS(O)₂(C₁₋₄ acyl), OS(O)₂(C₁₋₄ alkyl), OS(O)₂(C₂₋₄ alkynyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkenyl), NH(C₂₋₄ alkynyl), NH(C₁₋₄ acyl), N(C₁₋₄ alkyl)₂, or N(C₁₋₄ acyl)₂;

[0104] (i) R⁷, R⁸R⁹ are independently H, F, Cl, Br, I, OH, OR', SH, SR', NH₂, NHR', NR'₂, lower alkyl, halogenated (F, Cl, Br, I) lower alkyl, lower alkenyl of C₂-C₆, CO₂H, CO₂R', CONH₂, CONHR', CONR'₂, wherein R' is a C₁₋₂₀ alkyl; a C₁₋₂₀ cycloalkyl; a C₂-C₆ alkenyl, a C₂-C₆ alkynyl.

[0105] A second aspect of the first embodiment is directed to a compound represented by formula I-1

[0106] wherein

[0107] (a) R¹ is hydrogen, n-alkyl or a substituted or unsubstituted phenyl, where the substituent of the substituted phenyl is at least one of a C₁₋₃ alkyl, a C₁₋₃ alkoxy, F, Cl, Br, I, nitro, cyano, and a C₁₋₃ haloalkyl;

[0108] (b) R² is hydrogen or CH₃;

[0109] (c) R^{3a} and R^{3b} are independently (i) R^{3a} is hydrogen and R^{3b} and R² together are (CH₂)_n so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R² together are (CH₂)_n so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 3 to 5 n is 2 to 4, and where R³ is independently hydrogen or C₁₋₆ alkyl and R^{3'} is —OR' or —N(R³)₂; (vi) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl; or (viii) R^{3a} is CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂,

—CF₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)H₃, CH₂((4'-OH)-Ph), or CH₂SH and R^{3b} is H;

[0110] (d) R⁴ is hydrogen, CH₃, Et, 'Pr, "Pr, "Bu, 2-butyl, 'Bu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, di(lower alkyl)amino-lower alkyl, or aminoacyl;

[0111] (e) R⁵ is H, CN, CH₃, OCH₃, CH₂OH, CH₂(halo), such as CH₂F, N₃, CH₂CN, CH₂N₃, CH₂NH₂, CH₂NHCH₃, CH₂N(CH₃)₂, halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R⁶ is H, R⁵ cannot be N₃;

[0112] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

[0113] (g) X is H, OH, F, OCH₃, NH₂, or N₃;

[0114] (h) Y is OH, H, CH₃, vinyl, N₃, CN, Cl, Br, F, I, OC(O)CH₃, OCH₃, NH₂, NHCH₃, NH(vinyl), NH(acetyl), NH(C(O)CH₃), N(CH₃)₂, N(C(O)CH₃)₂, or O(aminoacyl);

[0115] (i) R⁷ and R⁸ are independently H, F, Cl, Br, I, OH, OCH₃, SH, SCH₃, NH₂, NHCH₃, N(CH₃)₂, CH₃, CH_{3-q}X_q, where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO₂H, CO₂CH₃, CONH₂, CONHCH₃, or CON(CH₃)₂; and

[0116] (j) R⁹ is selected from among OH, OCH₃, SH, SCH₃, NH₂, NHCH₃, N(CH₃)₂, OC(O)(C₁₋₂₀ alkyl), which include but are not limited to OC(O)(CH₂)_sCH₃, NHC(O)(C₁₋₂₀ alkyl), which include but are not limited to NHC(O)(CH₂)_sCH₃, and N(C(O)(CH₂)_sCH₃)₂, which include but is not limited to N(C(O)(CH₂)_sCH₃)₂, where s is an integer selected from 0, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, and 19.

[0117] A third aspect of the first embodiment is directed to a compound represented by formula I-1

[0118] wherein

[0119] (a) R¹ is hydrogen, n-alkyl or a substituted or unsubstituted phenyl, where the substituent of the substituted phenyl is at least one of a CR₃, OCH₃, F, Cl, Br, I, nitro, cyano, and a CH_{3-q}X_q, where X is F, Cl, Br, or I, and q is 1-3;

[0120] (b) R² is hydrogen or CH₃;

[0121] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl or R^{3a} is CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl and R^{3b} is H;

[0122] (d) R⁴ is hydrogen, CH₃, Et, 'Pr, "Pr, "Bu, 2-butyl, 'Bu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, lower haloalkyl, di(lower alkyl)amino-lower alkyl, or aminoacyl;

[0123] (e) R⁵ is H, CN, CH₃, OCH₃, CH₂OH, CH₂(halo), such as CH₂F, N₃, CH₂CN, CH₂N₃, CH₂NH₂, CH₂NHCH₃, CH₂N(CH₃)₂, halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R⁶ is H, R⁵ cannot be N₃;

[0124] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

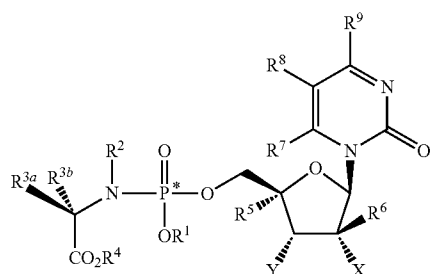
[0125] (g) X is H, OH, F, OCH₃, NH₂, or N₃;

[0126] (h) Y is OH, H, CH₃, vinyl, N₃, CN, Cl, Br, F, I, OC(O)CH₃, OCH₃, NH₂, NHCH₃, NH(vinyl), NH(acetyl), NH(C(O)CH₃), N(CH₃)₂, N(C(O)CH₃)₂, or O(aminoacyl);

[0127] (i) R⁷ and R⁸ are independently H, F, Cl, Br, I, OH, OCH₃, SH, SCH₃, NH₂, NHCH₃, N(CH₃)₂, CH₃, CH_{3-q}X_q, where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO₂H, CO₂CH₃, CONH₂, CONHCH₃, or CON(CH₃)₂, wherein R¹ is a C₁₋₂₀ alkyl; a C₁₋₂₀ cycloalkyl; a C₂-C₆ alkenyl, a C₂-C₆ alkynyl; and

[0128] (j) R⁹ is selected from among OH, OCH₃, SH, SCH₃, NH₂, NHCH₃, N(CH₃)₂, OC(O)(C₁₋₂₀ alkyl), which include but are not limited to OC(O)(CH₂)_sCH₃, NHC(O)(C₁₋₂₀ alkyl), which include but are not limited to NHC(O)(CH₂)_sCH₃, and N(C(O)(CH₂)_sCH₃)₂, which include but is not limited to N(C(O)(CH₂)_sCH₃)₂, where s is an integer selected from 0, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, and 19.

[0129] A fourth aspect of the first embodiment is directed to a compound represented by formula I-2



I-2

[0130] wherein

[0131] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, or a substituted or unsubstituted phenyl, where the substituent of the substituted phenyl is at least one of a CH₃, OCH₃, F, Cl, Br, I, nitro, cyano, and a CH_{3-q}X_q, where X is F, Cl, Br, or I, and q is 1-3;

[0132] (b) R² is hydrogen or CH₃;

[0133] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl or R^{3a} is CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH((4'-OH)-Ph), CH₂SH, or lower cycloalkyl and R^{3b} is H;

[0134] (d) R⁴ is hydrogen, C₁₋₁₀ alkyl, C₁₋₁₀ alkyl optionally substituted with a lower alkyl, alkoxy or halogen, C₁₋₁₀ haloalkyl, C₃₋₁₀ cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, di(lower alkyl)amino-lower alkyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;

[0135] (e) R⁵ is H, CN, CH₃, OCH₃, CH₂OH, CH₂F, N₃, CH₂CN, CH₂N₃, CH₂NH₂, CH₂NHCH₃, CH₂N(CH₃)₂, halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R⁶ is H, R⁵ cannot be N₃;

[0136] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

[0137] (g) X is H, OH, F, OCH₃, Cl, Br, I, NH₂, or N₃;

[0138] (h) Y is OH, H, CH₃, vinyl, N₃, CN, Cl, Br, F, I, OC(O)CH₃, OCH₃, NH₂, NHCH₃, NH(vinyl), NH(acetyl), NH(C(O)CH₃), N(CH₃)₂, N(C(O)CH₃)₂, or O(aminoacyl);

[0139] (i) R⁷ and R⁸ are independently H, F, Cl, Br, I, OH, OCH₃, SH, SCH₃, NH₂, NHCH₃, N(CH₃)₂, CH₃, CH_{3-q}X_q, where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO₂H, CO₂CH₃, CONH₂, CONHCH₃, or CON(CH₃)₂, wherein R¹ is a C₁₋₂₀ alkyl; a C₁₋₂₀ cycloalkyl; a C₂-C₆ alkenyl, a C₂-C₆ alkynyl; and

[0140] (j) R⁹ is selected from among OH, OCH₃, SH, SCH₃, NH₂, NHCH₃, N(CH₃)₂, OC(O)(C₁₋₂₀ alkyl), which include but are not limited to OC(O)(CH₂)_sCH₃, NHC(O)(C₁₋₂₀ alkyl), which include but are not limited to NHC(O)(CH₂)_sCH₃, and N(C(O)(CH₂)_sCH₃)₂, which include but is not limited to N(C(O)(CH₂)_sCH₃)₂, where s is an integer selected from 0, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, and 19.

[0141] A fifth aspect of the first embodiment is directed to a compound represented by formula I-2

[0142] wherein

[0143] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chlorophenyl, p-fluorophenyl;

[0144] (b) R² is hydrogen or CH₃;

[0145] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl;

[0146] (d) R⁴ is hydrogen, CH₃, Et, 'Pr, 'Pr, 'Bu, 2-butyl, 'Bu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, lower haloalkyl, di(lower alkyl)amino-lower alkyl, or aminoacyl;

[0147] (e) R⁵ is H, CN, CH₃, OCH₃, CH₂OH, CH₂F, halogen, including F, Cl, Br, or I;

[0148] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

[0149] (g) X is H, OH, F, OCH₃, NH₂, or N₃;

[0150] (h) Y is OH, H, CH₃, vinyl, N₃, CN, Cl, Br, F, I, OC(O)CH₃, OCH₃, or O(aminoacyl);

[0151] (i) R⁷ and R⁸ are independently H, F, Cl, Br, I, OH, OCH₃, SH, SCH₃, NH₂, NHCH₃, N(CH₃)₂, CH₃, CH_{3-q}X_q, where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO₂H, CO₂CH₃, CONH₂, CONHCH₃, or CON(CH₃)₂, wherein R¹ is a C₁₋₂₀ alkyl; a C₁₋₂₀ cycloalkyl; a C₂-C₆ alkenyl, a C₂-C₆ alkynyl; and

[0152] (j) R⁹ is selected from among OH, OCH₃, SH, SCH₃, NH₂, NHCH₃, N(CH₃)₂, OC(O)(C₁₋₂₀ alkyl), which include but are not limited to OC(O)(CH₂)_sCH₃, NHC(O)(C₁₋₂₀ alkyl), which include but are not limited to NHC(O)(CH₂)_sCH₃, and N(C(O)(CH₂)_sCH₃)₂, which include but is not limited to N(C(O)(CH₂)_sCH₃)₂, where s is an integer selected from 0, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, and 19.

[0153] A sixth aspect of the first embodiment is directed to a compound represented by formula I-2

[0154] wherein

[0155] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0156] (b) R² is hydrogen or CH₃;

[0157] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0158] (d) R^4 is hydrogen, C_{1-10} alkyl, C_{1-10} alkyl optionally substituted with a lower alkyl, alkoxy or halogen, C_{1-10} haloalkyl, C_{3-10} cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, di(lower alkyl)amino-lower alkyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;

[0159] (e) R^5 is H, OMe, CN, CH_2F , F, Cl, Br, or I;

[0160] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0161] (g) X is H, OH, F, OCH_3 , F, Cl, Br, I, or N_3 ;

[0162] (h) Y is H, OH, CH_3 , F, Cl, Br, I, or N_3 , OCH_3 , $OC(O)CH_3$, or O(aminoacyl);

[0163] (i) R^7 and R^8 are independently H, F, Br, SCH_3 , CH_3 , CH_3-X_q , where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO_2CH_3 , $CONH_2$, $CONHCH_3$, or $CON(CH_3)_2$; and

[0164] (j) R^9 is selected from among OH, OCH_3 , SH, SCH_3 , NH_2 , $NHCH_3$, $N(CH_3)_2$, $OC(O)(C_{1-20}$ alkyl), which include but are not limited to $OC(O)(CH_2)_sCH_3$, $NHC(O)(C_{1-20}$ alkyl), which include but are not limited to $NHC(O)(CH_2)_sCH_3$, and $N(C(O)(CH_2)_sCH_3)_2$, which include but is not limited to $N(C(O)(CH_2)_sCH_3)_2$, where s is an integer selected from 0, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, and 19.

[0165] A seventh aspect of the first embodiment is directed to a compound represented by formula I-2

[0166] wherein

[0167] (a) R^1 is hydrogen, n methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0168] (b) R^2 is hydrogen;

[0169] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0170] (d) R^4 is hydrogen, CH_3 , Et, 'Pr, 'Pr, 'Bu, 2-butyl, 'Bu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl.

[0171] (e) R^5 is H;

[0172] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0173] (g) X is H, OH, OCH_3 , F, or N_3 ;

[0174] (h) Y is OH, OCH_3 , $OC(O)CH_3$, or O(aminoacyl);

[0175] (i) R^7 and R^8 are independently H, F, Br, SCH_3 , CH_3 , CH_3-X_q , where X is F, Cl, Br or I and q is 1 to 3, vinyl, CO_2CH_3 , $CONH_2$, $CONHCH_3$, or $CON(CH_3)_2$; and

[0176] (j) R^9 is selected from among OH, OCH_3 , NH_2 , $NHCH_3$, $N(CH_3)_2$, $OC(O)(C_{1-20}$ alkyl), which include but are not limited to $OC(O)(CH_2)_sCH_3$, $NHC(O)(C_{1-20}$ alkyl), which include but are not limited to $NHC(O)(CH_2)_sCH_3$, and $N(C(O)(CH_2)_sCH_3)_2$, which include but is not limited to $N(C(O)(CH_2)_sCH_3)_2$, where s is an integer selected from among 0, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, and 19.

[0177] An eighth aspect of the first embodiment is directed to a compound represented by formula I-2

[0178] wherein

[0179] (a) R^1 is hydrogen, methyl, phenyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0180] (b) R^2 is hydrogen;

[0181] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0182] (d) R^4 is hydrogen, CH_3 , Et, 'Pr, 'Pr, 'Bu, 2-butyl, 'Bu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclo-

hexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0183] (e) R^5 is H;

[0184] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0185] (g) X is H, OH, OCH_3 , F, or N_3 ;

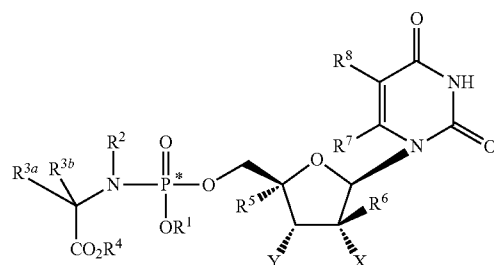
[0186] (h) Y is OH, OCH_3 , $OC(O)CH_3$, or O(aminoacyl);

[0187] (i) R^7 and R^8 are independently H, F, Br, SCH_3 , CH_3 , CH_3-X_q , where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO_2CH_3 , $CONH_2$, $CONHCH_3$, or $CON(CH_3)_2$;

[0188] (j) R^9 is selected from among OH, OCH_3 , NH_2 , $NHCH_3$, $N(CH_3)_2$, $OC(O)(C_{1-20}$ alkyl), which include but are not limited to $OC(O)(CH_2)_sCH_3$, $NHC(O)(C_{1-20}$ alkyl), which include but are not limited to $NHC(O)(CH_2)_sCH_3$, and $N(C(O)(CH_2)_sCH_3)_2$, which include but is not limited to $N(C(O)(CH_2)_sCH_3)_2$, where s is an integer selected from among 0, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, and 19.

[0189] A second embodiment of the invention is directed to a compound represented by formula I in which the base is a structure represented by formula b above, wherein R^1 , R^2 , R^{3a} , R^{3b} , R^4 , R^5 , R^6 , X, Y, R^7 , and R^8 are defined in the Summary of the Invention section above.

[0190] A first aspect of the second embodiment is directed to a compound represented by formula I-3



I-3

[0191] wherein

[0192] (a) R^1 is hydrogen, n-alkyl; branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-6} alkoxy, F, Cl, Br, I, nitro, cyano, C_{1-6} haloalkyl, $-N(R^1)_2$, C_{1-6} acylamino, $-NHSO_2C_{1-6}$ alkyl, $-SO_2N(R^1)_2$, COR^1 , and $-SO_2C_{1-6}$ alkyl; (R^1 is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, R^1 is $-OR^1$ or $-N(R^1)_2$);

[0193] (b) R^2 is hydrogen or CH_3 ;

[0194] (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C_{1-10} alkyl, cycloalkyl, $-(CH)_c(NR^3)_2$, C_{1-6} hydroxyalkyl, $-CH_2SH$, $-(CH_2)_2S(O)_dMe$, $-(CH_2)_3NHC(=NH)NH_2$, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, $-(CH_2)_eCOR^{3'}$, aryl and aryl C_{1-3} alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C_{1-10} alkyl, C_{1-6} alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C_{1-6} alkyl; (iii) R^{3a} and R^{3b} together are $(CH_2)_n$, so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R^2 together are $(CH_2)_n$, so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R^2 together are $(CH_2)_n$, so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where R^3 is independently hydrogen or C_{1-6} alkyl and $R^{3'}$ is $-OR^1$ or

ing N and C atoms, $C(O)CR^{3a}R^{3b}NHR^1$, where n is 2 to 4 and R^1 , R^{3a} , and R^{3b} are as defined herein;

[0216] (c) R^{3a} and R^{3b} are independently (i) R^{3a} is hydrogen and R^{3b} and R^2 together are $(CH_2)_n$, so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R^2 together are $(CH_2)_n$, so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3 f is 3 to 5, n is 2 to 4, and where R^1 is independently hydrogen or C_{1-6} alkyl and R^{3a} is $-OR^1$ or $-N(R^{3a})_2$; (vi) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl; or (viii) R^{3a} is CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl and R^{3b} is H;

[0217] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0218] (e) R^5 is H, CN, CH_3 , OCH_3 , CH_2OH , $CH_2(halo)$, such as CH_2F , N_3 , CH_2CN , CH_2N_3 , CH_2NH_2 , CH_2NHCH_3 , $CH_2N(CH_3)_2$, halogen, including F, Cl, Br, or I;

[0219] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

[0220] (g) X is H, OH, F, OCH_3 , halogen, NH_2 , or N_3 ;

[0221] (h) Y is OH, H, CH_3 , vinyl, N_3 , CN, Cl, Br, F, I, $OC(O)CH_3$, OCH_3 , NH_2 , $NHCH_3$, $NH(vinyl)$, $NH(acetyl)$, $NH(C(O)CH_3)$, $N(CH_3)_2$, $N(C(O)CH_3)_2$;

[0222] (i) R^7 and R^8 are independently H, F, Cl, Br, I, OH, OCH_3 , SH, SCH_3 , NH_2 , $NHCH_3$, $N(CH_3)_2$, CH_3 , CH_3-qX_q , where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO_2H , CO_2CH_3 , $CONH_2$, $CONHCH_3$, or $CON(CH_3)_2$.

[0223] The fourth aspect of the second embodiment is directed to a compound represented by formula I-3

[0224] wherein

[0225] (a) R^1 is hydrogen, n-alkyl or a substituted or unsubstituted phenyl, where the substituent of the substituted phenyl is at least one of a CH_3 , OCH_3 , F, Cl, Br, I, nitro, cyano, and a CH_3-qX_q , where X is F, Cl, Br, or I, and q is 1-3;

[0226] (b) R^2 is hydrogen or CH_3 ;

[0227] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl or R^{3a} is CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl and R^{3b} is H;

[0228] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl,

N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0229] (e) R^5 is H, CN, CH_3 , OCH_3 , CH_2OH , $CH_2(halo)$, such as CH_2F , N_3 , CH_2CN , CH_2N_3 , CH_2NH_2 , CH_2NHCH_3 , $CH_2N(CH_3)_2$, halogen, including F, Cl, Br, or I;

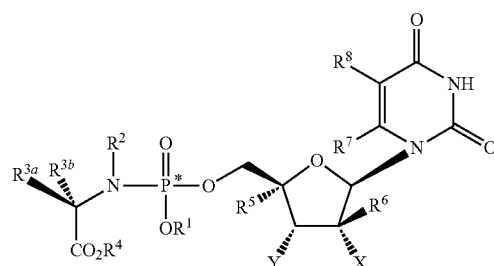
[0230] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

[0231] (g) X is H, OH, F, OCH_3 , NH_2 , or N_3 ;

[0232] (h) Y is OH, H, CH_3 , vinyl, N_3 , CN, Cl, Br, F, I, $OC(O)CH_3$, OCH_3 , NH_2 , $NHCH_3$, $NH(vinyl)$, $NH(acetyl)$, $NH(C(O)CH_3)$, $N(CH_3)_2$, $N(C(O)CH_3)_2$;

[0233] (i) R^7 and R^8 are independently H, F, Cl, Br, I, OH, OCH_3 , SH, SCH_3 , NH_2 , $NHCH_3$, $N(CH_3)_2$, CH_3 , CH_3-qX_q , where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO_2H , CO_2CH_3 , $CONH_2$, $CONHCH_3$, or $CON(CH_3)_2$, wherein R^1 is a C_{1-20} alkyl; a C_{1-20} cycloalkyl; a C_2-C_6 alkenyl, a C_2-C_6 alkynyl.

[0234] The fifth aspect of the second embodiment is directed to a compound represented by formula I-4



I-4

[0235] wherein

[0236] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, or a substituted or unsubstituted phenyl, where the substituent of the substituted phenyl is at least one of a CH_3 , OCH_3 , F, Cl, Br, I, nitro, cyano, and a CH_3-qX_q , where X is F, Cl, Br, or I, and q is 1-3;

[0237] (b) R^2 is hydrogen or CH_3 ;

[0238] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl or R^{3a} is CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl and R^{3b} is H;

[0239] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0240] (e) R^5 is H, CN, CH_3 , OCH_3 , CH_2OH , CH_2F , N_3 , CH_2CN , CH_2N_3 , CH_2NH_2 , CH_2NHCH_3 , $CH_2N(CH_3)_2$, halogen, including F, Cl, Br, or I;

[0241] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

[0242] (g) X is H, OH, F, OCH_3 , Cl, Br, I, NH_2 , or N_3 ;

[0243] (h) Y is OH, H, CH₃, vinyl, N₃, CN, Cl, Br, F, I, OC(O)CH₃, OCH₃, NH₂, NHCH₃, NH(vinyl), NH(acetyl), NH(C(O)CH₃), N(CH₃)₂, N(C(O)CH₃)₂;

[0244] (i) R⁷ and R⁸ are independently H, F, Cl, Br, I, OH, OCH₃, SH, SCH₃, NH₂, NHCH₃, N(CH₃)₂, CH₃, CH_{3-q}X_q, where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO₂H, CO₂CH₃, CONH₂, CONHCH₃, or CON(CH₃)₂, wherein R¹ is a C₁₋₂₀ alkyl; a C₁₋₂₀ cycloalkyl; a C₂-C₆ alkenyl, a C₂-C₆ alkynyl,

[0245] The sixth aspect of the second embodiment is directed to a compound represented by formula I-4

[0246] wherein

[0247] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0248] (b) R² is hydrogen or CH₃;

[0249] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl;

[0250] (d) R⁴ is hydrogen, CH₃, Et, ⁱPr, ⁿPr, ^tBu, 2-butyl, ^tBu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0251] (e) R⁵ is H, CN, CH₃, OCH₃, CH₂OH, CH₂F, halogen, including F, Cl, Br, or I;

[0252] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

[0253] (g) X is H, OH, F, OCH₃, NH₂, or N₃;

[0254] (h) Y is OH, H, CH₃, vinyl, NH₂, N₃, CN, Cl, Br, F, I, OC(O)CH₃, OCH₃;

[0255] (i) R⁷ and R⁸ are independently H, F, Cl, Br, I, OH, OCH₃, SH, SCH₃, NH₂, NHCH₃, N(CH₃)₂, CH₃, CH_{3-q}X_q, where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO₂H, CO₂CH₃, CONH₂, CONHCH₃, or CON(CH₃)₂, wherein R¹ is a C₁₋₂₀ alkyl; a C₁₋₂₀ cycloalkyl; a C₂-C₆ alkenyl, a C₂-C₆ alkynyl.

[0256] The seventh aspect of the second embodiment is directed to a compound represented by formula I-4

[0257] wherein

[0258] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0259] (b) R² is hydrogen or CH₃;

[0260] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0261] (d) R⁴ is hydrogen, CH₃, Et, ⁱPr, ⁿPr, ^tBu, 2-butyl, ^tBu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0262] (e) R⁵ is H, OMe, CN, CH₂F, F, Cl, Br, or I;

[0263] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0264] (g) X is H, OH, F, OCH₃, F, Cl, Br, I, NH₂ or N₃;

[0265] (h) Y is H, OH, CH₃, F, Cl, Br, I, NH₂ or N₃, OCH₃, or OC(O)CH₃;

[0266] (i) R⁷ and R⁸ are independently H, F, Br, SCH₃, CH₃, CH_{3-q}X_q, where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO₂CH₃, CONH₂, CONHCH₃, or CON(CH₃)₂;

[0267] The eighth aspect of the second embodiment is directed to a compound represented by formula I-4

[0268] wherein

[0269] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chlorophenyl, p-fluorophenyl;

[0270] (b) R² is hydrogen;

[0271] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0272] (d) R⁴ is hydrogen, CH₃, Et, ⁱPr, ⁿPr, ^tBu, 2-butyl, ^tBu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0273] (e) R⁵ is H;

[0274] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0275] (g) X is H, OH, OCH₃, F, NH₂ or N₃;

[0276] (h) Y is OH, NH₂, OCH₃, or OC(O)CH₃;

[0277] (i) R⁷ and R⁸ are independently H, F, Br, SCH₃, CH₃, CH_{3-q}X_q, where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO₂CH₃, CONH₂, CONHCH₃, or CON(CH₃)₂.

[0278] The ninth aspect of the second embodiment is directed to a compound represented by formula I-4

[0279] wherein

[0280] (a) R¹ is hydrogen, methyl, phenyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0281] (b) R² is hydrogen;

[0282] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0283] (d) R⁴ is hydrogen, CH₃, Et, ⁱPr, ⁿPr, ^tBu, 2-butyl, ^tBu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0284] (e) R⁵ is H;

[0285] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0286] (g) X is H, OH, OCH₃, F, or N₃;

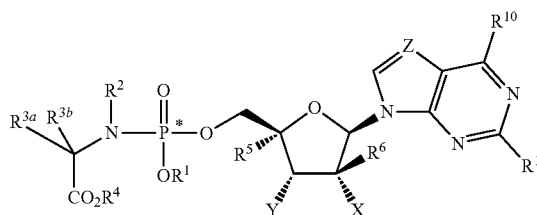
[0287] (h) Y is OH, OCH₃, or OC(O)CH₃;

[0288] (i) R⁷ and R⁸ are independently H, F, Br, SCH₃, CH₃, CH_{3-q}X_q, where X is F, Cl, Br, or I and q is 1 to 3, vinyl, CO₂CH₃, CONH₂, CONHCH₃, or CON(CH₃)₂.

[0289] A third embodiment of the invention is directed to a compound represented by formula I in which the base is a structure represented by formula c above, wherein R¹, R², R^{3a}, R^{3b}, R⁴, R⁵, R⁶, X, Y, Z, R¹⁰, R¹¹, and R¹² are defined in the Summary of the Invention section above; with the proviso that R¹¹ is not H.

[0290] A first aspect of the third embodiment is directed to a compound represented by formula I-5

I-5



[0291] wherein

[0292] (a) R^1 is hydrogen, n-alkyl; branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-6} alkoxy, F, Cl, Br, I, nitro, cyano, C_{1-6} haloalkyl, $-N(R^{1'})_2$, C_{1-6} acylamino, $-NHSO_2C_{1-6}$ alkyl, $-SO_2N(R^{1'})_2$, $COR^{1'}$, and $-SO_2C_{1-6}$ alkyl; ($R^{1'}$ is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, $R^{1''}$ is $-OR'$ or $-N(R^{1'})_2$);

[0293] (b) R^2 is hydrogen or CH_3 ;

[0294] (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C_{1-10} alkyl, cycloalkyl, $-(CH_2)_c(NR^{3'})_2$, C_{1-6} hydroxyalkyl, $-CH_2SH$, $-(CH_2)_2S(O)_2Me$, $-(CH_2)_3NHC(=NH)NH_2$, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, $-(CH_2)_cCOR^{3''}$, aryl and aryl C_{1-3} alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C_{1-10} alkyl, C_{1-6} alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C_{1-6} alkyl; (iii) R^{3a} and R^{3b} together are $(CH_2)_n$ so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where $R^{3'}$ is independently hydrogen or C_{1-6} alkyl and $R^{3''}$ is $-OR'$ or $-N(R^{3'})_2$; (vi) R^{3a} is H and R^{3b} is from H, CH_3 , CH_2CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl; or (viii) R^{3a} is CH_3 , $-CH_2CH_3$, $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl and R^{3b} is H, where $R^{3'}$ is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, $R^{3''}$ is $-OR'$ or $-N(R^{3'})_2$;

[0295] (d) R^4 is hydrogen, C_{1-10} alkyl, C_{1-10} alkyl optionally substituted with a lower alkyl, alkoxy or halogen, C_{1-10} haloalkyl, C_{3-10} cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, di(lower alkyl)amino-lower alkyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;

[0296] (e) R^5 is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., $-(CH_2)_pOH$, where p is 1-6, including hydroxyl methyl (CH_2OH), CH_2F , N_3 , CH_2CN , CH_2NH_2 , CH_2NHCH_3 , $CH_2N(CH_3)_2$, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R^6 is H, R^5 cannot be N_3 and when X is OH, R^6 is CH_3 or CH_2F and B is a purine base, R^5 cannot be H;

[0297] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

[0298] (g) X is H, OH, F, OMe, halogen, NH_2 , or N_3 ;

[0299] (h) Y is OH, H, C_{1-4} alkyl, C_{2-4} alkenyl, C_{2-4} alkynyl, vinyl, N_3 , CN, Cl, Br, F, I, NO_2 , $OC(O)O(C_{1-4}$ alkyl), $OC(O)O(C_{1-4}$ alkyl), $OC(O)O(C_{2-4}$ alkynyl), $OC(O)O(C_{2-4}$ alkenyl), OC_{1-10} haloalkyl, O(aminoacyl), O(C_{1-10} acyl), O(C_{1-4} alkyl), O(C_{2-4} alkenyl), S(C_{1-4} acyl), S(C_{1-4} alkyl), S(C_{2-4} alkynyl), S(C_{2-4} alkenyl), SO(C_{1-4} acyl), SO(C_{1-4} alkyl),

SO(C_{2-4} alkynyl), SO(C_{2-4} alkenyl), $SO_2(C_{1-4}$ acyl), $SO_2(C_{1-4}$ alkyl), $SO_2(C_{2-4}$ alkynyl), $SO_2(C_{2-4}$ alkenyl), OS(O)₂(C_{1-4} acyl), OS(O)₂(C_{1-4} alkyl), OS(O)₂(C_{2-4} alkenyl), NH_2 , NH(C_{1-4} alkyl), NH(C_{2-4} alkenyl), NH(C_{2-4} alkynyl), NH(C_{1-4} acyl), N(C_{1-4} alkyl)₂, N(C_{1-18} acyl)₂, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N_3 , CN, one to three halogen (Cl, Br, F, I), NO_2 , C(O)O(C_{1-4} alkyl), C(O)O(C_{1-4} alkyl), C(O)O(C_{2-4} alkynyl), C(O)O(C_{2-4} alkenyl), O(C_{1-4} acyl), O(C_{1-4} alkyl), O(C_{2-4} alkenyl), S(C_{1-4} acyl), S(C_{1-4} alkyl), S(C_{2-4} alkynyl), S(C_{2-4} alkenyl), SO(C_{1-4} acyl), SO(C_{1-4} alkyl), SO(C_{2-4} alkynyl), SO(C_{2-4} alkenyl), $SO_2(C_{1-4}$ acyl), $SO_2(C_{1-4}$ alkyl), $SO_2(C_{2-4}$ alkynyl), $SO_2(C_{2-4}$ alkenyl), OS(O)₂(C_{1-4} acyl), OS(O)₂(C_{1-4} alkyl), OS(O)₂(C_{2-4} alkenyl), NH_2 , NH(C_{1-4} alkyl), NH(C_{2-4} alkenyl), NH(C_{2-4} alkynyl), NH(C_{1-4} acyl), N(C_{1-4} alkyl)₂, N(C_{1-4} acyl)₂;

[0300] (i) R^{10} and R^{11} are independently H, F, Cl, Br, I, OH, OR', SH, SR', NH_2 , NHR', NR', lower alkyl of C_1-C_6 , halogenated (F, Cl, Br, I) lower alkyl of C_1-C_6 , lower alkenyl of C_2-C_6 , halogenated (F, Cl, Br, I) lower alkenyl of C_2-C_6 , lower alkynyl of C_2-C_6 such as $C\equiv CH$, halogenated (F, Cl, Br, I) lower alkynyl of C_2-C_6 , lower alkoxy of C_1-C_6 , halogenated (F, Cl, Br, I) lower alkoxy of C_1-C_6 , CO_2H , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$, with the proviso that when R^{10} is OH and R^{11} is not NH_2 ;

[0301] wherein R' is an optionally substituted alkyl, which includes, but is not limited to, an optionally substituted C_{1-20} alkyl, an optionally substituted C_{1-10} alkyl, an optionally substituted lower alkyl; an optionally substituted cycloalkyl; an optionally substituted alkynyl of C_2-C_6 , an optionally substituted lower alkenyl of C_2-C_6 , or optionally substituted acyl, which includes but is not limited to C(O) alkyl, C(O)(C_{1-20} alkyl), C(O)(C_{1-10} alkyl), or C(O)(lower alkyl) or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0302] (j) Z is N or CR^{12} ; and

[0303] R^{12} is an H, halogen (including F, Cl, Br, I), OH OR', SH, SR', NH_2 , NHR', NR', NO_2 lower alkyl of C_1-C_6 halogenated (F, Cl, Br, I) lower alkyl of C_1-C_6 , lower alkenyl of C_2-C_6 , halogenated (F, Cl, Br, I) lower alkenyl of C_2-C_6 , lower alkynyl of C_2-C_6 , halogenated (F, Cl, Br, I) lower alkynyl of C_2-C_6 , lower alkoxy of C_1-C_6 , halogenated (F, Cl, Br, I) lower alkoxy of C_1-C_6 , CO_2H , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0304] A second aspect of the third embodiment is directed to a compound represented by formula 1-5

[0305] wherein

[0306] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0307] (b) R^2 is hydrogen or CH_3 ;

[0308] (c) R^{3a} is H and R^{3b} is H CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-C_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl;

[0309] (d) R^4 is hydrogen, CH_3 , Et, iPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0310] (e) R⁵ is H, CN, CH₃, OCH₃, CH₂OH, CH₂F, halogen, including F, Cl, Br, or I, with the provisos that when X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H.

[0311] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

[0312] (g) X is H, OH, F, OCH₃, Cl, Br, I, NH₂, or N₃;

[0313] (h) Y is OH, H, CH₃, vinyl, NH₂, N₃, CN, Cl, Br, F, I, OC(O)CH₃, OCH₃;

[0314] (i) R¹⁰ and R¹¹ are independently H, F, Br, I, OH, OR', NH₂, NHR', NR'₂, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R', with the proviso that when R¹⁰ is OH and R¹¹ is not NH₂;

[0315] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0316] (j) Z is N or CR¹²; and

[0317] R¹² is an H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'.

[0318] A third aspect of the third embodiment is directed to a compound represented by formula I-5

[0319] wherein

[0320] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0321] (b) R² is hydrogen or CH₃;

[0322] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0323] (d) R⁴ is hydrogen, CH₃, Et, ^tPr, ⁿPr, ⁱBu, 2-butyl, ^tBu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0324] (e) R⁵ is H, CN, CH₂F, F, Cl, Br, or I; with the proviso that X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H;

[0325] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0326] (g) X is H, OH, F, OCH₃, F, Cl, Br, I, NH₂ or N₃;

[0327] (h) Y is H, OH, CH₃, F, Cl, Br, I, NH₂ or N₃, OCH₃, or OC(O)CH₃;

[0328] (i) R¹⁰ and R¹¹ are independently H, F, Br, I, OH, OR', NH₂, NHR', NR'₂, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R', with the proviso that when R¹⁰ is OH and R¹¹ is not NH₂;

[0329] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0330] (j) Z is N or CR¹²; and

[0331] R¹² is an H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'.

[0332] A fourth aspect of the third embodiment is directed to a compound represented by formula I-5

[0333] wherein

[0334] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0335] (b) R² is hydrogen;

[0336] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0337] (d) R⁴ is hydrogen, CH₃, Et, ^tPr, ⁿPr, ⁱBu, 2-butyl, ^tBu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-aziridin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0338] (e) R⁵ is H, with the provisos that when X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H;

[0339] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0340] (g) X is H, OH, OCH₃, F, NH₂ or N₃;

[0341] (h) Y is OH, NH₂, OCH₃, or OC(O)CH₃;

[0342] (i) R¹⁰ and R¹¹ are independently H, F, Br, I, OH, OR', NH₂, NHR', NR'₂, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R', with the proviso that when R¹⁰ is OH and R¹¹ is not NH₂;

[0343] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms, and

[0344] (j) Z is N or CR¹²; and

[0345] R¹² is an H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'.

[0346] A fifth aspect of the third embodiment is directed to a compound represented by formula I-5

[0347] wherein

[0348] (a) R¹ is hydrogen, methyl, phenyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0349] (b) R² is hydrogen;

[0350] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0351] (d) R⁴ is hydrogen, CH₃, Et, ^tPr, ⁿPr, ⁱBu, 2-butyl, ^tBu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-aziridin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0352] (e) R⁵ is H, with the provisos that when X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H;

[0353] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0354] (g) X is H, OH, OCH₃, F, NH₂ or N₃;

[0355] (h) Y is OH, NH₂, OCH₃, or OC(O)CH₃;

[0356] (i) R¹⁰ and R¹¹ are independently H, F, Br, I, OH, OR', NH₂, NHR', NR'₂, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R', with the proviso that when R¹⁰ is OH and R¹¹ is not NH₂;

[0357] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0358] (j) Z is N or CR¹²; and

[0359] R¹² is an H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'.

[0360] A sixth aspect of the third embodiment is directed to a compound represented by formula I-5

[0361] wherein

[0362] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0363] (b) R² is hydrogen or CH₃;

[0364] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl;

[0365] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-aziridin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0366] (e) R^5 is H, CN, CH_3 , OCH_3 , CH_2OH , CH_2F , halogen, including F, Cl, Br, or I, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0367] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

[0368] (g) X is H, OH, F, OCH_3 , Cl, Br, I, NH_2 , or N_3 ;

[0369] (h) Y is OH, H, CH_3 , vinyl, NH_2 , N_3 , CN, Cl, Br, F, I, $OC(O)CH_3$, OCH_3 ;

[0370] (i) R^{10} is NH_2 and R^{11} is H, F, Br, I, OH, OR' , NH_2 , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$;

[0371] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0372] (j) Z is N or CR^{12} , and

[0373] R^{12} is an H, halogen (including F, Cl, Br, I), OR' , NH_2 , NHR' , NR'_2 , NO_2 , lower alkyl of C_1 - C_6 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0374] A seventh aspect of the third embodiment is directed to a compound represented by formula I-5

[0375] wherein

[0376] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0377] (b) R^2 is hydrogen or CH_3 ;

[0378] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0379] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-aziridin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0380] (e) R^5 is H, CN, CH_2F , F, Cl, Br, or I, with the proviso that X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0381] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0382] (g) X is H, OH, F, OCH_3 , F, Cl, Br, I, NH_2 or N_3 ;

[0383] (h) Y is H, OH, CH_3 , F, Cl, Br, I, NH_2 or N_3 , OCH_3 , or $OC(O)CH_3$;

[0384] (i) R^{10} is NH_2 and R^{11} is H, F, Br, I, OH, OR' , NH_2 , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$;

[0385] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0386] (j) Z is N or CR^{12} ; and

[0387] R^{12} is an H, halogen (including F, Cl, Br, I), OR' , NH_2 , NHR' , NR'_2 , NO_2 , lower alkyl of C_1 - C_6 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0388] An eighth aspect of the third embodiment is directed to a compound represented by formula I-5

[0389] wherein

[0390] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0391] (b) R^2 is hydrogen;

[0392] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0393] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-aziridin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl;

[0394] (e) R^5 is H, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0395] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0396] (g) X is H, OH, OCH_3 , F, NH_2 or N_3 ;

[0397] (h) Y is OH, NH_2 , OCH_3 , or $OC(O)CH_3$;

[0398] (i) R^{10} is NH_2 and R^{11} is H, F, Br, I, OH, OR' , NH_2 , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$;

[0399] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'_2 each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0400] (j) Z is N or CR^{12} ; and

[0401] R^{12} is an H, halogen (including F, Cl, Br, I), OR' , NH_2 , NHR' , NR'_2 , NO_2 , lower alkyl of C_1 - C_6 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0402] A ninth aspect of the third embodiment is directed to a compound represented by formula I-5

[0403] wherein

[0404] (a) R^1 is hydrogen, methyl, phenyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0405] (b) R^2 is hydrogen;

[0406] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0407] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, or N-methyl-pyrrolidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0408] (e) R^5 is H, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0409] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0410] (g) X is H, OH, OCH_3 , F, NH_2 or N_3 ;

[0411] (h) Y is OH, NH_2 , OCH_3 , or $OC(O)CH_3$;

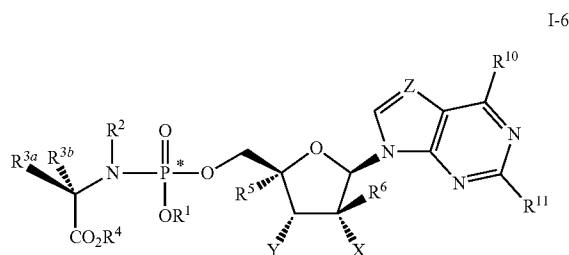
[0412] (i) R^{10} is NH_2 and R^{11} is H, F, Br, I, OH, OR' , NH_2 , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$;

[0413] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0414] (j) Z is N or CR¹²; and

[0415] R¹² is an H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'.

[0416] A tenth aspect of the third embodiment is directed to a compound represented by formula I-6



[0417] wherein

[0418] (a) R¹ is hydrogen, n-alkyl; branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C₁₋₆ alkyl, C₂₋₆ alkenyl, C₂₋₆ alkynyl, C₁₋₆ alkoxy, F, Cl, Br, I, nitro, cyano, C₁₋₆ haloalkyl, —N(R^{1'})₂, C₁₋₆ acylamino, —NHSO₂C₁₋₆ alkyl, —SO₂N(R^{1'})₂, COR^{1'}, and —SO₂C₁₋₆ alkyl; (R^{1'} is independently hydrogen or alkyl, which includes, but is not limited to, C₁₋₂₀ alkyl, C₁₋₁₀ alkyl, or C₁₋₆ alkyl, R^{1''} is —OR' or —N(R^{1'})₂);

[0419] (b) R² is hydrogen or CH₃;

[0420] (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C₁₋₁₀ alkyl, cycloalkyl, —(CH₂)_c(NR^{3'})₂, C₁₋₆ hydroxyalkyl, —CH₂SH, —CH₂S(O)_dMe, —(CH₂)₂NHC(=NH)NH₂, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, —(CH₂)_cCOR^{3''}, aryl and aryl C₁₋₃ alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C₁₋₁₀ alkyl, C₁₋₆ alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C₁₋₆ alkyl; (iii) R^{3a} and R^{3b} together are (CH₂)_e so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R² together are (CH₂)_n so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R² together are (CH₂)_n so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where R^{3'} is independently hydrogen or C₁₋₆ alkyl and R^{3''} is —OR' or —N(R^{3'})₂; (vi) R^{3a} is H and R^{3b} is H, CH₃, CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl; or (viii) R^{3a} is CH₃, —CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), Ph), CH₂SH, or lower cycloalkyl and R^{3b} is H, where R^{3'} is independently hydrogen or alkyl, which includes, but is not limited to, C₁₋₂₀ alkyl, C₁₋₁₀ alkyl, or C₁₋₆ alkyl, R^{3''} is —OR' or —N(R^{3'})₂;

[0421] (d) R⁴ is hydrogen, C₁₋₁₀ alkyl, C₁₋₁₀ alkyl optionally substituted with a lower alkyl, alkoxy or halogen, C₁₋₁₀ haloalkyl, C₃₋₁₀ cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, di(lower alkyl)amino-lower alkyl, aryl,

such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;

[0422] (e) R⁵ is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., —(CH₂)_pOH, where p is 1-6, including hydroxyl methyl (CH₂OH), CH₂F, N₃, CH₂CN, CH₂NH₂, CH₂NHCH₃, CH₂N(CH₃)₂, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OR, base is cytosine and R⁶ is H, R⁵ cannot be N₃ and when X is OH, R⁶ is CH₃ or CH₂F and B is a purine base, R⁵ cannot be H;

[0423] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

[0424] (g) X is H, OH, F, OMe, Cl, Br, I, NH₂, or N₃;

[0425] (h) Y is OH, H, C₁₋₄ alkyl, C₂₋₄ alkenyl, C₂₋₄ alkynyl, vinyl, N₃, CN, Cl, Br, F, I, NO₂, OC(O)O(C₁₋₄ alkyl), OC(O)O(C₁₋₄ alkyl), OC(O)O(C₂₋₄ alkynyl), OC(O)O(C₂₋₄ alkenyl), OC₁₋₁₀ haloalkyl, O(aminoacyl), O(C₁₋₁₀ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₄ alkyl), S(C₂₋₄ alkynyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl), SO(C₁₋₄ alkyl), SO(C₂₋₄ alkynyl), SO(C₂₋₄ alkenyl), SO₂(C₁₋₄ acyl), SO₂(C₁₋₄ alkyl), SO₂(C₂₋₄ alkynyl), SO₂(C₂₋₄ alkenyl), OS(O)₂(C₁₋₄ acyl), OS(O)₂(C₁₋₄ alkyl), OS(O)₂(C₂₋₄ alkenyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkenyl), NH(C₂₋₄ alkynyl), NH(C₁₋₄ acyl), N(C₁₋₄ alkyl)₂, N(C₁₋₁₈ acyl)₂, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N₃, CN, one to three halogen (Cl, Br, F, I), NO₂, C(O)O(C₁₋₄ alkyl), C(O)O(C₁₋₄ alkyl), C(O)O(C₂₋₄ alkynyl), C(O)O(C₂₋₄ alkenyl), O(C₁₋₄ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₄ alkyl), S(C₂₋₄ alkynyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl), SO(C₁₋₄ alkyl), SO(C₂₋₄ alkynyl), SO(C₂₋₄ alkenyl), SO₂(C₁₋₄ acyl), SO₂(C₁₋₄ alkyl), SO₂(C₂₋₄ alkynyl), SO₂(C₂₋₄ alkenyl), OS(O)₂(C₁₋₄ acyl), OS(O)₂(C₁₋₄ alkyl), OS(O)₂(C₂₋₄ alkenyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkenyl), NH(C₂₋₄ alkynyl), NH(C₁₋₄ acyl), N(C₁₋₄ alkyl)₂, N(C₁₋₄ acyl)₂;

[0426] (i) R¹⁰ and R¹¹ are independently H, F, Br, I, OH, OR', NH₂, NHR', NR'₂, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R', with the proviso that when R¹⁰ is OH and R¹¹ is not NH₂;

[0427] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0428] (j) Z is N or CR¹²; and

[0429] R¹² is an H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'.

[0430] An eleventh aspect of the third embodiment is directed to a compound represented by formula I-6

[0431] wherein

[0432] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0433] (b) R² is hydrogen or CH₃;

[0434] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, C₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl;

[0435] (d) R⁴ is hydrogen, CH₃, Et, ⁱPr, ⁿPr, ⁿBu, 2-butyl, ^tBu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl,

N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0436] (e) R^5 is H, CN, CH_3 , OCH_3 , CH_2OH , CH_2F , halogen, including F, Cl, Br, or I, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0437] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

[0438] (g) X is H, OH, F, OCH_3 , Cl, Br, I, NH_2 , or N_3 ;

[0439] (h) Y is OH, H, CH_3 , vinyl, NH_2 , N_3 , CN, Cl, Br, F, I, $OC(O)CH_3$, OCH_3 ;

[0440] (i) R^{10} and R^{11} are independently H, F, Br, I, OH, OR' , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$, with the proviso that when R^{10} is OH and R^{11} is not NH_2 ;

[0441] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0442] (j) Z is N or CR^{12} ; and

[0443] R^{12} is an H, halogen (including F, Cl, Br, I), OR' , NH_2 , NHR' , NR'_2 , NO_2 , lower alkyl of C_1 - C_6 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0444] A twelfth aspect of the third embodiment is directed to a compound represented by formula I-6

[0445] wherein

[0446] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0447] (b) R^2 is hydrogen or CH_3 ;

[0448] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0449] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0450] (e) R^5 is H, CN, CH_2F , F, Cl, Br, or I, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0451] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0452] (g) X is H, OH, F, OCH_3 , F, Cl, Br, I, NH_2 or N_3 ;

[0453] (h) Y is H, OH, CH_3 , F, Cl, Br, I, NH_2 or N_3 , OCH_3 , or $OC(O)CH_3$;

[0454] (i) R^{10} and R^{11} are independently H, F, Br, I, OH, OR' , NH_2 , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$, with the proviso that when R^{10} is OH and R^{11} is not NH_2 ;

[0455] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0456] (j) Z is N or CR^{12} ; and

[0457] R^{12} is a H, halogen (including F, Cl, Br, I), OR' , NH_2 , NHR' , NR'_2 , NO_2 , lower alkyl of C_1 - C_6 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0458] A thirteenth aspect of the third embodiment is directed to a compound represented by formula I-6

[0459] wherein

[0460] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0461] (b) R^2 is hydrogen;

[0462] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0463] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0464] (e) R^5 is H, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0465] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0466] (g) X is H, OH, OCH_3 , F, NH_2 or N_3 ;

[0467] (h) Y is OH, NH_2 , OCH_3 , or $OC(O)CH_3$;

[0468] (i) R^{10} and R^{11} are independently H, F, Br, I, OH, OR' , NH_2 , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$, with the proviso that when R^{10} is OH and R^{11} is not NH_2 ;

[0469] wherein R' is a lower alkyl, a lower cycloalkyl, or C(Q)(lower alkyl) or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0470] (j) Z is N or CR^{12} ; and

[0471] R^{12} is an H, halogen (including F, Cl, Br, I), OR' , NH_2 , NHR' , NR'_2 , NO_2 , lower alkyl of C_1 - C_6 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0472] A fourteenth aspect of the third embodiment is directed to a compound represented by formula I-6

[0473] wherein

[0474] (a) R^1 is hydrogen, methyl, phenyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0475] (b) R^2 is hydrogen;

[0476] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0477] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0478] (e) R^5 is H, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0479] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0480] (g) X is H, OH, OCH_3 , F, NH_2 or N_3 ;

[0481] (h) Y is OH, NH_2 , OCH_3 , or $OC(O)CH_3$;

[0482] (i) R^{10} and R^{11} are independently H, F, Br, I, OH, OR' , NH_2 , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$, with the proviso that when R^{10} is OH and R^{11} is not NH_2 ;

[0483] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0484] (j) Z is N or CR^{12} ; and

[0485] R^{12} is an H, halogen (including F, Cl, Br, I), OR' , NH_2 , NHR' , NR'_2 , NO_2 , lower alkyl of C_1 - C_6 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0486] A fifteenth aspect of the third embodiment is directed to a compound represented by formula I-6

[0487] wherein

[0488] (a) R^1 is hydrogen, n-alkyl, branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally

substituted with at least one of C_{1-6} alkyl, C_{2-4} alkenyl, C_{2-6} alkynyl, C_{1-6} alkoxy, F, Cl, Br, I, nitro, cyano, C_{1-6} haloalkyl, $-N(R^{1'})_2$, C_{1-6} acylamino, $-NHSO_2C_{1-6}$ alkyl, $-SO_2N(R^{1'})_2$, $COR^{1'}$, and $-SO_2C_{1-6}$ alkyl, ($R^{1'}$ is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, $R^{1'}$ is $-OR'$ or $-N(R^{1'})_2$);

[0489] (b) R^2 is hydrogen or CH_3 ;

[0490] (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C_{1-10} alkyl, cycloalkyl, $-(CH_2)_c(NR^3)_2$, C_{1-6} hydroxyalkyl, $-CH_2SH$, $-(CH_2)_2S(O)_dMe$, $-(CH_2)_3NHC(=NH)NH_2$, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, $-(CH_2)_cCOR^{3''}$, aryl and aryl C_{1-3} alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C_{1-10} alkyl, C_{1-6} alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C_{1-6} alkyl; (iii) R^{3a} and R^{3b} together are $(CH_2)_c$ so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where $R^{3''}$ is independently hydrogen or C_{1-6} alkyl and $R^{3''}$ is $-OR'$ or $-N(R^{3''})_2$; (vi) R^{3b} is H and R^{3b} is H, CH_3 , CH_2CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl; or (viii) R^{3a} is CH_3 , $-CH_2CH_3$, $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl and R^{3b} is H, where $R^{3'}$ is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, $R^{3''}$ is $-OR'$ or $-N(R^{3''})_2$;

[0491] (d) R^4 is hydrogen, C_{1-10} alkyl, C_{1-10} alkyl optionally substituted with a lower alkyl, alkoxy or halogen, C_{1-10} haloalkyl, C_{3-10} cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, di(lower alkyl)amino-lower alkyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;

[0492] (e) R^5 is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., $-(CH_2)_pOH$, where p is 1-6, including hydroxyl methyl (CH_2OH), CH_2F , N_3 , CH_2CN , CH_2NH_2 , CH_2NHCH_3 , $CH_2N(CH_3)_2$, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R^6 is H, R^5 cannot be N_3 and when X is OH, R^6 is CH_3 or CH_2F and R^5 is a purine base, R^5 cannot be H;

[0493] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

[0494] (g) X is H, OH, F, OMe, halogen, NH_2 , or N_3 ;

[0495] (h) Y is OH, H, C_{1-4} alkyl, C_{2-4} alkenyl, C_{2-4} alkynyl, vinyl, N_3 , CN, Cl, Br, F, I, NO_2 , $OC(O)O(C_{1-4}$ alkyl), $OC(O)O(C_{1-4}$ alkyl), $OC(O)O(C_{2-4}$ alkynyl), $OC(O)O(C_{2-4}$ alkenyl), OC_{1-10} haloalkyl, O(aminoacyl), O(C_{1-10} acyl), O(C_{1-4} alkyl), O(C_{2-4} alkenyl), S(C_{1-4} acyl), S(C_{1-4} alkyl), S(C_{2-4} alkynyl), S(C_{2-4} alkenyl), SO(C_{1-4} acyl), SO(C_{1-4} alkyl), SO(C_{2-4} alkynyl), SO(C_{2-4} alkenyl), $SO_2(C_{1-4}$ acyl), $SO_2(C_{1-4}$ alkyl), $SO_2(C_{2-4}$ alkynyl), $SO_2(C_{2-4}$ alkenyl), OS(O_2 (C_{1-4} acyl), OS(O_2 (C_{1-4} alkyl), OS(O_2 (C_{2-4} alkenyl), NH_2 , NH(C_{1-4} alkyl), NH(C_{2-4} alkenyl), NH(C_{2-4} alkynyl),

NH(C_{1-4} acyl), N(C_{1-4} alkyl) $_2$, N(C_{1-18} acyl) $_2$, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N_3 , CN, one to three halogen (Cl, Br, F, I), NO_2 , C(O)O(C_{1-4} alkyl), C(O)O(C_{1-4} alkyl), C(O)O(C_{2-4} alkynyl), C(O)O(C_{2-4} alkenyl), O(C_{1-4} acyl), O(C_{1-4} alkyl), O(C_{2-4} alkynyl), S(C_{1-4} acyl), S(C_{1-4} alkyl), S(C_{2-4} alkynyl), S(C_{2-4} alkenyl), SO(C_{1-4} acyl), SO(C_{1-4} alkyl), SO(C_{2-4} alkynyl), SO(C_{2-4} alkenyl), $SO_2(C_{1-4}$ acyl), $SO_2(C_{2-4}$ alkyl), $SO_2(C_{2-4}$ alkynyl), $SO_2(C_{2-4}$ alkenyl), OS(O_2 (C_{1-4} acyl), OS(O_2 (C_{1-4} alkyl), OS(O_2 (C_{2-4} alkenyl), NH_2 , NH(C_{1-4} alkyl), NH(C_{2-4} alkenyl), NH(C_{2-4} alkynyl), NH(C_{1-4} acyl), N(C_{1-4} alkyl) $_2$, N(C_{1-4} acyl) $_2$;

[0496] (i) R^{10} is NH_2 and R^{11} is H, F, Br, I, OH, OR' , NH_2 , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$;

[0497] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0498] (j) Z is N or CR^{12} ; and

[0499] R^{12} is an H, halogen (including F, Cl, Br, I), OR' , NH_2 , NHR' , NR'_2 , NO_2 , lower alkyl of C_1-C_6 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0500] An sixteenth aspect of the third embodiment is directed to a compound represented by formula I-6

[0501] wherein

[0502] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0503] (b) R^2 is hydrogen or CH_3 ;

[0504] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl;

[0505] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, iBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0506] (e) R^5 is H, CN, CH_3 , OCH_3 , CH_2OH , CH_2F , halogen, including F, Cl, Br, or I, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0507] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

[0508] (g) X is H, OH, F, OCH_3 , Cl, Br, I, NH_2 , or N_3 ;

[0509] (h) Y is OH, H, CH_3 , vinyl, NH_2 , N_3 , CN, Cl, Br, F, I, $OC(O)CH_3$, OCH_3 ;

[0510] (i) R^{10} is NH_2 and R^{11} is H, F, Br, I, OH, OR' , NH_2 , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$;

[0511] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0512] (j) Z is N or CR^{12} ; and

[0513] R^{12} is an H, halogen (including F, Cl, Br, I), OR' , NH_2 , NHR' , NR'_2 , NO_2 , lower alkyl of C_1-C_6 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0514] A seventeenth aspect of the third embodiment is directed to a compound represented by formula I-6

[0515] wherein

[0516] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0517] (b) R^2 is hydrogen or CH_3 ;

[0518] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0519] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , tBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0520] (e) R^5 is H, CN, CH_2F , F, Cl, Br, or I, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0521] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0522] (g) X is H, OH, F, OCH_3 , F, Cl, Br, I, NH_2 or N_3 ;

[0523] (h) Y is H, OH, CH_3 , F, Cl, Br, I, NH_2 or N_3 , OCH_3 , or $OC(O)CH_3$;

[0524] (i) R^{10} is NH_2 and R^{11} is H, F, Br, I, OH, OR' , NH_2 , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$;

[0525] wherein R' is a lower alkyl, a lower cycloalkyl, or $C(O)(\text{lower alkyl})$ or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0526] (j) Z is N or CR^{12} ; and

[0527] R^{12} is an H, halogen (including F, Cl, Br, I), OR' , NH_2 , NHR' , NR'_2 , NO_2 , lower alkyl of C_1 - C_6 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0528] An eighteenth aspect of the third embodiment is directed to a compound represented by formula I-6

[0529] wherein

[0530] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0531] (b) R^2 is hydrogen;

[0532] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0533] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , tBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0534] (e) R^5 is H, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0535] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0536] (g) X is H, OH, OCH_3 , F, NH_2 or N_3 ;

[0537] (h) Y is OH, NH_2 , OCH_3 , or $OC(O)CH_3$;

[0538] (i) R^{10} is NH_2 and R^{11} is H, F, Br, I, OH, OR' , NH_2 , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$;

[0539] wherein R' is a lower alkyl, a lower cycloalkyl, or $C(O)(\text{lower alkyl})$ or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0540] (j) Z is N or CR^{12} ; and

[0541] R^{12} is an H, halogen (including F, Cl, Br, I), OR' , NH_2 , NHR' , NR'_2 , NO_2 , lower alkyl of C_1 - C_6 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0542] A nineteenth aspect of the third embodiment is directed to a compound represented by formula I-6

[0543] wherein

[0544] (a) R^1 is hydrogen, methyl, phenyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0545] (b) R^2 is hydrogen;

[0546] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0547] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , tBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0548] (e) R^5 is H, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0549] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0550] (g) X is H, OH, OCH_3 , F, NH_2 or N_3 ;

[0551] (h) Y is OH, NH_2 , OCH_3 , or $OC(O)CH_3$;

[0552] (i) R^{10} is NH_2 and R^{11} are independently H, F, Br, I, OH, OR' , NH_2 , NHR' , NR'_2 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$;

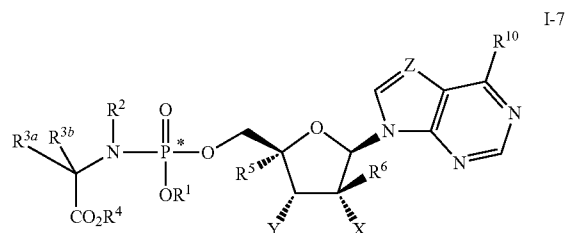
[0553] wherein R' is a lower alkyl, a lower cycloalkyl, or $C(O)(\text{lower alkyl})$ or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0554] (j) Z is N or CR^{12} ; and

[0555] R^{12} is an H, halogen (including F, Cl, Br, I), OR' , NH_2 , NHR' , NR'_2 , NO_2 , lower alkyl of C_1 - C_6 , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$.

[0556] A fourth embodiment of the invention is directed to a compound represented by formula I in which the base is a structure represented by formula c above, where R^{11} is H and R^2 , R^{3a} , R^{3b} , R^4 , R^5 , R^6 , X, and Y are defined in the Summary of the Invention section above.

[0557] A first aspect of the fourth embodiment is directed to a compound represented by formula I-7



[0558] wherein

[0559] (a) R^1 is hydrogen, n-alkyl; branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-6} alkoxy, F, Cl, Br, I, nitro, cyano, C_{1-6} haloalkyl, $-N(R^{11})_2$, C_{1-6} acylamino, $-NHSO_2C_{1-6}$ alkyl, $-SO_2N(R^{11})_2$, COR^{11} , and $-SO_2C_{1-6}$ alkyl; (R^{11} is independently

hydrogen or alkyl, which includes, but is not limited to, C₁₋₂₀ alkyl, C₁₋₁₀ alkyl, or C₁₋₆ alkyl, R^{1'} is —OR' or —N(R^{1'})₂;

[0560] (b) R² is hydrogen or CH₃;

[0561] (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C₁₋₁₀ alkyl, cycloalkyl, —(CH₂)_c(NR³)₂, C₁₋₆ hydroxyalkyl, —CH₂SH, —(CH₂)₂S(O)_dMe, —(CH₂)₃NHC(=NH)NH₂, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, —(CH₂)₂COR³, aryl and aryl C₁₋₃ alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C₁₋₁₀ alkyl, C₁₋₆ alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C₁₋₆ alkyl; (iii) R^{3a} and R^{3b} together are (CH₂)_n so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R² together are (CH₂)_n so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R² together are (CH₂)_n so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where R^{3'} is independently hydrogen or C₁₋₆ alkyl and R^{3''} is —OR' or —N(R^{3'})₂; (vi) R^{3a} is H and R^{3b} is H, CH₃, CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CHCOOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl; or (viii) R^{3a} is CH₃, —CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CHCOOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl and R^{3b} is H, where R^{3'} is independently hydrogen or alkyl, which includes, but is not limited to, C₁₋₂₀ alkyl, C₁₋₁₀ alkyl, or C₁₋₆ alkyl, R^{3''} is —OR' or N(R^{3'})₂;

[0562] (d) R⁴ is hydrogen, C₁₋₁₀ alkyl, C₁₋₁₀ alkyl optionally substituted with a lower alkyl, alkoxy or halogen, C₁₋₁₀ haloalkyl, C₁₋₁₀ cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, di(lower alkyl)amino-lower alkyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;

[0563] (e) R⁵ is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., —(CH₂)_pOH, where p is 1-6, including hydroxyl methyl (CH₂OH), CH₂F, N₃, CH₂CN, CH₂NH₂, CH₂NHCH₃, CH₂N(CH₃)₂, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I with the provisos that when X is OH, base is cytosine and R⁶ is H, R⁵ cannot be N₃ and when X is OH, R⁶ is CH₃ or CH₂F and B is a purine base, R⁵ cannot be H;

[0564] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

[0565] (g) X is H, OH, F, OMe, halogen, NH₂, or N₃;

[0566] (h) Y is OH, H, C₁₋₄ alkyl, C₂₋₄ alkenyl, C₂₋₄ alkynyl, vinyl, N₃, CN, Cl, Br, F, I, NO₂, OC(O)O(C₁₋₄ alkyl), OC(O)O(C₁₋₄ alkyl), OC(O)O(C₂₋₄ alkynyl), OC(O)O(C₂₋₄ alkenyl), OC₁₋₁₀ haloalkyl, O(aminoacyl), O(C₁₋₁₀ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₄ alkyl), S(C₂₋₄ alkynyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl) SO(C₁₋₄ alkyl), SO(C₂₋₄ alkynyl), SO(C₂₋₄ alkenyl), SO₂(C₁₋₄ acyl), SO₂(C₁₋₄ alkyl) SO₂(C₁₋₄ alkynyl), SO₂(C₂₋₄ alkenyl), OS(O)₂(C₁₋₄ acyl), OS(O)₂(C₁₋₄ alkyl), OS(O)₂(C₂₋₄ alkenyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkenyl), NH(C₂₋₄ alkynyl), NH(C₁₋₄ acyl) N(C₁₋₄ alkyl)₂, N(C₁₋₁₈ acyl)₂, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N₃, CN, one to three halogen (Cl, Br, F, I), NO₂, C(O)O(C₁₋₄ alkyl), C(O)O(C₁₋₄ alkyl), C(O)O(C₂₋₄ alkynyl), C(O)O

(CO₂₋₄ alkenyl), O(C₁₋₄ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₄ alkyl), S(C₂₋₄ alkynyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl) SO(C₁₋₄ alkyl), SO(C₁₋₄ alkynyl), SO(C₂₋₄ alkenyl), SO₂(C₁₋₄ acyl), SO₂(C₁₋₄ alkyl), SO₂(C₂₋₄ alkynyl), SO₂(C₂₋₄ alkenyl), OS(O)₂(C₁₋₄ acyl), OS(O)₂(C₁₋₄ alkyl), OS(O)₂(C₂₋₄ alkenyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkenyl), NH(C₂₋₄ alkynyl), NH(C₁₋₄ acyl), N(C₁₋₄ alkyl)₂, N(C₁₋₄ acyl)₂;

[0567] (i) R¹⁰ is H, F, Cl, Br, I, OH, OR', SH, SR', NH₂, NHR', NR'₂, lower alkyl of C₁-C₆, halogenated (F, Cl, Br, I) lower alkyl of C₁-C₆, lower alkenyl of C₂-C₆, halogenated (F, Cl, Br, I) lower alkenyl of C₂-C₆, lower alkynyl of C₂-C₆, such as C≡CH, halogenated (F, Cl, Br, I) lower alkynyl of C₂-C₆ lower alkoxy of C₁-C₆, halogenated (F, Cl, Br, I) lower alkoxy of C₁-C₆, CO₂H, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=OCHCO₂R'.

[0568] wherein R' is an optionally substituted alkyl, which includes, but is not limited to, an optionally substituted C₁₋₂₀ alkyl, an optionally substituted C₁₋₁₀ alkyl, an optionally substituted lower alkyl; an optionally substituted cycloalkyl; an optionally substituted alkynyl of C₂-C₆, an optionally substituted lower alkenyl of C₂-C₆, or optionally substituted acyl, which includes but is not limited to C(O) alkyl, C(O)(C₁₋₂₀ alkyl), C(O)(C₁₋₁₀ alkyl), or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0569] (j) Z is N or CR¹², and

[0570] R¹² is H, halogen (including F, Cl, Br, I), OH, OR', SH, SR', NH₂, NHR', NR'₂, NO₂ lower alkyl of C₁-C₆, halogenated (F, Cl, Br, I) lower alkyl of C₁-C₆, lower alkenyl of C₂-C₆, halogenated (F, Cl, Br, I) lower alkenyl of C₂-C₆, lower alkynyl of C₂-C₆, halogenated (F, Cl, Br, I) lower alkynyl of C₂-C₆, lower alkoxy of C₁-C₆, halogenated (F, Cl, Br, I) lower alkoxy of C₁-C₆, CO₂H, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'; with the proviso that R¹² is not a: (i) —C≡C—H, (ii) —C=CR₂, or (iii) —NO₂.

[0571] A second aspect of the fourth embodiment is directed to a compound represented by formula I-7

[0572] wherein

[0573] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromino-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0574] (b) R² is hydrogen or CH₃;

[0575] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl;

[0576] (d) R⁴ is hydrogen, CH₃, Et, 'Pr, 'Pr, 'Bu, 2-butyl, 'Bu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0577] (e) R⁵ is H, CN, CH₃, OCH₃, CH₂OH, CH₂F, halogen, including F, Cl, Br, or I, with the provisos that when X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H.

[0578] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

[0579] (g) X is H, OH, F, OCH₃, halogen, NH₂, or N₃;

[0580] (h) Y is OH, H, CH₃, vinyl, NH₂, N₃, CN, Cl, Br, F, I, OC(O)CH₃, OCH₃;

[0581] (i) R¹⁰ is H, F, Br, I, OH, OR', NH₂, NHR', NR'₂, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R';

[0582] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0583] (j) Z is N or CR¹²; and

[0584] R¹² is H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'; with the proviso that R¹² is not a: (i) —C≡C—H, (ii) —C=CH₂, or (iii) —NO₂.

[0585] A third aspect of the third embodiment is directed to a compound represented by formula I-7

[0586] wherein

[0587] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0588] (b) R² is hydrogen or CH₃;

[0589] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0590] (d) R⁴ is hydrogen, CH₃, Et, ⁱPr, ⁿPr, ⁿBu, 2-butyl, ^tBu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0591] (e) R⁵ is H, CN, CH₂F, F, Cl, Br, or I; with the proviso that X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H;

[0592] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0593] (g) X is H, OH, F, OCH₃, F, Cl, Br, I, NH₂ or N₃;

[0594] (h) Y is H, OH, CH₃, F, Cl, Br, I, NH₂ or N₃, OCH₃, or OC(O)CH₃;

[0595] (i) R¹⁰ is H, F, Br, I, OH, OR', NH₂, NHR', NR'₂, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R';

[0596] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0597] (j) Z is N or CR¹²; and

[0598] R¹² is H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'; with the proviso that R¹² is not a: (i) —C≡C—H, (ii) —C=CH₂, or (iii) —NO₂.

[0599] A fourth aspect of the fourth embodiment is directed to a compound represented by formula I-7

[0600] wherein

[0601] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0602] (b) R² is hydrogen;

[0603] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0604] (d) R⁴ is hydrogen, CH₃, Et, ⁱPr, ⁿPr, ⁿBu, 2-butyl, ^tBu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0605] (e) R⁵ is H, with the provisos that when X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H;

[0606] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0607] (g) X is H, OH, OCH₃, F, NH₂ or N₃;

[0608] (h) Y is OH, NH₂, OCH₃, or OC(O)CH₃;

[0609] (i) R¹⁰ and R¹¹ H, F, Br, I, OH, OR', NH₂, NHR', NR'₂, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R';

[0610] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of RN'₂, each R' comprise at least one C atom that are joined to form a heterocyclic comppnnsng at least two carbon atoms; and

[0611] (j) Z is N or CR¹²; and

[0612] R¹² is an H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'; with the proviso that R¹² is not a: (i) —C≡C—H, (ii) —C=CH₂, or (iii) —NO₂.

[0613] A flth aspect of the fourth embodiment is directed to a compound represented by formula I-7

[0614] wherein

[0615] (a) R¹ is hydrogen, methyl, phenyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl.

[0616] (b) R² is hydrogen;

[0617] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0618] (d) R⁴ is hydrogen, CH₃, Et, ⁱPr, ⁿPr, ⁿBu, 2-butyl, ^tBu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0619] (e) R⁵ is H, with the provisos that when X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H;

[0620] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0621] (g) X is H, OH, OCH₃, F, NH₂ or N;

[0622] (h) Y is OH, NH₂, OCH₃, or OC(O)CH₃;

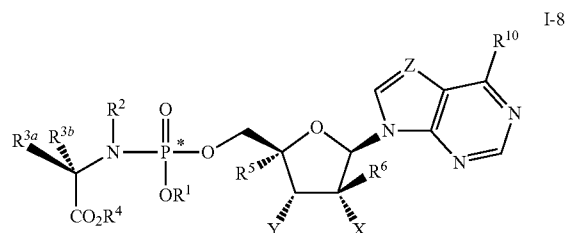
[0623] (i) R¹⁰ is H, F, Br, I, OH, OR', NH₂, NHR', NR'₂, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R';

[0624] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0625] (j) Z is N or CR¹²; and

[0626] R¹² is H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'.

[0627] A sixth aspect of the fourth embodiment is directed to a compound represented by formula I-8



[0628] wherein

[0629] (a) R¹ is hydrogen, n-alkyl; branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally

substituted with at least one of C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-6} alkoxy, F, Cl, Br, I, nitro, cyano, C_{1-6} haloalkyl, $-N(R^{1'})_2$, C_{1-6} acylamino, $-NHSO_2C_{1-6}$ alkyl, $-SO_2N(R^{1'})_2$, $COR^{1'}$, and $-SO_2C_{1-6}$ alkyl; ($R^{1'}$ is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, $R^{1'}$ is $-OR'$ or $-N(R^{1'})_2$);

[0630] (b) R^2 is hydrogen or CH_3 ;

[0631] (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C_{1-10} alkyl, cycloalkyl, $-(CH_2)_c(NR^3)_2$, C_{1-6} hydroxyalkyl, $-CH_2SH$, $-(CH_2)_2S(O)_dMe$, $-(CH_2)_3NHC(=NH)NH_2$, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, $-(CH_2)_cCOR^{3'}$, aryl and aryl C_{1-3} alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C_{1-10} alkyl, C_{1-6} alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C_{1-6} alkyl; (iii) R^{3a} and R^{3b} together are $(CH_2)_n$ so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where R^3 is independently hydrogen or C_{1-6} alkyl and $R^{3'}$ is $-OR'$ or $-N(R^{3'})_2$; (vi) R^{3a} is H and R^{3b} is H, CH_3 , CH_2CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl; or (viii) R^{3a} is CH_3 , $-CH_2CH_3$, $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl and R^{3b} is H, where R^3 is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, $R^{3'}$ is $-OR'$ or $-N(R^{3'})_2$;

[0632] (d) R^4 is hydrogen, C_{1-10} alkyl, C_{1-10} alkyl optionally substituted with a lower alkyl, alkoxy or halogen, C_{1-10} haloalkyl, C_{3-10} cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, di(lower alkylamino-lower alkyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl,

[0633] (e) R^5 is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., $-(CH_2)_pOH$, where p is 1-6, including hydroxyl methyl (CH_2OH), CH_2F , N_3 , CH_2CN , CH_2NH_2 , CH_2NHCH_3 , $CH_2N(CH_3)_2$, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R^6 is H, R^5 cannot be N_3 and when X is OH, R^6 is CH_3 or CH_2F and B is a purine base, R^5 cannot be H;

[0634] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN,

[0635] (g) X is H, OH, F, OMe, halogen, NH_2 , or N_3 ;

[0636] (h) Y is OH, H, C_{1-4} alkyl, C_{2-4} alkenyl, C_{2-4} alkynyl, vinyl, N_3 , CN, Cl, Br, F, I, NO_2 , $OC(O)O(C_{1-4}$ alkyl), $OC(O)O(C_{2-4}$ alkynyl), $OC(O)O(C_{2-4}$ alkenyl), OC_{1-10} haloalkyl, O(aminoacyl), O(C_{1-10} acyl), O(C_{1-4} alkyl), O(C_{2-4} alkenyl), S(C_{1-4} acyl), S(C_{1-4} alkyl), S(C_{2-4} alkynyl), S(C_{2-4} alkenyl), SO(C_{1-4} acyl), SO(C_{1-4} alkyl), SO(C_{2-4} alkynyl), SO(C_{2-4} alkenyl), $SO_2(C_{1-4}$ acyl), $SO_2(C_{1-4}$ alkyl), $SO_2(C_{2-4}$ alkynyl), $SO_2(C_{2-4}$ alkenyl), OS(O_2 (C_{1-4} acyl), OS(O_2 (C_{1-4} alkyl), OS(O_2 (C_{2-4} alkenyl), NH_2 , NH(C_{1-4} alkyl), NH(C_{2-4} alkenyl), NH(C_{2-4} alkynyl), NH(C_{1-4} acyl), N(C_{1-4} alkyl),

N(C_{1-18} acyl) $_2$, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N_3 , CN, one to three halogen (Cl, Br, F, I), NO_2 , C(O)O(C_{1-4} alkyl), C(O)O(C_{1-4} alkyl), C(O)O(C_{2-4} alkynyl), C(O)O(C_{2-4} alkenyl), O(C_{1-4} acyl), O(C_{1-4} alkyl), O(C_{2-4} alkenyl), S(C_{1-4} acyl), S(C_{1-4} alkyl), S(C_{2-4} alkynyl), S(C_{2-4} alkenyl), SO(C_{1-4} acyl), SO(C_{1-4} alkyl), SO(C_{2-4} alkynyl), SO(C_{2-4} alkenyl), $SO_2(C_{1-4}$ acyl), $SO_2(C_{1-4}$ alkyl), $SO_2(C_{2-4}$ alkynyl), $SO_2(C_{2-4}$ alkenyl), OS(O_2 (C_{1-4} acyl), OS(O_2 (C_{1-4} alkyl), OS(O_2 (C_{2-4} alkenyl), NH_2 , NH(C_{1-4} alkyl), NH(C_{2-4} alkenyl), NH(C_{2-4} alkynyl), NH(C_{1-4} acyl), N(C_{1-4} alkyl) $_2$, N(C_{1-4} acyl) $_2$;

[0637] (i) R^{10} is H, F, Br, I, OH, OR', NH_2 , NHR', NR' $_2$, CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$,

[0638] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR' $_2$, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0639] (j) Z is N or CR 12 ; and

[0640] R^{12} is H, halogen (including F, Cl, Br, I), OR', NH_2 , NHR', NR' $_2$, NO_2 , lower alkyl of C_{1-6} , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$; with the proviso that when base is represented by the structure c with R^{11} being hydrogen, R^{12} is not a: (i) $-C\equiv C-H$, (ii) $-C=CH_2$, or (iii) $-NO_2$.

[0641] A seventh aspect of the fourth embodiment is directed to a compound represented by formula I-8

[0642] wherein

[0643] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0644] (b) R^2 is hydrogen or CH_3 ;

[0645] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CHR)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl;

[0646] (d) R^4 is hydrogen, CH_3 , Et, 'Pr, "Pr, "Bu, 2-butyl, 'Bu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0647] (e) R^5 is H, CN, CH_3 , OCH_3 , CH_2OH , CH_2F , halogen, including F, Cl, Br, or I, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0648] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

[0649] (g) X is H, OH, F, OCH_3 , halogen, NH_2 , or N_3 ;

[0650] (h) Y is OH, H, CH_3 , vinyl, NH_2 , N_3 , CN, Cl, Br, F, I, $OC(O)CH_3$, OCH_3 ;

[0651] (i) R^{10} is H, F, Br, I, OH, OR', NH_2 , NHR' NR' $_2$, CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CHCO_2H$, or $CH=CHCO_2R'$,

[0652] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR' $_2$, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0653] (j) Z is N or CR 12 ; and

[0654] R^{12} is an H, halogen (including F, Cl, Br, I), OR', NH_2 , NHR', NR' $_2$, NO_2 , lower alkyl of C_{1-6} , CO_2R' , $CONH_2$, $CONHR'$, $CONR'_2$, $CH=CH_4CO_2H$, or

CH=CHCO₂R'; with the proviso that R¹² is not a: (i) —C≡C—H, (ii) —C=CH₂, or (iii) —NO₂.

[0655] An eighth aspect of the fourth embodiment is directed to a compound represented by formula I-8

[0656] wherein

[0657] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0658] (b) R² is hydrogen or CH₃;

[0659] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0660] (d) R⁴ is hydrogen, CH₃, Et, 'Pr, "Pr, "Bu, 2-butyl, 'Bu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0661] (e) R⁵ is H, CN, CH₂F, F, Cl, Br, or I, with the provisos that when X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H;

[0662] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0663] (g) X is H, OH, F, OCH₃, F, Cl, Br, I, NH₂ or N₃;

[0664] (h) Y is H, OH, CH₃, F, Cl, Br, I, NH₂ or N₃, OCH₃, or OC(O)CH₃;

[0665] (i) R¹⁰ is H, F, Br, I, OH, OR', NH₂, NHR', NR'₂, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R';

[0666] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

[0667] (j) Z is N or CR¹²; and

[0668] R¹² is an H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'; with the proviso that R¹² is not a: (i) —C≡C—H, (ii) —C=CH₂, or (iii) —NO₂.

[0669] A ninth aspect of the fourth embodiment is directed to a compound represented by formula I-8

[0670] wherein

[0671] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0672] (b) R² is hydrogen;

[0673] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0674] (d) R⁴ is hydrogen, CH₃, Et, 'Pr, "Pr, "Bu, 2-butyl, 'Bu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl,

[0675] (e) R⁵ is H, with the provisos that when X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H;

[0676] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0677] (g) X is H, OH, OCH₃, F, NH₂ or N₃;

[0678] (h) Y is OH, NH₂, OCH₃, or OC(O)CH₃;

[0679] (i) R¹⁰ is H, F, Br, I, OH, OR', NH₂, NHR', NR'₂, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R';

[0680] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising, at least two carbon atoms; and

[0681] (j) Z is N or CR¹²; and

[0682] R¹² is an H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'; with the proviso that R¹² is not a: (i) —C≡C—H, (ii) —C=CH₂, or (iii) —NO₂.

[0683] A tenth aspect of the fourth embodiment is directed to a compound represented by formula I-8

[0684] wherein

[0685] (a) R¹ is hydrogen, methyl, phenyl, p-bromo-phenyl, p-chlorophenyl, p-fluorophenyl;

[0686] (b) R² is hydrogen;

[0687] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0688] (d) R⁴ is hydrogen, CH₃, Et, 'Pr, "Pr, "Bu, 2-butyl, 'Bu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0689] (e) R⁵ is H, with the provisos that when X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H;

[0690] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0691] (g) X is H, OH, OCH₃, F, NH₂ or N₃;

[0692] (h) Y is OH, NH₂, OCH₃, or OC(O)CH₃;

[0693] (i) R¹⁰ is H, F, Br, I, OH, OR', NH₂, NHR', NR'₂, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R';

[0694] wherein R' is a lower alkyl, a lower cycloalkyl, or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

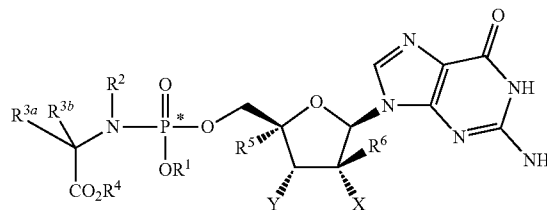
[0695] (j) Z is N or CR¹²; and

[0696] R¹² is an H, halogen (including F, Cl, Br, I), OR', NH₂, NHR', NR'₂, NO₂, lower alkyl of C₁-C₆, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'; with the proviso that R¹² is not a: (i) —C≡C—H, (ii) —C=CH₂, or (iii) —NO₂.

[0697] A fifth embodiment of the invention is directed to a compound represented by formula I in which the base is a structure represented by formula d above, wherein R¹, R², R^{3a}, R^{3b}, R⁴, R⁵, R⁶, X, and Y are defined in the Summary of the Invention section above.

[0698] The first aspect of the fifth embodiment is directed to a compound represented by formula I-9

I-9



[0699] wherein

[0700] (a) R¹ is hydrogen, n-alkyl; branched alkyl; cycloalkyl, or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C₁₋₆ alkyl, C₂₋₆ alkenyl, C₂₋₆ alkynyl, C₁₋₆ alkoxy, F, Cl, Br, I, nitro, cyano, C₁₋₆ haloalkyl,

—N(R¹)₂, C₁₋₆ acylamino, —NHSO₂C₁₋₆ alkyl, —SO₂N(R¹)₂, COR¹, and —SO₂C₁₋₆ alkyl; (R¹ is independently hydrogen or alkyl, which includes, but is not limited to, C₁₋₂₀ alkyl, C₁₋₁₀ alkyl, or C₁₋₆ alkyl, R¹ is —OR' or —N(R¹)₂);

[0701] (b) R² is hydrogen or CH₃;

[0702] (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C₁₋₁₀ alkyl, cycloalkyl, —(CH₂)_c(NR³)₂, C₁₋₆ hydroxyalkyl, —CH₂SH, —(CH₂)₂S(O)_dMe, —(CH₂)₃NHC(=NH)NH₂, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, —(CH₂)_cCOR³, aryl and aryl C₁₋₃ alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C₁₋₁₀ alkyl, C₁₋₆ alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C₁₋₆ alkyl; (iii) R^{3a} and R^{3b} together are (CH₂)₂, so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R² together are (CH₂)_n, so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R² together are (CH₂)_n, so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where R³ is independently hydrogen or C₁₋₆ alkyl and R³ is —OR' or N(R³)₂; (vi) R^{3a} is H and R^{3b} is H, CH₃, CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl; or (viii) R^{3a} is CH₃, —CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl and R^{3b} is H, where R³ is independently hydrogen or alkyl, which includes, but is not limited to, C₁₋₂₀ alkyl, C₁₋₁₀ alkyl, or C₁₋₆ alkyl, R³ is —OR' or —N(R³)₂);

[0703] (d) R⁴ is hydrogen, C₁₋₁₀ alkyl, C₁₋₁₀ alkyl optionally substituted with a lower alkyl, alkoxy or halogen, C₁₋₁₀ haloalkyl, C₃₋₁₀ cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, di(lower alkyl)amino-lower alkyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;

[0704] (e) R⁵ is H, a lower alkyl, CN, vinyl, O-lower alkyl), hydroxyl lower alkyl, i.e., —(CH₂)_pOH, where p is 1-6, including hydroxyl methyl (CH₂OH), CH₂F, N₃, CH₂CN, CH₂NH₂, CH₂NHCH₃, CH₂N(CH₃)₂, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R⁶ is H, R⁵ cannot be Niand when X is OH, R⁶ is CH₃ or CH₂F and B is a purine base, R⁵ cannot be H;

[0705] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

[0706] (g) X is H, OH, F, OMe, halogen, NH₂, or N₃;

[0707] (h) Y is OH, H, C₁₋₄ alkyl, C₂₋₄ alkenyl, C₂₋₄ alkynyl, vinyl, N₃, CN, Cl, Br, F, I, NO₂, OC(O)O(C₁₋₄alkyl), OC(O)O(C₂₋₄ alkyl), OC(O)O(C₂₋₄ alkynyl), OC(O)O(C₂₋₄ alkenyl), OC₁₋₁₀ haloalkyl, O(aminoacyl), O(C₁₋₁₀ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₄ alkyl), S(C₂₋₄ alkynyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl), SO(C₁₋₄ alkyl), SO₂(C₂₋₄ alkynyl), SO(C₂₋₄ alkenyl), SO₂(C₁₋₄ acyl), SO₂(C₁₋₄ alkyl), SO₂(C₂₋₄ alkynyl), SO₂(C₂₋₄ alkenyl), OS(O)₂(C₁₋₄ acyl), OS(O)₂(C₁₋₄ alkyl), OS(O)₂(C₂₋₄ alkenyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkenyl), NH(C₂₋₄ alkynyl), NH(C₁₋₄ acyl), N(C₁₋₄ alkyl)₂, N(C₁₋₁₈ acyl), wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N₃, CN, one to three halogen (Cl, Br, F, I), NO₂, C(O)O(C₁₋

4alkyl), C(O)O(C₁₋₄ alkyl), C(O)O(C₂₋₄ alkynyl), C(O)O(C₂₋₄ alkenyl), O(C₁₋₄ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₄ alkyl), S(C₂₋₄ alkynyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl), SO(C₁₋₄ alkyl), SO(C₂₋₄ alkynyl), SO(C₂₋₄ alkenyl), SO₂(C₁₋₄ acyl), SO₂(C₁₋₄ alkyl), SO₂(C₂₋₄ alkynyl), SO₂(C₂₋₄ alkenyl), OS(O)₂(C₁₋₄ acyl), OS(O)₂(C₁₋₄ alkyl), OS(O)₂(C₂₋₄ alkenyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkenyl), NH(C₂₋₄ alkynyl), NH(C₁₋₄ acyl), N(C₁₋₄ alkyl)₂, N(C₁₋₄ acyl)₂.

[0708] A second aspect of the fifth embodiment is directed to a compound represented by formula I-9

[0709] wherein

[0710] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0711] (b) R² is hydrogen or CH₃;

[0712] (c) R^{3a} is H and R_{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl;

[0713] (d) R⁴ is hydrogen, CH₃, Et, 'Pr, 'Pr, 'Bu, 2-butyl, 'Bu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0714] (e) R⁵ is H, CN, CH₃, OCH₃, CH₂OH, CH₂F, halogen, including F, Cl, Br, or I, with the provisos that when X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H.

[0715] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;

[0716] (g) X is H, OH, F, OCH₃, halogen, NH₂, or N₃;

[0717] (h) Y is OH, H, CH₃, vinyl, NH₂, N₃, CN, Cl, Br, F, I, OC(O)CH₃, OCH₃;

[0718] A third aspect of the fifth embodiment is directed to a compound represented by formula I-9

[0719] wherein

[0720] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0721] (b) R² is hydrogen or CH₃;

[0722] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;

[0723] (d) R⁴ is hydrogen, CH₃, Et, 'Pr, 'Pr, 'Bu, 2-butyl, 'Bu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0724] (e) R⁵ is H, CN, CH₂F, F, Cl, Br, or I; with the proviso that X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H;

[0725] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;

[0726] (g) X is H, OH, F, OCH₃, F, Cl, Br, I, NH₂ or N₃;

[0727] (h) Y is H, OH, CH₃, F, Cl, Br, I, NH₂, N₃, OCH₃, or OC(O)CH₃;

[0728] A fourth aspect of the fifth embodiment is directed to a compound represented by formula I-9

[0729] wherein

[0730] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;

[0731] (b) R² is hydrogen;

[0732] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0733] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0734] (e) R^5 is H, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0735] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0736] (g) X is H, OH, OCH_3 , F, NH_2 or N_3 ;

[0737] (h) Y is OH, NH_2 , OCH_3 , or $OC(O)CH_3$;

[0738] A fifth aspect of the fifth embodiment is directed to a compound represented by formula I-9

[0739] wherein

[0740] (a) R^1 is hydrogen, methyl, phenyl, p-bromo-phenyl, p-fluoro-phenyl, p-fluorophenyl;

[0741] (b) R^2 is hydrogen;

[0742] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0743] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl) amino-lower alkyl;

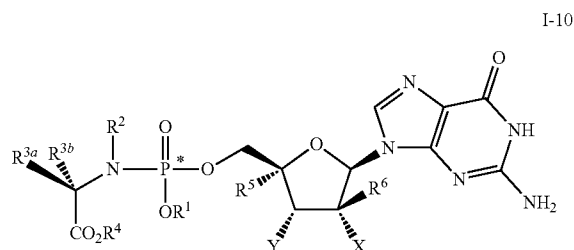
[0744] (e) R^5 is H, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0745] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0746] (g) X is H, OH, OCH_3 , F, NH_2 or N_3 ;

[0747] (h) Y is OH, NH_2 , OCH_3 , or $OC(O)CH_3$;

[0748] A sixth aspect of the fifth embodiment is directed to a compound represented by formula I-10



[0749] wherein

[0750] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, or a substituted or unsubstituted phenyl, where the substituent of the substituted phenyl is at least one of a CH_3 , OCH_3 , F, Cl, Br, I, nitro, cyano, and a CH_3-qX_q , where X is F, Cl, Br, or I, and q is 1-3;

[0751] (b) R^2 is hydrogen or CH_3 ;

[0752] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl or R^{3a} is CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$,

$-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl and R^{3b} is H;

[0753] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0754] (e) R^5 is H, CN, CH_3 , OCH_3 , CH_2OH , CH_2F , N_3 , CH_2CN , CH_2N_3 , CH_2NH_2 , CH_2NHCH_3 , $CH_2N(CH_3)_2$, halogen, including F, Cl, Br, or I, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0755] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

[0756] (g) X is H, OH, F, OCH_3 , Cl, Br, I, NH_2 , or N_3 ;

[0757] (h) Y is OH, H, CH_3 , vinyl, N_3 , CN, Cl, Br, F, I, $OC(O)CH_3$, OCH_3 , NH_2 , $NHCH_3$, $NH(vinyl)$, $NH(acetyl)$, $NH(C(O)CH_3)$, $N(CH_3)_2$, $N(C(O)CH_3)_2$;

[0758] A seventh aspect of the fifth embodiment is directed to a compound represented by formula I-10

[0759] wherein

[0760] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chlorophenyl, p-fluorophenyl;

[0761] (b) R^2 is hydrogen or CH_3 ;

[0762] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl;

[0763] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;

[0764] (e) R^5 is H, CN, CH_3 , OCH_3 , CH_2OH , CH_2F , halogen, including F, Cl, Br, or I, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0765] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

[0766] (g) X is H, OH, F, OCH_3 , Cl, Br, I, NH_2 , or N_3 ; and

[0767] (h) Y is OH, H, CH_3 , vinyl, NH_2 , N_3 , CN, Cl, Br, F, I, $OC(O)CH_3$, OCH_3 .

[0768] An eighth aspect of the fifth embodiment is directed to a compound represented by formula I-10

[0769] wherein

[0770] (a) R^1 is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, p-tolyl, p-bromo-phenyl, p-chlorophenyl, p-fluorophenyl;

[0771] (b) R^2 is hydrogen or CH_3 ;

[0772] (c) R^{3a} is H and R^{3b} is H, CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , or lower cycloalkyl;

[0773] (d) R^4 is hydrogen, CH_3 , Et, iPr , nPr , nBu , 2-butyl, tBu , benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl) amino-lower alkyl;

[0774] (e) R^5 is H, CN, CH_2F , F, Cl, Br, or I, with the provisos that when X is OH, R^6 is CH_3 or CH_2F , R^5 cannot be H;

[0775] (f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , or F;

[0776] (g) X is H, OH, F, OCH₃, F, Cl, Br, I, NH₂ or N₃;
 [0777] (h) Y is H, OH, CH₃, F, Cl, Br, I, NH₂ or N₃, OCH₃, or OC(O)CH₃;
 [0778] A ninth aspect of the fifth embodiment is directed to a compound represented by formula I-10
 [0779] wherein
 [0780] (a) R¹ is hydrogen, methyl, ethyl, n-propyl, i-propyl, phenyl, tolyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;
 [0781] (b) R² is hydrogen;
 [0782] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, or lower cycloalkyl;
 [0783] (d) R⁴ is hydrogen, CH₃, Et, ^tPr, ⁿPr, ⁱBu, 2-butyl, ^tBu benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;
 [0784] (e) R⁵ is H, with the provisos that when X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H;
 [0785] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;
 [0786] (g) X is H, OH, OCH₃, F, NH₂ or N₃;
 [0787] (h) Y is OH, NH₂, OCH₃, or OC(O)CH₃;
 [0788] A tenth aspect of the fifth embodiment is directed to a compound represented by formula I-10
 [0789] wherein
 [0790] (a) R¹ is hydrogen, methyl, phenyl, p-bromo-phenyl, p-chloro-phenyl, p-fluorophenyl;
 [0791] (b) R² is hydrogen;
 [0792] (c) R^{3a} is H and R^{3b} is H, CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CR₂CH₃, CH₂Ph, or lower cycloalkyl;
 [0793] (d) R⁴ is hydrogen, CH₃, Et, ^tPr, ⁿPr, ⁱBu, 2-butyl, ^tBu, benzyl, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, N-methyl-aziridin-2-yl, N-methyl-azetidin-3-yl, N-methyl-pyrrolidin-3-yl, N-methyl-pyrrolidin-4-yl, N-methyl-piperidin-4-yl, lower haloalkyl, or di(lower alkyl)amino-lower alkyl;
 [0794] (e) R⁵ is H, with the provisos that when X is OH, R⁶ is CH₃ or CH₂F, R⁵ cannot be H;
 [0795] (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, or F;
 [0796] (g) X is H, OH, OCH₃, F, NH₂ or N₃;
 [0797] (h) Y is OH, NH₂, OCH₃, or OC(O)CH₃.
 [0798] The following tables contain numeric identifiers associated with various substituent designators that should be viewed in light of the accompanying structure. These structures are contemplated species of the various aspects of the disclosed embodiments and are not intended to be limiting on full breadth of the contemplated compound represented by the structure of formula I. However, it is contemplated that any one of the exemplified nucleoside bases can be used in combination with any one of contemplated species that specify a particular combination of R¹, R², R^{3a}, R^{3b}, R⁴, R⁵, R⁶, X, and Y. In each of the presented tables, the phosphoramidate substituent containing the substituents R^{3a} and R^{3b} are depicted without reference to stereochemical structure (cf. structures I-1, I-3, I-5, I-7, and I-9 above). It is contemplated that the compounds recited below embody compounds in which R^{3a} projects toward the viewer while R^{3b} projects away from the viewer (cf. structures I-2, I-4, I-6, I-8, and I-10). Moreover, it is contemplated that the compounds recited below also embody compounds in which R^{3a} projects away from the viewer while R^{3b} projects towards the viewer. Not

meant to be limiting, however, it is contemplated that preferred compounds are those in which R^{3a} projects towards the viewer and R^{3b} projects away from the viewer such that the natural L-amino acid (S)-configuration is presented. Additionally, the inventors recognize that the phosphorus atom of the phosphoramidate moiety is another source of chirality. Although the structures below do not specifically depict chirality at phosphorus, the inventors recognize that stereochemical configurations are possible such that in a staggered (or zig-zag) line structure the oxo-substituent projects towards the viewer while the OR¹ substituent projects away from the viewer, and vice versa, i.e., where the Cahn-Ingold-Prelog stereochemical designation of phosphorous is either R or S. Therefore, the structures below include all possible stereochemical configurations possible for phosphorus.

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Dosage, Administration, and Use

[0799] A sixth embodiment of the present invention is directed to a composition for the treatment of any of the viral agents disclosed herein said composition comprising a pharmaceutically acceptable medium selected from among an excipient, carrier, diluent, and equivalent medium and a compound, that is intended to include its salts (acid or basic addition salts), hydrates, solvates, and crystalline forms can be obtained, represented by formula I.

[0800] It is contemplated that the formulation of the sixth embodiment can contain any of the compounds contemplated in any of the aspects of the first, second, third, fourth, and fifth embodiments or those specifically recited in the tables above or exemplified herein, either alone or in combination with another compound of the present invention.

[0801] The compounds of the present invention may be formulated in a wide variety of oral administration dosage forms and carriers. Oral administration can be in the form of tablets, coated tablets, hard and soft gelatin capsules, solutions, emulsions, syrups, or suspensions. Compounds of the present invention are efficacious when administered by suppository administration, among other routes of administration. The most convenient manner of administration is generally oral using a convenient daily dosing regimen which can be adjusted according to the severity of the disease and the patient's response to the antiviral medication.

[0802] A compound or compounds of the present invention, as well as their pharmaceutically acceptable salts, together with one or more conventional excipients, carriers, or diluents, may be placed into the form of pharmaceutical compositions and unit dosages. The pharmaceutical compositions and unit dosage forms may be comprised of conventional ingredients in conventional proportions, with or without additional active compounds and the unit dosage forms may contain any suitable effective amount of the active ingredient commensurate with the intended daily dosage range to be employed. The pharmaceutical compositions may be employed as solids, such as tablets or filled capsules, semi-solids, powders, sustained release formulations, or liquids such as suspensions, emulsions, or filled capsules for oral use; or in the form of suppositories for rectal or vaginal administration. A typical preparation will contain from about 5% to about 95% active compound or compounds (w/w). The term "preparation" or "dosage form" is intended to include both solid and liquid formulations of the active compound and one skilled in the art will appreciate that an active ingredient can exist in different preparations depending on the desired dose and pharmacokinetic parameters.

[0803] The term "excipient" as used herein refers to a compound that is used to prepare a pharmaceutical composition, and is generally safe, non-toxic and neither biologically nor otherwise undesirable, and includes excipients that are acceptable for veterinary use as well as human pharmaceutical use. The compounds of this invention can be administered alone but will generally be administered in admixture with

one or more suitable pharmaceutical excipients, diluents or carriers selected with regard to the intended route of administration and standard pharmaceutical practice.

[0804] A “pharmaceutically acceptable salt” form of an active ingredient may also initially confer a desirable pharmacokinetic property on the active ingredient which were absent in the non-salt form, and may even positively affect the pharmacodynamics of the active ingredient with respect to its therapeutic activity in the body. The phrase “pharmaceutically acceptable salt” of a compound as used herein means a salt that is pharmaceutically acceptable and that possesses the desired pharmacological activity of the parent compound. Such salts include: (1) acid addition salts, formed with inorganic acids such as hydrochloric acid, hydrobromic acid, sulfuric acid, nitric acid, phosphoric acid, and the like; or formed with organic acids such as glycolic acid, pyruvic acid, lactic acid, malonic acid, malic acid, inaleic acid, fumaric acid, tartaric acid, citric acid, 3-(4-hydroxybenzoyl)benzoic acid, cinnamic acid, mandelic acid, methanesulfonic acid, ethanesulfonic acid, 1,2-ethane-disulfonic acid, 2-hydroxy-ethanesulfonic acid, benzenesulfonic acid, 4-chlorobenzene-sulfonic acid, 2-naphthalenesulfonic acid, 4-toluenesulfonic acid, camphorsulfonic acid, lauryl sulfuric acid, gluconic acid, glutamic acid, salicylic acid, muconic acid, and the like or (2) basic addition salts formed with the conjugate bases of any of the inorganic acids listed above, wherein the conjugate bases comprise a cationic component selected from among Na^+ , K^+ , Mg^{2+} , Ca^{2+} , and NH_4^+ , in which R^- is a C_{1-3} alkyl and g is a number selected from among 0, 1, 2, 3, or 4. It should be understood that all references to pharmaceutically acceptable salts include solvent addition forms (solvates) or crystal forms (polymorphs) as defined herein, of the same acid addition salt.

[0805] Solid form preparations include powders, tablets, pills, capsules, suppositories, and dispersible granules. A solid carrier may be one or more substances which may also act as diluents, flavoring agents, solubilizers, lubricants, suspending agents, binders, preservatives, tablet disintegrating agents, or an encapsulating material. In powders, the carrier generally is a finely divided solid which is a mixture with the finely divided active component. In tablets, the active component generally is mixed with the carrier having the necessary binding capacity in suitable proportions and compacted in the shape and size desired. Suitable carriers include but are not limited to magnesium carbonate, magnesium stearate, talc, sugar, lactose, pectin, dextrin, starch, gelatin, tragacanth, methylcellulose, sodium carboxymethylcellulose, a low melting wax, cocoa butter, and the like. Solid form preparations may contain, in addition to the active component, colorants, flavors, stabilizers, buffers, artificial and natural sweeteners, dispersants, thickeners, solubilizing agents, and the like.

[0806] Liquid formulations also are suitable for oral administration include liquid formulation including emulsions, syrups, elixirs and aqueous suspensions. These include solid form preparations which are intended to be converted to liquid form preparations shortly before use. Emulsions may be prepared in solutions, for example, in aqueous propylene glycol solutions or may contain emulsifying agents such as lecithin, sorbitan monooleate, or acacia. Aqueous suspensions can be prepared by dispersing the finely divided active component in water with viscous materials such as natural or synthetic gums, resins, methylcellulose, sodium carboxymethylcellulose, and other well known suspending agents.

[0807] The compounds of the present invention may be formulated for administration as suppositories. A low melting wax, such as a mixture of fatty acid glycerides or cocoa butter is first melted and the active component is dispersed homogeneously, for example, by stirring. The molten homogeneous mixture is then poured into convenient sized molds, allowed to cool, and to solidify.

[0808] The compounds of the present invention may be formulated for vaginal administration. Pessaries, tampons, creams, gels, pastes, foams or sprays containing in addition to the active ingredient such carriers as are known in the art to be appropriate.

[0809] Suitable formulations along with pharmaceutical carriers diluent and excipients are described in *Remington: The Science and Practice of Pharmacy* 1995, edited by E. W. Martin, Mack Publishing Company, 19th edition, Easton, Pa., which is hereby incorporated by reference. The compounds of the present invention can also be encapsulated in liposomes, such as those disclosed in U.S. Pat. Nos. 6,180,134, 5,192,549, 5,376,380, 6,060,080, 6,132,763, each of which is incorporated by reference. A skilled formulation scientist may modify the formulations within the teachings of the specification to provide numerous formulations for a particular route of administration without rendering the compositions of the present invention unstable or compromising their therapeutic activity.

[0810] The modification of the present compounds to render them more soluble in water or other vehicle, for example, may be easily accomplished by minor modifications (e.g., salt formulation), which are well within the ordinary skill in the art. It is also well within the ordinary skill of the art to modify the route of administration and dosage regimen of a particular compound in order to manage the pharmacokinetics of the present compounds for maximum beneficial effect in patients.

[0811] A seventh embodiment of the present invention is directed to a use of the compound represented by formula I in the manufacture of a medicament for the treatment of any condition the result of an infection by any one of the following viral agents: hepatitis C virus, West Nile virus, yellow fever virus, dengue virus, rhinovirus, polio virus, hepatitis A virus, bovine viral diarrhea virus and Japanese encephalitis virus.

[0812] The term “medicament” means a substance used in a method of treatment and/or prophylaxis of a subject in need thereof, wherein the substance includes, but is not limited to, a composition, a formulation, a dosage form, and the like, comprising the compound of formula I. It is contemplated that the compound of the use of the compound represented by formula I in the manufacture of a medicament for the treatment of any of the antiviral conditions disclosed herein of the seventh embodiment can be any of the compounds contemplated in any of the aspects of the first, second, third, fourth, fifth embodiments or those specifically recited in the tables above or exemplified herein, either alone or in combination with another compound of the present invention. A medicament includes, but is not limited to, any one of the compositions contemplated by the sixth embodiment of the present invention.

[0813] A eighth embodiment of the present invention is directed to a method of treatment and/or prophylaxis in a subject in need thereof said method comprises administering a therapeutically effective amount of the compound represented by formula I to the subject.

[0814] A first aspect of the eighth embodiment is directed to a method of treatment and/or prophylaxis in a subject in need thereof said method comprises administering a therapeutically effective of at least two or more different compounds falling within the scope of the compound represented by formula I to the subject.

[0815] A second aspect of the eighth embodiment is directed to a method of treatment and/or prophylaxis in a subject in need thereof said method comprises alternatively or concurrently administering a therapeutically effective of at least two compounds falling within the scope of the compound represented by formula I to the subject.

[0816] It is intended that a subject in need thereof is one that has any condition the result of an infection by any of the viral agents disclosed herein, which includes, but is not limited to, hepatitis C virus, West Nile virus, yellow fever virus, dengue virus, rhinovirus, polio virus, hepatitis A virus, bovine viral diarrhea virus or Japanese encephalitis virus, flaviviridae viruses or pestiviruses or hepaciviruses or a viral agent causing symptoms equivalent or comparable to any of the above-listed viruses.

[0817] The term “subject” means a mammal, which includes, but is not limited to, cattle, pigs, sheep, chicken, turkey, buffalo, llama, ostrich, dogs, cats, and humans, preferably the subject is a human. It is contemplated that in the method of treating a subject thereof of the sixth embodiment can be any of the compounds contemplated in any of the aspects of the first, second, and third embodiments or those specifically recited in the tables above, either alone or in combination with another compound of the present invention.

[0818] The term “therapeutically effective amount” as used herein means an amount required to reduce symptoms of the disease in an individual. The dose will be adjusted to the individual requirements in each particular case. That dosage can vary within wide limits depending upon numerous factors such as the severity of the disease to be treated, the age and general health condition of the patient, other medicaments with which the patient is being treated, the route and form of administration and the preferences and experience of the medical practitioner involved. For oral administration, a daily dosage of between about 0.1 and about 10 g, including all values in between, such as 0.25, 0.5, 0.75, 1, 1.5, 2, 2.5, 3, 3.5, 4, 4.5, 5, 5.5, 6, 6.5, 7, 7.5, 8, 8.5, 9, and 9.5, per day should be appropriate in monotherapy and/or in combination therapy. A preferred daily dosage is between about 0.5 and about 7.5 g per day, more preferred 1.5 and about 6.0 g per day. Generally, treatment is initiated with a large initial “loading dose” to rapidly reduce or eliminate the virus following by a decreasing the dose to a level sufficient to prevent resurgence of the infection. One of ordinary skill in treating diseases described herein will be able, without undue experimentation and in reliance on personal knowledge, experience and the disclosures of this application, to ascertain a therapeutically effective amount of the compounds of the present invention for a given disease and patient.

[0819] Therapeutic efficacy can be ascertained from tests of liver function including, but not limited to protein levels such as serum proteins (e.g., albumin, clotting factors, alkaline phosphatase, aminotransferases (e.g., alanine transaminase, aspartate transaminase), 5'-nucleosidase, γ -glutamyl-transpeptidase, etc.), synthesis of bilirubin, synthesis of cholesterol, and synthesis of bile acids; a liver metabolic function, including, but not limited to, carbohydrate metabolism, amino acid and ammonia metabolism. Alternatively the

therapeutic effectiveness may be monitored by measuring HCV-RNA. The results of these tests will allow the dose to be optimized.

[0820] A third aspect of the eighth embodiment, is directed to a method of treatment and/or prophylaxis in a subject in need thereof said method comprises administering to the subject a therapeutically effective of a compound represented by formula I and a therapeutically effective amount of another antiviral agent; wherein the administration is concurrent or alternative. It is understood that the time between alternative administration can range between 1-24 hours, which includes any sub-range in between including, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, and 23 hours. Examples of “another antiviral agents” include, but are not limited to: HCV NS3 protease inhibitors (see WO 2008010921, WO 2008010921, EP 1881001, WO 2007015824, WO 2007014925, WO 2007014926, WO 20070149221, WO 2007014920, WO 2007014922, US 2005267018, WO 2005095403, WO 2005037214, WO 2004094452, US 2003187018, WO 200364456, WO 2005028502, and WO 2003006490); HCV NS5B Inhibitors (see US 2007275947, US20072759300, WO2007095269, WO 2007092000, WO 2007076034, WO 200702602, US 2005-98125, WO 2006093801, US 2006166964, WO 2006065590, WO 2006065335, US 2006040927, US 2006040890, WO 2006020082, WO 2006012078, WO 2005123087, US 2005154056, US 2004229840, WO 2004065367, WO 2004003138, WO 2004002977, WO 2004002944, WO 2004002940, WO 2004000858, WO 2003105770, WO 2003010141, WO 2002057425, WO 2002057287, WO 2005021568, WO 20040401201, US 20060293306, US 20060194749, US 20060241064, U.S. Pat. No. 6,784,166, WO 2007088148, WO 2007039142, WO 2005103045, WO 2007039145, WO 2004096210, and WO 2003037895); HCV NS4 Inhibitors (see WO 2007070556 and WO 2005067900); HCV NS5a Inhibitors (see US 2006276511, WO 2006120252, WO 2006120251, WO 2006100310, WO 2006035061); Toll-like receptor agonists (see WO 2007093901); and other inhibitors (see WO 2004035571, WO 2004014852, WO 2004014313, WO 2004009020, WO 2003101993, WO 2000006529).

[0821] A fourth aspect of the eighth embodiment, is directed to a method of treatment in a subject in need thereof said method comprises alternatively or concurrently administering a therapeutically effective of a compound represented by formula I and another antiviral agent to the subject. It is understood that the time between alternative administration can range between 1-24 hours, which includes any sub-range in between including, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, and 23 hours.

[0822] A fifth aspect of the eighth embodiment, is directed to a method of treatment and/or prophylaxis in a subject in need thereof said method comprises administering to the subject a therapeutically effective of at least one compound represented by formula I and a therapeutically effective amount of another antiviral agent; wherein the administration is concurrent or alternative. It is understood that the time between alternative administration can range between 1-24 hours, which includes any sub-range in between including, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, and 23 hours.

[0823] A sixth aspect of the eighth embodiment, is directed to a method of treatment in a subject in need thereof said method comprises alternatively or concurrently administer-

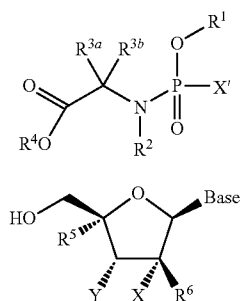
ing a therapeutically effective of at least one compound represented by formula I and another antiviral agent to the subject. It is understood that the time between alternative administration can range between 1-24 hours, which includes any sub-range in between including, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, and 23 hours.

[0824] It is contemplated that the another antiviral agent includes, but is not limited to interferon- α , interferon- β , pegylated interferon- α , ribavirin, levovirin, viramidine, another nucleoside HCV polymerase inhibitor, a HCV non-nucleoside polymerase inhibitor, a HCV protease inhibitor, a HCV helicase inhibitor or a HCV fusion inhibitor. When the active compound or its derivative or salt are administered in combination with another antiviral agent the activity may be increased over the parent compound. When the treatment is combination therapy, such administration may be concurrent or sequential with respect to that of the nucleoside derivatives. "Concurrent administration" as used herein thus includes administration of the agents at the same time or at different times. Administration of two or more agents at the same time can be achieved by a single formulation containing two or more active ingredients or by substantially simultaneous administration of two or more dosage forms with a single active agent.

[0825] It will be understood that references herein to treatment extend to prophylaxis as well as to the treatment of existing conditions. Furthermore, the term "treatment" of a HCV infection, as used herein, also includes treatment or prophylaxis of a disease or a condition associated with or mediated by HCV infection, or the clinical symptoms thereof.

Process for Preparation

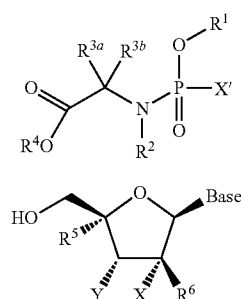
[0826] A ninth embodiment of the present invention is directed to a process for preparing the compound of formula I, which comprises reacting a suitably substituted phosphorochloridate compound 4 with a nucleoside analog 5



[0827] wherein the substituents R^1 , R^{2a} , R^{3a} , R^{3b} , R^4 , R^5 , X , Y , R^6 , and base have their meanings as disclosed in the Detailed Description of the Invention and X' is a leaving group, such as Cl, Br, I, tosylate, mesylate, trifluoroacetate, trifluorosulfonate, pentafluorophenoxide, p-NO_2 -phenoxide, or other commonly used leaving groups as disclosed in *Advanced Organic Chemistry* by March, Fourth Edition. Leaving groups and methods that can be used to effect the formation of a phosphoramidate nucleoside conjugate are found in US 20060142238 and WO 2007095269. Preferably, the leaving group is Cl.

[0828] This reaction is performed in an anhydrous aprotic solvent such tetrahydrofuran, dioxane, or both tetrahydrofuran and dioxane, or any functional equivalent thereof, with tetrahydrofuran being the preferred solvent. The reaction is typically initiated at a temperature range from -78°C . to 40°C . with the preferred reaction temperature being between 0°C . and room temperature. The nucleoside is first stirred with a base (5 to 12 equivalents) such as N-methylimidazole, collidine, pyridine, 2,6-lutidine, 2,6-'Bu-pyridine, etc. a tertiary amine base, such as triethylamine, diisopropylethylamine, etc., or an alkyl Grignard reagent, such as tBuMgCl, tBuMgBr, MeMgCl, MeMgBr, etc. The phosphorochloridate (3-10 equivalents) is dissolved in the reaction solvent and added to the mixture of the nucleoside and base. The reaction is then allowed to stir over a period of time at a temperature between room temperature and 40°C . for a period of 30 min to 24 hr. with the preferred reaction temperature being room temperature and time being 24 hr. The solvent is removed from the reaction mixture and the product is purified by chromatography on silica gel.

[0829] A tenth embodiment of the present invention is directed to a product obtained by a process which comprises reacting a suitably substituted phosphochloridate compound 4 with a nucleoside analog 5



[0830] wherein the substituents R^1 , R^2 , R^{3a} , R^{3b} , R^4 , R^5 , X , Y , R^6 , X' , and base have their meanings as disclosed in the Detailed Description of the Invention.

[0831] This reaction can be performed in an anhydrous aprotic solvent or other suitable solvent, such as tetrahydrofuran, dioxane, or a mixture of tetrahydrofuran and dioxane, with tetrahydrofuran being the preferred solvent. The reaction is typically initiated at a temperature range from -78°C . to 40°C . with the preferred reaction temperature being between 0°C . and room temperature. The nucleoside is first stirred with a base (5 to 12 equivalents) such as N-methylimidazole, a tertiary amine base or tButyl Magnesium Chloride. A phosphorochloridate (3-10 equivalents (or suitable "phosphoro-(leaving group)-date")) is dissolved in the reaction solvent and added to the mixture of the nucleoside and base. The reaction is then allowed to stir over a period of time at a temperature between room temperature and 40°C . for a period of 30 min to 24 hr. with the preferred reaction temperature being room temperature and time being 24 hr. The solvent is removed from the reaction mixture and the product is purified by chromatography on silica gel.

Compounds and Preparation

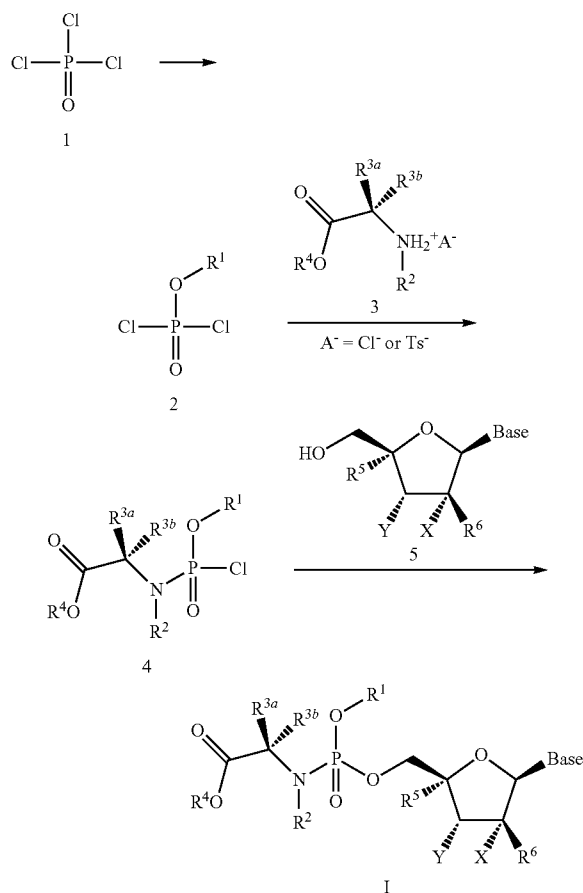
[0832] Phosphoramidate compounds of the present invention can be prepared by condensation of a nucleoside analog

5 with a suitably substituted phosphochloridate compound 4 (Scheme 1). The nucleoside analog is made by conventional procedures disclosed in any one of U.S. Published Application Nos. 2005/0009737, 2006/0199783, 2006/0122146, and 2007/0197463, each of which is incorporated by reference in its entirety.

[0833] Disclosed ¹H-NMR values were recorded on a Varian AS-400 instrument. Mass spectral data were obtained using either a Micromass-Quattro micro API or a Waters Acquity.

[0834] Thus, by way of example only, a suitably substituted phenol can be reacted with phosphorus oxychloride (1) to afford an aryloxy phosphorodichloridate 2 (see Example 1) which is subsequently treated with an acid addition salt of an α -amino acid ester in the presence of TEA to afford an aryloxy phosphorochloridate 4. This aryloxy-amino acid phosphoramidate is reacted with the nucleoside analog to provide the product I (for procedure see, e.g., C. McGuigan et al. *Antiviral Res.* 1992 17:311-321; D. Curley et al. *Antiviral Res.* 1990 14:345-356; McGuigan et al. *Antiviral Chem. Chemother* 1990 1(2):107-113).

Scheme 1



[0835] The preparation of nucleoside phosphoramidates requires reacting an appropriately substituted phosphochloridate with a nucleoside containing a free 5'-hydroxyl moiety. In cases where only one hydroxyl group is present, preparation of the phosphoramidate usually proceeds smoothly when the phosphochloridate is reacted with the desired nucleoside. In cases where the nucleoside contains more than one free hydroxyl group, preparation of the appropriately protected

nucleoside might be required. Silyl, acetonide or other alcohol protecting groups known in the art might be warranted for protection of the sugar moiety. For protection of the nucleoside base, protecting a free amino group may require amidine protection strategy.

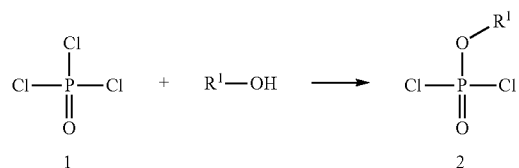
[0836] Condensation of the phosphochloridate can be carried out on the unprotected nucleoside. Since the 5'-OH group of a nucleoside is much less hindered than the 3'-OH group, selective phosphoramidation is possible under carefully controlled conditions. After condensation to form a protected phosphoramidate nucleoside, deprotection to obtain the free phosphoramidate nucleoside can be carried out using standard protocols for nucleic acid chemistry. In many cases, the desired product is readily separated from the starting material using column chromatography on silica gel. The synthetic scheme is summarized in Scheme 1.

[0837] A further understanding of the disclosed embodiments will be appreciated by consideration of the following examples, which are only meant to be illustrative, and not limit the disclosed invention.

Example 1

General Procedure for Preparation of Phosphorodichloridates

[0838]

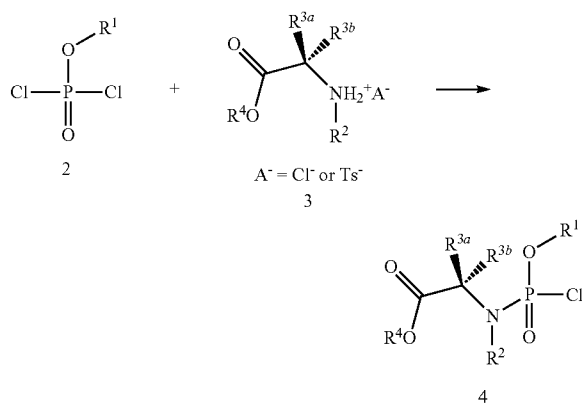


[0839] A solution of the appropriate phenol R^1-OH (1 eq) and triethylamine (1 eq.) in anhydrous ether was added dropwise to a stirred solution of phosphoryl trichloride 1 (1 eq) at 0° C. over a period of 3 hours under nitrogen. Then the temperature was warmed to room temperature, and the reaction was stirred overnight. The triethylamine salt was quickly removed with suction filtration and the filtrate concentrated in vacuo to dryness to afford 2 as an oil which was used without further purification.

Example 2

General Procedure for Preparation of Phosphorochloridates

[0840]

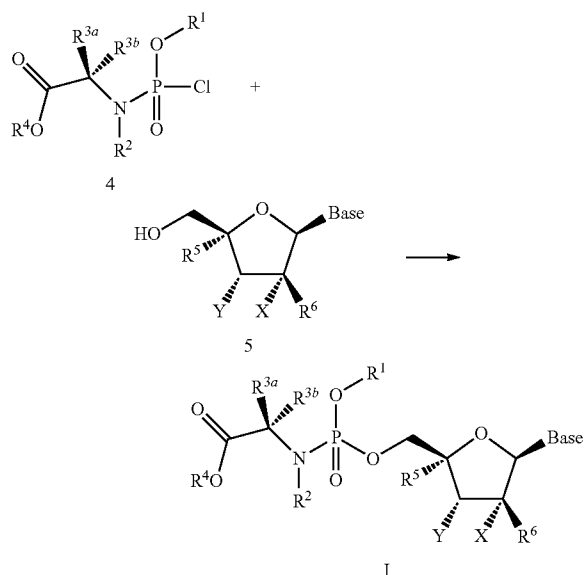


[0841] A solution of triethylamine (2 eq) in anhydrous dichloromethane was added dropwise to a solution of aryloxy-phosphodichloridate 2 (1 eq) and the appropriate amino ester 3 (1 eq) in anhydrous dichloromethane with vigorous stirring at -78°C . over a period of 30 to 120 minutes. Then the reaction temperature was allowed to warm to room temperature and stirred over night. Solvent was removed. The residue was washed with ethyl ether and filtered, the filtrate was dried over reduced pressure to give 4.

Example 3

General Procedures for Nucleoside Phosphoramidate Derivatives

[0842]

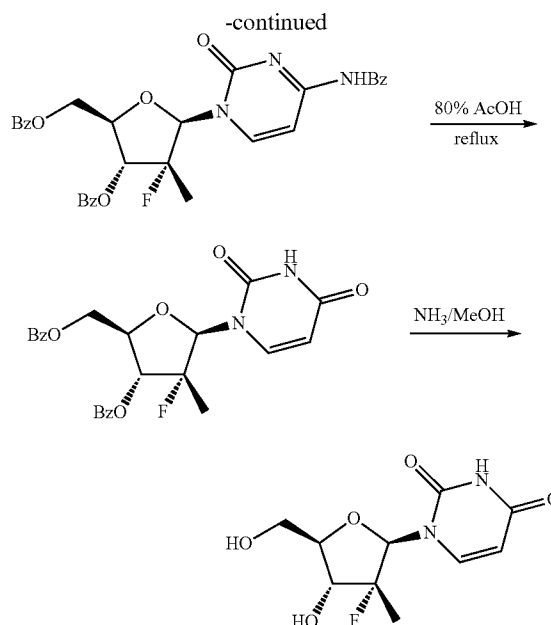
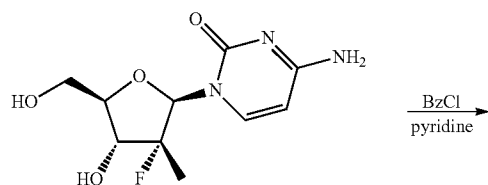


[0843] A solution of the appropriate phosphorochloridate 4 (6.5 equivalents) in anhydrous tetrahydrofuran (THF) was added to a mixture of nucleoside 5 (1 equivalent) and N-methylimidazole (8 equivalents) in anhydrous THF with vigorous stirring at room temperature and the reaction mixture was stirred overnight. The solvent was removed in vacuo and the crude was purified by column chromatography and/or preparative thin layer chromatography to give I.

Example 4

Preparation of 2'-deoxy-2'-fluoro-2'-C-methyluridine

[0844]



[0845] 2'-Deoxy-2'-fluoro-2'-C-methylcytidine (1.0 g, 1 eq) (Clark, J., et al., J. Med. Chem., 2005, 48, 5504-5508) was dissolved in 10 ml of anhydrous pyridine and concentrated to dryness in vacuo. The resulting syrup was dissolved in 20 ml of anhydrous pyridine under nitrogen and cooled to 0°C . with stirring. The brown solution was treated with benzoyl chloride (1.63 g, 3 eq) dropwise over 10 min. The ice bath was removed and stirring continued for 1.5 h whereby thin-layer chromatography (TLC) showed no remaining starting material. The mixture was quenched by addition of water (0.5 ml) and concentrated to dryness. The residue was dissolved in 50 mL of dichloromethane (DCM) and washed with saturated NaHCO_3 aqueous solution and H_2O . The organic phase was dried over NaSO_4 and filtered, concentrated to dryness to give N^4 , 3',5'-tribenzoyl-2'-Deoxy-2'-fluoro-2'-C-methylcytidine (2.0 g, Yield: 91%).

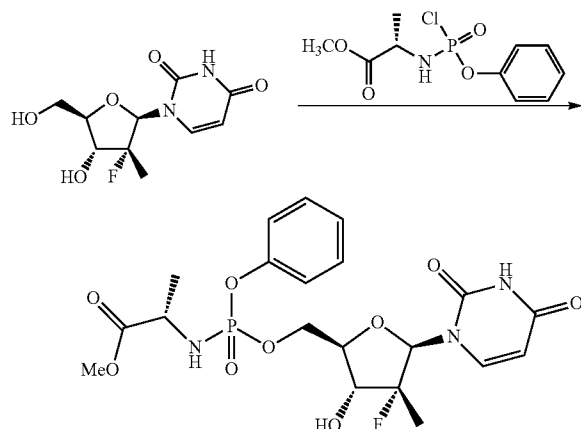
[0846] N^4 ,3',5'-tribenzoyl-2'-Deoxy-2'-fluoro-2'-C-methylcytidine (2.0 g, 1 eq) was refluxed in 80% aqueous AcOH overnight. After cooling and standing at room temperature (15°C .), most of the product precipitated and then was filtered through a sintered funnel. White precipitate was washed with water and co-evaporated with toluene to give a white solid. The filtrate was concentrated and co-evaporated with toluene to give additional product which was washed with water to give a white solid. Combining the two batches of white solid gave 1.50 g of 3',5'-dibenzoyl-2'-Deoxy-2'-fluoro-2'-C-methyluridine (Yield: 91%).

[0847] To a solution of 3',5'-dibenzoyl-2'-Deoxy-2'-fluoro-2'-C-methyluridine (1.5 g, 1 eq) in MeOH (10 mL) was added a solution of saturated ammonia in MeOH (20 mL). The reaction mixture was stirred at 0°C . for 30 min, and then warmed to room temperature slowly. After the reaction mixture was stirred for another 18 hours, the reaction mixture was evaporated under reduced pressure to give the residue, which was purified by column chromatography to afford pure compound 2'-deoxy-2'-fluoro-2'-C-methyluridine (500 mg, Yield: 60 %).

Example 5

Preparation of 2'-Deoxy-2'-fluoro-2'-C-methyluridine
5'-(phenyl methoxy-alanyl phosphate)

[0848]

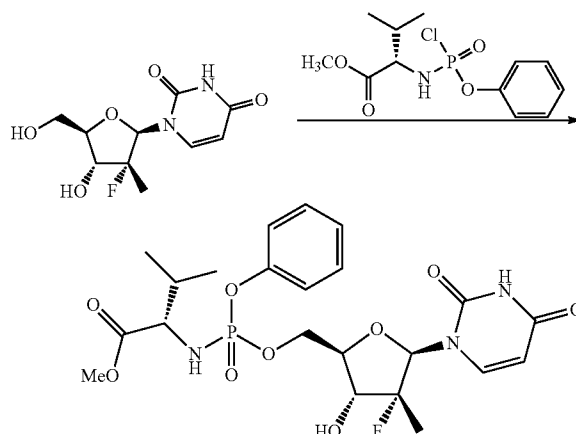


[0849] Phenyl methoxyalanyl phosphorochloridate (1 g, 6.5 eq) dissolved in 3 mL of THF was added to a mixture of 2'-Deoxy-2'-fluoro-2'-C-methyluridine (0.15 g, 1 eq) and N-methylimidazole (0.3 g, 8 eq) in 3 mL THF with vigorous stirring at room temperature, then the reaction was stirred overnight. Solvent was removed by reduced pressure. The resulting crude product was dissolved in methanol purified by prep-HPLC on a YMC 25×30×2 mm column using a water/acetonitrile gradient elution mobile phase. The acetonitrile and water were removed under reduced pressure to give the desired product (50.1 mg, 15.6%). ¹H NMR (DMSO-d₆) δ 1.20-1.27 (m, 6H), 3.58 (d, J=16.0 Hz, 3H), 3.75-3.92 (m, 2H), 4.015-4.379 (m, 2H), 5.54 (t, J=10.2 Hz, 1H), 5.83-5.91 (m, 1H), 6.00-6.16 (m, 1H), 7.18 (d, J=8.0 Hz, 2H), 7.22 (s, 1H), 7.35 (t, J=4.4 Hz, 2H), 7.55 (s, 1H), 11.52 (s, 1H); MS, m/e 502 (M+1)⁺.

Example 6

Preparation of 2'-Deoxy-2'-fluoro-2'-C-methyluridine
5'-(phenyl methoxy-valyl phosphate)

[0850]

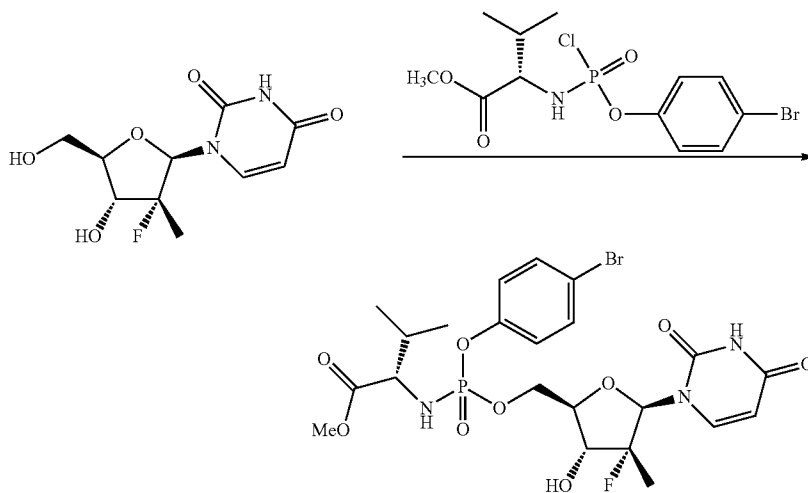


[0851] Phenyl methoxy-valyl phosphorochloridate (0.6 g, 3.6 eq) dissolved in 3 mL of THF was added to a mixture of 2'-Deoxy-2'-fluoro-2'-C-methyluridine (0.15 g, 1 eq) and N-methylimidazole (0.44 g, 9 eq) in 3 mL THF with vigorous stirring at room temperature, then the reaction was stirred overnight. Solvent was removed by reduced pressure. The resulting crude product was dissolved in methanol purified by prep-HPLC on a YMC 25×30×2 mm column using a water/acetonitrile gradient elution mobile phase. The acetonitrile and water were removed under reduced pressure to give the desired product (60 mg, 20.9%). ¹H NMR (DMSO-d₆) δ 0.74-0.947 (m, 6H), 1.20-1.28 (m, 3H), 1.89-1.92 (m, 1H), 3.50-3.54 (m, 1H), 3.58 (d, J=10.4 Hz, 3H), 3.72-3.95 (m, 1H), 4.03-4.05 (m, 1H), 4.23-4.43 (m, 2H), 5.56 (t, J=16.0 Hz, 1H), 5.85-5.92 (m, 1H), 6.01-6.07 (m, 1H), 7.16-7.21 (m, 3H), 7.37 (t, J=8 Hz, 2H), 7.55-7.60 (m, 1H), 11.52 (s, 1H); MS, m/e 530 (M+1)⁺.

Example 7

Preparation of 2'-Deoxy-2'-fluoro-2'-C-methyluridine
5'-(4-bromophenyl methoxy-valyl phosphate)

[0852]

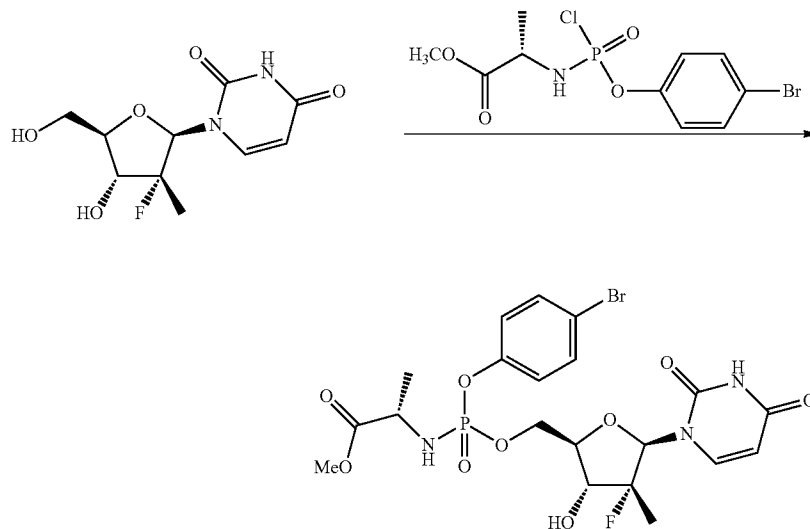


[0853] 4-Bromophenyl methoxy-valyl phosphorochloride (1 g, 3.4 eq) dissolved in 3 mL of THF was added to a mixture of 2'-deoxy-2'-fluoro-2'-C-methyluridine (0.2 g, 1 eq) and N-methylimidazole (0.35 g, 6 eq) in 3 mL THF with vigorous stirring at room temperature, then the reaction was stirred overnight. Solvent was removed by reduced pressure. The resulting crude product was dissolved in methanol purified by prep-HPLC on a YMC 25×30×2 mm column using a water/acetonitrile gradient elution mobile phase. The acetonitrile and water were removed reduced pressure to give the desired product (120 mg, 26%). ¹H NMR (DMSO-d₆) δ 0.72-0.82 (m, 6H), 1.19-1.26 (m, 3H), 1.86-1.92 (m, 1H), 3.48-3.50 (m, 1H), 3.56 (d, J=12.0 Hz, 3H), 3.72-3.89 (m, 1H), 3.96-4.03 (m, 1H), 4.22-4.37 (m, 2H), 5.54-5.60 (m, 1H), 5.85-5.91 (m, 1H), 5.98-6.13 (m, 1H), 7.15 (d, J=8.0 Hz, 2H), 7.49-7.56 (m, 3H), 11.53 (s, 1H); MS, m/e 608 (M+1)³⁰.

Example 8

Preparation of 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-alanyl phosphate)

[0854]

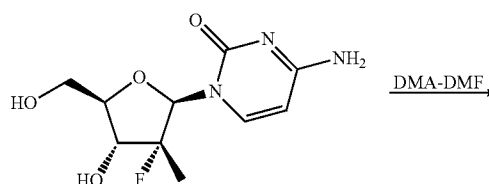


[0855] 4-Bromophenyl methoxy-alanyl phosphorochloride (0.6 g, 5 eq) dissolved in 3 mL of THF was added to a mixture of 2'-deoxy-2'-fluoro-2'-C-methyluridine (0.15 g, 1 eq) and N-methylimidazole (0.3 g, 7.8 eq) in 3 mL THF with vigorous stirring at room temperature, then the reaction was stirred overnight. Solvent was removed by reduced pressure. The resulting crude product was dissolved in methanol purified by prep-HPLC on a YMC 25×30×2 mm column using a water/acetonitrile gradient elution mobile phase. The acetonitrile and water were removed under reduced pressure to give the desired product (40 mg, 12%); ¹H NMR (DMSO-d₆) δ 1.20-1.26 (m, 6H), 3.57 (d, J=2.8 Hz, 3H), 3.84 (s, 1H), 3.97-4.03 (m, 1H), 4.21-4.25 (m, 1H), 4.33-4.37 (m, 2H), 5.54-5.60 (m, 1H), 5.83-5.89 (m, 1H), 5.98-6.19 (m, 1H), 7.16 (t, J=10.2 Hz, 2H), 7.52-7.57 (m, 3H), 11.52 (s, 1H); MS, m/e 580(M+1)⁺.

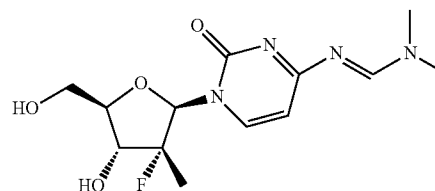
Example 9

Preparation of N⁴-(N,N-dimethylformamidinyl)-2'-deoxy-2'-fluoro-2'-C-methylcytidine

[0856]



-continued

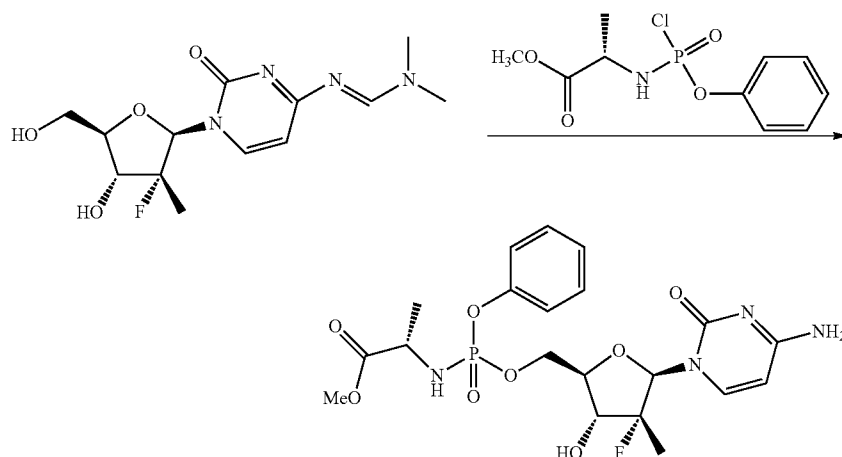


[0857] 2'-Deoxy-2'-fluoro-2'-C-methylcytidine (500 mg, 1.9 mmol) was stirred with dimethylformamide dimethyl acetal in DMF (10 mL). The resulting mixture was stirred at room temperature overnight. After solvent removal the crude product was used for next step without further purification.

Example 10

Preparation of 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(phenyl methoxy-alanyl phosphate)

[0858]



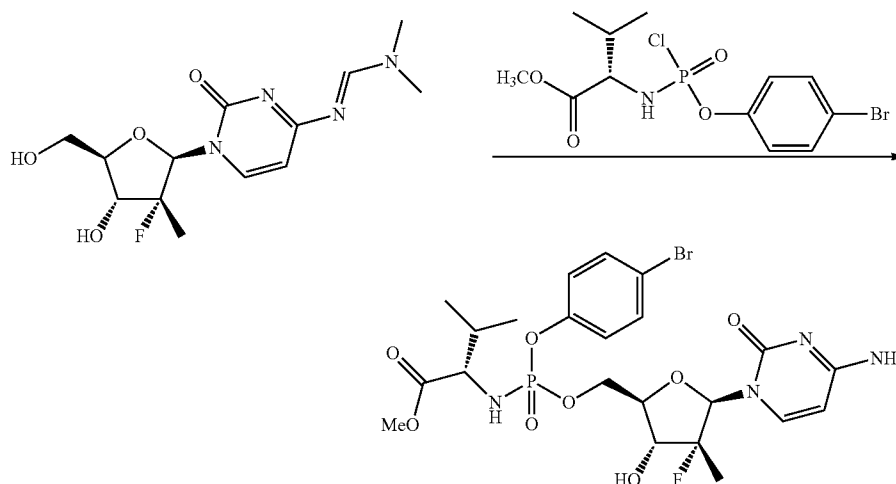
[0859] Phenyl methoxyalanyl phosphorochloridate (0.6 g, 6 eq) dissolved in 3 mL of THF was added to a mixture of N⁴-(N,N-dimethylformamidinyl)-2'-deoxy-2'-fluoro-2'-C-methylcytidine (0.15 g, 1 eq) and N-methylimidazole (0.3 g, 7.8 eq) in 3 mL THF with vigorous stirring at room temperature, then the reaction was stirred overnight. Solvent was removed by reduced pressure. The resulting crude product was dissolved in methanol purified by prep-HPLC on a YMC 25×30×2 mm column using a water/acetonitrile gradient elution mobile phase. The acetonitrile and water were removed under reduced pressure to give the desired product (62 mg, 20.6%). ¹H NMR (DMSO-d₆), δ 1.16 (d, J=23.2 Hz, 3H),

1.22 (d, J=7.2 Hz, 3H), 3.56 (s, 3H), 3.69-3.75 (d, J=25.6 Hz, 1H), 3.82-3.86 (m, 1H), 3.96-3.98 (m, 1H), 4.21-4.34 (m, 2H), 5.68 (d, J=7.2 Hz, 1H), 5.75-5.77 (m, 1H), 6.07-6.16 (m, 1H), 7.15-7.19 (m, 3H), 7.2 (d, J=9.2 Hz, 2H), 7.39 (t, J=7.8 Hz, 2H), 7.48 (d, J=9.2 Hz, 1H); MS, m/e 501(M+1)³⁰.

Example 11

Preparation of 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-4-bromophenyl methoxy-valyl phosphate

[0860]



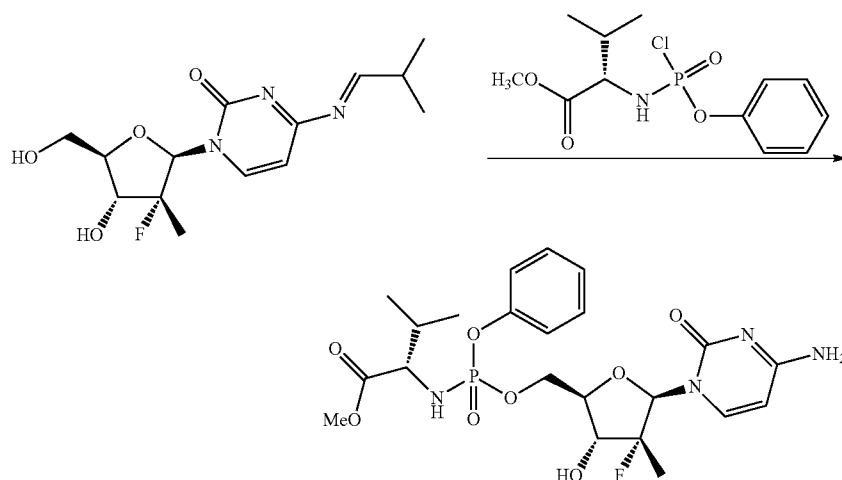
[0861] 4-Bromophenyl methoxy-valyl phosphorochloridate (1.0 g, 3.4 eq.) dissolved in 3 mL of THF was added to a mixture of N⁴-(N,N-dimethylformamidinyl)-2'-deoxy-2'-fluoro-2'-C-methylcytidine (0.2 g, 1 eq.) and N-methylimidazole (0.35 g, 6 eq.) in 3 mL THF with vigorous stirring at room temperature, then the reaction was stirred overnight. Solvent was removed by reduced pressure. The resulting crude product was dissolved in methanol purified by prep-

HPLC on a YMC 25×30×2 mm column using a water/acetonitrile gradient elution mobile phase. The acetonitrile and water were removed under reduced pressure to give the desired product as a white solid (59 mg, 13%); ¹H NMR (DMSO-d₆) δ 0.74-0.83 (m, 6H), 1.12-1.20 (m, 3H), 1.89-1.92 (m, 1H), 3.49-3.51 (m, 1H), 3.55 (s, 3H), 3.59-3.68 (m, 1H), 3.72-3.83 (m, 1H), 4.21-4.39 (m, 2H), 5.70-5.72 (m, 1H), 5.76-5.83 (m, 1H), 6.04-6.16 (m, 1H), 7.15 (d, J=13.0 Hz, 2H), 7.26 (s, 1H), 7.33 (s, 1H), 7.46-7.55 (m, 1H), 7.56 (d, J=4.4 Hz, 2H); MS, m/e 607 (M+1)⁺.

Example 12

Preparation of 2'-deoxy-2'-fluoro-2'-C-methylcytidine 5'-(phenyl methoxy-valyl phosphate)

[0862]

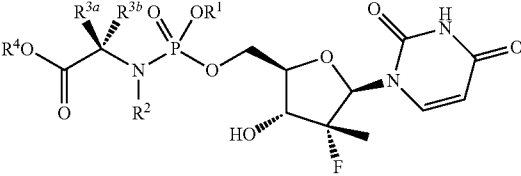


[0863] Phenyl methoxy-valyl phosphorochloridate (0.6 g, 6 eq) dissolved in 3 mL of THF was added to a mixture of N⁴-(N,N-dimethylformamidine)-2'-deoxy-2'-fluoro-2'-C-methylcytidine (0.15 g, 1 eq) and N-methylimidazole (0.3 g, 7.8 eq) in 3 mL THF with vigorous stirring at room temperature, then the reaction was stirred overnight. Solvent was removed by reduced pressure. The resulting crude product was dissolved in methanol purified by prep-HPLC on a YMC 25×30×2 mm column using a water/acetonitrile gradient elution mobile phase. The acetonitrile and water were removed under reduced pressure to give the desired product as a white solid (86 mg, 42.9 %). ¹H NMR (DMSO-d₆) δ 0.72-0.80 (m,

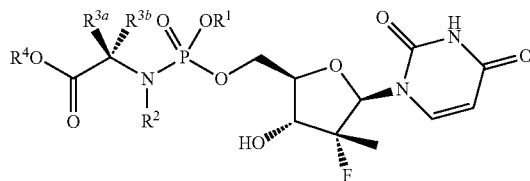
6H), 1.09-1.18 (m, 3H), 1.87-1.92 (m, 1H), 3.47-3.51 (m, 1H), 3.58 (s, 3H), 3.71-3.75 (m, 1H), 3.97 (t, J=11.2 Hz, 1H), 4.22-4.37 (m, 2H), 5.70 (d, J=8.0 Hz, 1H), 5.76-5.84 (m, 1H), 6.01-4.15 (m, 1H), 7.13-7.18 (m, 3H), 7.27 (s, 2H), 7.34 (d, J=4.0 Hz, 2H), 7.46-7.50 (m, 1H); MS, m/e 529 (M+1)⁺.

Examples

[0864] Example numbers 13-54 and 56-66 are prepared using similar procedures described for examples 5-8. The example number, compound identification, and NMR/MS details are shown below:

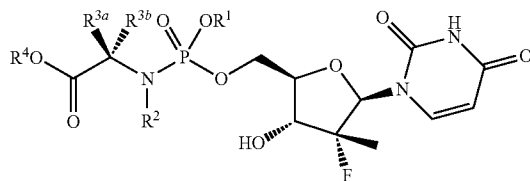
						
Ex.	R ¹	R ²	R ^{3a}	R ^{3b}	R ⁴	NMR/MS
13	Ph	H	H	Me	Et	¹ H NMR(DMSO-d ₆) δ 1.12-1.16 (m, 3H), 1.20-1.28 (m, 6H), 3.70-3.90 (m, 2H), 4.00-4.08 (m, 3H), 4.18-4.45 (m, 2H), 5.52-5.58 (m, 1H), 5.85-5.98 (m, 1H), 6.00-6.20 (m, 2H), 7.16-7.23 (m, 3H), 7.37-7.40 (m, 2H), 7.54-7.60 (m, 1H), 11.54 (s, 1H); MS, m/e 516.1 (M + 1) ⁺

-continued



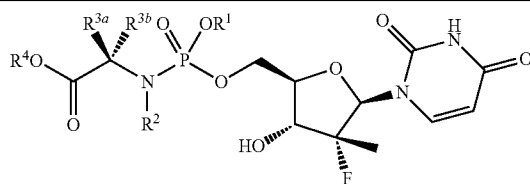
Ex.	R ¹	R ²	R ^{3a}	R ^{3b}	R ⁴	NMR/MS
14	1-Naph	H	H	Me	Bn	1H NMR (DMSO-d ₆) δ 1.18-1.30 (m, 6H), 3.78-4.10 (m, 3H), 4.38-4.49 (m, 2H), 4.99-5.11 (m, 2H), 5.28-5.40 (m, 1H), 5.85-6.10 (m, 2H), 6.30-6.41 (m, 1H), 7.28-7.32 (m, 5H), 7.41-7.60 (m, 5H), 7.73-7.76 (m, 1H), 7.94-8.11 (m, 1H), 8.13-8.15 (m, 1H), 11.50 (s, 1H); MS, m/e 628.4 (M + 1)+
15	Ph	H	H	H	Me	1H NMR (DMSO-d ₆) δ 1.22 (d, J = 22.4 Hz, 3H), 3.59 (s, 3H), 3.63-3.69 (m, 2H), 3.74-3.8 (m, 1H), 4.02 (d, J = 11.2 Hz, 1H), 4.23-4.28 (m, 1H), 4.40-4.43 (m, 1H), 5.57-5.60 (m, 1H), 5.89 (d, J = 6.8 Hz, 1H), 6.00-6.06 (m, 2H), 7.15-7.23 (m, 3H), 7.35-7.39 (m, 2H), 7.52 (d, J = 8 Hz, 1H), 11.52 (s, 1H); MS, m/e 487.97 (M + 1)+
16	2,4-Cl-Ph	H	H	Me	Me	1H NMR (DMSO-d ₆) δ 1.22-1.28 (m, 6H), 3.57-3.60 (m, 3H), 3.84-3.92 (m, 2H), 4.00-4.04 (m, 1H), 4.31-4.44 (m, 2H), 5.54-5.61 (m, 1H), 5.85-6.10 (m, 2H), 6.32-6.43 (m, 1H), 7.44-7.54 (m, 3H), 7.72-7.75 (m, 1H), 11.54 (s, 1H); MS, m/e 570.2 (M + 1)+
17	1-Naph	H	H	Me	Me	1H NMR (DMSO-d ₆) δ 1.15-1.27 (m, 6H), 3.51-3.55 (d, 3H), 3.85-3.96 (m, 2H), 4.00-4.10 (m, 1H), 4.30-4.46 (m, 2H), 5.31-5.39 (m, 1H), 5.89-6.05 (m, 2H), 6.22-6.34 (m, 1H), 7.44-7.60 (m, 5H), 7.73-7.77 (m, 1H), 7.93-7.96 (m, 1H), 8.12-8.14 (m, 1H), 11.50 (s, 1H); MS, m/e 552.1 (M + 1)+
18	Ph	*	H	*	Me	1H NMR (DMSO-d ₆) δ 1.19 (d, J = 22.8 Hz, 3H), 1.69-1.84 (m, 3H), 1.99-2.04 (m, 1H), 3.16-3.21 (m, 2H), 3.58 (s, 3H), 3.63-3.8 (m, 1H), 4.00 (m, 1H), 4.01-4.13 (m, 1H), 4.22-4.25 (m, 1H), 4.5 (d, J = 11.2 Hz, 1H), 5.54 (d, J = 8.0 Hz, 1H), 5.86 (s, 1H), 5.6 (d, J = 19.6 Hz, 1H), 7.15-7.2 (m, 3H), 7.34 (t, J = 8.0 Hz, 2H), 7.51 (d, J = 8.0 Hz, 1H), 11.38 (s, 1H); MS, m/e 527.93 (M + 1)+
19	Ph	H	H	Me	n-Bu	1H NMR (DMSO-d ₆) δ 0.80-0.90 (m, 3H), 1.20-1.35 (m, 8H), 1.48-1.55 (m, 2H), 3.78-3.88 (m, 2H), 3.95-4.08 (m, 3H), 4.22-4.45 (m, 2H), 5.55-5.57 (t, 1H), 5.85-6.18 (m, 3H), 7.14-7.23 (m, 3H), 7.35-7.40 (m, 2H), 7.51-7.60 (d, 1H), 11.50 (s, 1H); MS, m/e 544.2 (M + 1)+
20	Ph	H	H	Me	Bn	1H NMR (DMSO-d ₆) δ 1.20-1.30 (m, 6H), 3.72-4.05 (m, 3H), 4.23-4.27 (m, 1H), 4.32-4.45 (m, 1H), 5.07-5.10 (t, 2H), 5.52-5.56 (t, 1H), 5.86-6.10 (m, 2H), 6.13-6.21 (m, 1H), 7.15-7.21 (m, 3H), 7.29-7.40 (m, 7H), 7.51-7.56 (d, 1H), 11.50 (s, 1H); MS, m/e 578.2 (M + 1)+
21	4-F-Ph	H	H	Me	Me	1H NMR (DMSO-d ₆) δ 1.28-1.34 (m, 6H), 3.65 (d, J = 4 Hz, 3H), 3.85-3.96 (m, 2H), 4.06-4.12 (m, 1H), 4.30-4.34 (m, 1H), 4.40-4.47 (m, 1H), 5.62-5.67 (m, 1H), 5.94-6.01 (m, 1H), 6.09 (d, J = 18.8 Hz, 1H), 6.17-6.26 (m, 1H), 7.27-7.33 (m, 4H), 7.62 (d, J = 7.6 Hz, 1H), 11.61 (s, 1H); MS, m/e 519.94 (M + 1)+
22	4-Cl-Ph	H	H	Me	Me	1H NMR (DMSO-d ₆) δ 1.22-1.28 (m, 6H), 3.58 (d, 2H), 3.70-3.95 (m, 2H), 3.95-4.08 (m, 1H), 4.23-4.45 (m, 2H), 5.55-5.61 (t, 1H), 5.85-6.10 (m, 2H), 6.15-6.23 (m, 1H), 7.20-7.26 (m, 2H), 7.43-7.46 (m, 2H), 7.54-7.57 (d, 1H), 11.50 (s, 1H); MS, m/e 536.1 (M + 1)+
23	3,4-Cl-Ph	H	H	Me	Me	1H NMR (DMSO-d ₆) δ 1.13 (m, 6H), 3.49 (s, 3H), 3.61-3.85 (m, 2H), 3.90-3.93 (m, 1H), 4.16-4.22 (m, 1H), 4.27-4.31 (m, 1H), 5.47-5.52 (m, 1H), 5.82 (d, J = 11.6 Hz, 1H), 5.93 (d, J = 19.2 Hz, 1H), 6.15-6.25 (m, 1H), 7.13 (t, J = 9.6 Hz, 1H), 7.43 (d, J = 12 Hz, 2H), 7.57 (d, J = 6.0 Hz, 1H), 11.43 (s, 1H); MS, m/e 569.85 (M + 1)+
24	Ph	H	H	Me	2-Bu	1H NMR (DMSO-d ₆) δ 0.83 (d, J = 6.8 Hz, 6H), 1.20-1.26 (m, 6H), 1.79-1.86 (m, 1H), 3.73-3.90 (m, 4H), 4.01 (t, J = 11.2 Hz, 1H), 4.21-4.28 (m, 1H), 4.33-4.42 (m, 1H), 5.54 (t, J = 7.6 Hz, 1H), 5.85-5.92 (m, 1H), 5.99-6.13 (m, 2H), 7.19 (t, J = 8 Hz, 3H), 7.36 (t, J = 7.6 Hz, 2H), 7.53 (d, J = 7.6 Hz, 1H), 11.52 (s, 1H); MS, m/e 544.00 (M + 1)+
25	Ph	H	H	Me	i-Pr	1H NMR (DMSO-d ₆) δ 1.13-1.28 (m, 12H), 3.74-3.81 (m, 2H), 3.95-4.08 (m, 1H), 4.20-4.45 (m, 2H), 4.83-4.87 (m, 1H), 5.52-5.58 (m, 1H), 5.84-6.15 (m, 3H), 7.17-7.23 (m, 3H), 7.35-7.39 (m, 2H), 7.54-7.57 (m, 1H), 11.50 (s, 1H); MS, m/e 530.2 (M + 1)+
26	4-MeO-Ph	H	H	Me	n-Bu	1HNMR (400 MHz, DMSO-d ₆): δ = 0.78-0.82 (m, 3H), 1.29-1.47 (m, 8H), 1.49-1.54 (m, 2H), 3.66-3.87 (m, 5H), 3.96-4.02 (m, 3H), 4.21-4.39 (m, 2H), 5.57 (t, J = 12.0 Hz, 1H), 5.84-6.05 (m, 3H), 6.90 (dd, J1 = 8.0 Hz, J2 = 4.0 Hz, 2H), 7.09-7.14 (dd, J1 = 16.0 Hz, J2 = 4.0 Hz, 2H), 7.55 (d, J = 8.0 Hz, 1H), 11.48-11.62 (s, 1H)
27	4-F-Ph	H	H	Me	Et	1H NMR (DMSO-d ₆) δ 1.12-1.28 (m, 9H), 3.72-3.94 (m, 2H), 3.98-4.10 (m, 3H), 4.21-4.42 (m, 2H), 5.55-5.61 (t, 1H), 5.85-6.20 (m, 3H), 7.18-7.25 (m, 4H), 7.55-7.58 (d, 1H), 11.50 (s, 1H); MS, m/e 533.90 (M + 1)+

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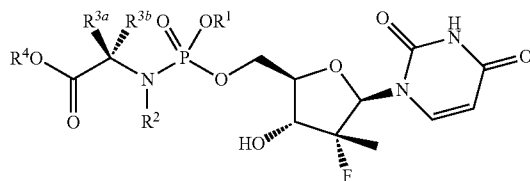
Ex.	R ¹	R ²	R ^{3a}	R ^{3b}	R ⁴	NMR/MS
28	4-F-Ph	H	H	Me	i-Pr	1H NMR (DMSO-d ₆) δ 1.13-1.30 (m, 12H), 3.74-3.85 (m, 2H), 3.98-4.06 (m, 1H), 4.23-4.41 (m, 2H), 4.83-4.87 (m, 1H), 5.55-5.61 (t, 1H), 5.85-6.12 (m, 3H), 7.18-7.24 (m, 4H), 7.55-7.58 (d, 1H), 11.50 (s, 1H); MS, m/e 547.91 (M + 1)+
29	4-F-Ph	H	H	Me	Bn	1H NMR (DMSO-d ₆) δ 1.10-1.23 (m, 6H), 3.65-3.89 (m, 3H), 4.10-4.30 (m, 2H), 4.96-5.00 (m, 2H), 5.46-5.50 (t, 1H), 5.75-5.96 (m, 2H), 6.04-6.12 (m, 1H), 7.05-7.11 (m, 4H), 7.20-7.24 (d, 5H), 7.42-7.45 (d, 1H), 11.50 (s, 1H); MS, m/e 595.94 (M + 1)+
30	4-MeO-Ph	H	H	Me	i-Pr	1HNMR (400 MHz, DMSO-d ₆): δ = 1.15-1.27 (m, 12H), 3.71-3.89 (m, 5H), 3.98-4.02 (m, 1H), 4.22-4.25 (m, 1H), 4.33-4.39 (m, 1H), 4.84-4.87 (m, 1H), 5.57 (t, J = 12.0 Hz, 1H), 5.91-6.03 (m, 3H), 6.90 (d, J = 8.0 Hz, 2H), 7.09-7.14 (m, 2H), 7.55 (d, J = 8.0 Hz, 1H), 11.51 (s, 1H)
31	2-Cl-Ph	H	H	Me	Bn	1H NMR (DMSO-d ₆) δ 1.23 (m, 6H), 3.93-4.00 (m, 3H), 4.27-4.40 (m, 2H), 5.0 (t, J = 7.2 Hz, 2H), 5.53 (m, 1H), 5.80-6.0 (m, 2H), 6.30 (m, 1H), 7.15 (d, J = 2.4 Hz, 1H), 7.27 (m, 6H), 7.51 (m, 3H), 11.5 (s, 1H); MS, m/e 579.87 (M + 1)+/596.78 (M + 18)+
32	2,4-Cl-Ph	H	H	Me	n-Bu	1H NMR (DMSO-d ₆) δ = 0.82 (m, 3H), 1.23 (m, 8H), 1.47 (m, 2H), 3.86 (m, 2H), 3.84 (m, 3H), 4.27-4.43 (m, 2H), 5.5 (m, 1H), 6.02 (m, 2H), 6.35 (m, 1H), 7.44 (m, 3H), 7.77 (m, 1H), 11.5 (s, 1H); MS, m/e 611.87 (M + 1)+
33	4-Me-Ph	H	H	Me	i-Pr	1H NMR (DMSO-d ₆) δ 1.14-1.27 (m, 12H), 2.17-2.26 (m, 3H), 3.73-3.82 (m, 1H), 3.99-4.02 (m, 1H), 4.23-4.26 (m, 1H), 4.37-4.40 (m, 1H), 4.82-4.88 (m, 1H), 5.52-5.58 (m, 1H), 5.85-6.07 (m, 3H), 7.01-7.20 (m, 4H), 7.55 (d, J = 16 Hz, 1H), 11.51 (s, 1H); MS, m/e 543.98 (M + 1)+; 1108.86 (2M + 23)+
34	4-F-Ph	H	H	Me	n-Bu	1H NMR (DMSO-d ₆) δ 0.82-0.89 (m, 3H), 1.20-1.31 (m, 8H), 1.48-1.53 (m, 2H), 3.77-3.90 (m, 2H), 3.95-4.10 (m, 3H), 4.21-4.45 (m, 2H), 5.56-5.61 (t, 1H), 5.83-6.20 (m, 3H), 7.18-7.25 (m, 4H), 7.55-7.58 (d, 1H), 11.50 (s, 1H); MS, m/e 584.1 (M + 23)+
35	3,4-diCl-Ph	H	H	Me	Et	1H NMR (DMSO-d ₆) δ 1.12-1.31 (m, 9H), 3.77-3.92 (m, 2H), 3.95-4.08 (m, 3H), 4.21-4.45 (m, 2H), 5.56-5.62 (t, 1H), 5.80-6.11 (m, 2H), 6.18-6.33 (m, 1H), 7.18-7.25 (m, 1H), 7.49-7.56 (d, 2H), 7.62-7.67 (m, 1H), 11.50 (s, 1H); MS, m/e 606.1 (M + 23)+
36	2-Cl-Ph	H	H	Me	i-Pr	1HNMR (400 MHz, DMSO-d ₆): δ = 1.12-1.16 (m, 6H), 1.21-1.27 (m, 6H), 3.79-3.85 (m, 2H), 4.00-4.07 (m, 1H), 4.28-4.32 (m, 1H), 4.38-4.43 (m, 1H), 4.83-4.87 (m, 1H), 5.56 (dd, J1 = 16.0 Hz, J2 = 8.0 Hz, 1H), 5.85-6.12 (m, 2H), 6.20-6.33 (m, 1H), 7.19-7.22 (m, 1H), 7.33 (t, J = 16.0 Hz, 1H), 7.48-7.55 (m, 3H), 11.55 (s, 1H)
37	4-MeO-Ph	H	H	Me	Bn	1HNMR (400 MHz, DMSO-d ₆): δ = 1.19-1.26 (m, 6H), 3.69-3.70 (s, 3H), 3.87 (m, 2H), 3.99 (m, 1H), 4.20-4.21 (m, 1H), 4.35 (m, 1H), 5.07-5.09 (m, 2H), 5.54 (t, J = 16.0 Hz, 1H), 5.85-5.92 (m, 1H), 6.04-6.10 (m, 2H), 6.86 (d, J = 8.0 Hz, 2H), 7.09 (dd, J1 = 16.0 Hz, J2 = 4.0 Hz, 2H), 7.30-7.34 (m, 5H), 7.53 (s, 1H), 11.52 (s, 1H)
38	Ph	H	H	Me	n-Pen	1H NMR (DMSO-d ₆) δ 0.79-0.81 (m, 3H), 1.17-1.23 (m, 10H), 3.74-3.81 (m, 2H), 3.94-3.96 (m, 3H), 4.19-4.36 (m, 2H), 5.49-5.54 (m, 1H), 5.27-6.08 (m, 3H), 7.14-7.33 (m, 3H), 7.31-7.35 (m, 2H), 7.51 (d, J = 8 Hz, 1H), 11.51 (s, 1H); Ms, m/e 557.9 (M + 1)+; 1136.88 (2M + 23)+
39	4-Cl-Ph	H	H	Me	i-Pr	1H NMR (DMSO-d ₆) δ 1.04-1.19 (m, 12H), 3.76-3.80 (m, 2H), 3.98-4.08 (m, 1H), 4.42-4.42 (m, 2H), 4.82-4.85 (m, 1H), 5.55-5.60 (m, 1H), 5.80-4.20 (m, 3H), 7.20-7.25 (m, 2H), 7.43 (d, J = 8.8 Hz, 1H), 7.54 (d, J = 8 Hz, 1H), 11.51 (s, 1H); MS, m/e 563.88 (M + 1)+; 1148.73 (2M + 23)+
40	4-Cl-Ph	H	H	Me	n-Bu	1H NMR (DMSO-d ₆) δ 0.85 (t, J = 7.2 Hz, 3H), 1.22-1.33 (m, 8H), 1.45-1.53 (m, 2H), 3.80-3.87 (m, 2H), 3.96-4.04 (m, 3H), 4.24-4.27 (m, 1H), 4.35-4.39 (m, 1H), 5.56-5.61 (m, 1H), 5.82-6.11 (m, 2H), 6.15-6.18 (m, 1H), 7.20-7.56 (m, 4H), 7.51-7.57 (m, 1H), 11.54 (s, 1H); MS, m/e 577.95 (M + 1)+
41	4-Cl-Ph	H	H	Me	Et	1H NMR (DMSO-d ₆) δ 1.14 (t, J = 7.0 Hz, 3H), 1.20-1.28 (m, 6H), 3.77-3.88 (m, 2H), 3.99-4.07 (m, 3H), 4.24-4.28 (m, 1H), 4.34-4.43 (m, 1H), 5.56-5.61 (m, 1H), 5.86-6.13 (m, 2H), 6.15-6.24 (m, 1H), 7.20-7.26 (m, 2H), 7.44 (d, J = 7.6 Hz, 2H), 7.55 (d, J = 7.6 Hz, 1H), 11.55 (s, 1H); MS, m/e 549.11 (M + 1)+

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Ex.	R ¹	R ²	R ^{3a}	R ^{3b}	R ⁴	NMR/MS
42	4-Me-Ph	H	H	Me	n-Bu	¹ H NMR (DMSO-d ₆) δ 0.79-0.83 (m, 3H), 1.17-1.28 (m, 8H), 1.45-1.47 (m, 2H), 2.22 (d, J = 2.8 Hz, 1H), 3.70-3.90 (m, 2H), 3.95-3.98 (m, 3H), 4.10-4.40 (m, 2H), 5.51 (t, 1H), 5.80-5.90 (m, 1H), 5.95-6.05 (m, 2H), 7.02-7.06 (m, 2H), 7.51 (t, J = 4.2 Hz, 4H), 7.51 (d, 1H), 11.51 (s, 1H); MS, m/e 557.99 (M + 1) ⁺ ; 1136.84 (2M + 23) ⁺
43	4-Me-Phe	H	H	Me	Bn	¹ H NMR (DMSO-d ₆) δ 1.16-1.24 (m, 6H), 2.22 (s, 3H), 3.65-4.03 (m, 3H), 4.11-4.38 (m, 2H), 5.04-5.05 (m, 2H), 5.48-5.50 (m, 1H), 5.77-5.87 (m, 1H), 5.90-6.11 (m, 2H), 6.98-7.10 (m, 4H), 7.28-7.32 (m, 5H), 7.50 (t, 1H), 11.48 (s, 1H); MS, m/e 592.00 (M + 1) ⁺
44	Ph	H	H	Et	Me	¹ H NMR (DMSO-d ₆) δ 0.70-0.80 (m, 3H), 1.11-1.26 (m, 3H), 1.42-1.61 (m, 2H), 3.50-3.54 (m, 3H), 3.58-3.80 (m, 2H), 3.91-4.02 (m, 1H), 4.12-4.38 (m, 2H), 5.47-5.52 (m, 1H), 5.90-6.03 (m, 2H), 7.08-7.16 (m, 3H), 7.26-7.35 (m, 2H), 7.48 (t, 1H), 11.45 (s, 1H); MS, m/e 515.95 (M + 1) ⁺ ; 1052.82 (2M + 23) ⁺
45	Ph	H	H	Me	4-F-Bn	¹ H NMR (400 MHz, DMSO-d ₆) δ 1.20-1.26 (m, 6H), 3.80-3.93 (m, 2H), 3.98 (s, 1H), 4.23-4.26 (m, 1H), 4.36-4.37 (m, 1H), 5.07 (s, 2H), 5.52-5.55 (m, 1H), 5.86-5.87 (m, 1H), 5.98-6.04 (m, 1H), 6.14-6.17 (m, 1H), 7.15-7.20 (m, 5H), 7.36 (dd, 20.0, 8.0 Hz, 4H), 7.54 (s, 1H), 11.55 (s, 1H)
46	4-Cl-Ph	H	H	Me	n-Bu	¹ H NMR (400 MHz, DMSO-d ₆) δ 1.21-1.28 (m, 6H), 3.71-3.88 (m, 1H), 3.91-3.98 (m, 1H), 4.00-4.01 (m, 1H), 4.23-4.27 (m, 1H), 4.35-4.38 (m, 1H), 5.08 (d, J = 4.0 Hz, 2H), 5.57 (dd, J = 12.0, 8.0 Hz, 1H), 5.91 (d, J = 8.0 Hz, 1H), 6.01 (d, J = 8.0 Hz, 1H), 6.22-6.24 (m, 1H), 7.17-7.23 (m, 2H), 7.31-7.40 (m, 7H), 7.53 (s, 1H), 11.50 (s, 1H)
47	Ph	H	H	Me	3-Me-I-Bu	¹ H NMR (DMSO-d ₆) δ 0.80-0.82 (m, 6H), 1.18-1.40 (m, 8H), 1.50-1.58 (m, 1H), 3.71-3.82 (m, 3H), 3.97-4.01 (m, 3H), 4.21-4.40 (m, 2H), 5.30 (t, J = 8.6 Hz, 1H), 5.81-6.10 (m, 3H), 7.15-7.20 (m, 3H), 7.32-7.36 (m, 2H), 7.48 (d, J = 8.4 Hz, 1H), 11.38 (s, 1H); MS, m/e 557.98 (M + 1) ⁺ ; 1136.88 (2M + 23) ⁺
48	3,4-diCl-Ph	H	H	Me	Bn	¹ H NMR (DMSO-d ₆) δ 1.05-1.37 (m, 6H), 3.71-3.82 (m, 1H), 3.87-4.02 (m, 2H), 4.28-4.29 (m, 1H), 4.36-4.38 (m, 1H), 5.04 (d, J = 5.2 Hz, 2H), 5.55-5.64 (m, 1H), 5.85-5.94 (m, 1H), 6.00-6.05 (m, 1H), 6.29-6.40 (m, 1H), 7.17-7.24 (m, 1H), 7.30-7.41 (m, 5H), 7.45-7.58 (m, 2H), 7.61 (d, J = 4.0 Hz, 1H), 11.53 (s, 1H); MS, m/e 545.80 (M + 1) ⁺
49	Ph	H	H	Me	c-Hex	¹ H NMR (DMSO-d ₆) δ 1.18-1.41 (m, 12H), 1.59-1.67 (m, 4H), 3.74-13.80 (m, 1H), 3.96-4.02 (m, 1H), 4.19-4.26 (m, 1H), 4.31-4.39 (m, 1H), 4.60 (s, 1H), 5.52 (t, J = 7.8 Hz, 1H), 5.80-6.09 (m, 3H), 7.15-7.20 (m, 3H), 7.32-7.36 (m, 2H), 7.52 (d, J = 8 Hz, 1H), 11.50 (s, 1H); MS, m/e 569.98 (M + 1) ⁺ ; 592.14 (M + 23) ⁺
50	Ph	H	Me	H	n-Bu	¹ H NMR (DMSO-d ₆) δ 0.76 (t, J = 7.2 Hz, 3H), 1.10-1.22 (m, 8H), 1.38-1.43 (m, 2H), 3.72-3.75 (m, 2H), 3.87-3.93 (m, 3H), 4.14-4.21 (m, 1H), 4.23-4.33 (m, 1H), 5.46-5.54 (m, 1H), 5.84-6.11 (m, 3H), 7.09-7.14 (m, 2H), 7.27-7.32 (m, 2H), 7.34-7.51 (m, 1H), 11.47 (s, 1H); MS, m/e 543.98 (M + 1) ⁺
51	Ph	H	Me	H	i-Pr	¹ H NMR (DMSO-d ₆) δ 1.39 (d, J = 7.2 Hz, 6H), 1.19-1.29 (m, 6H), 3.65-3.75 (m, 2H), 3.95-4.05 (m, 1H), 4.20-4.22 (m, 1H), 4.31-4.33 (m, 1H), 4.79-4.82 (m, 1H), 5.48-5.57 (m, 1H), 5.84-5.91 (m, 1H), 5.96-6.07 (m, 2H), 7.12-7.35 (m, 5H), 7.44-7.54 (m, 1H), 11.49 (s, 1H); MS, m/e 529.96 (M + 1) ⁺
52	Ph	H	Me	H	Bn	¹ H NMR (DMSO-d ₆) δ 1.18-1.28 (m, 6H), 3.70-3.83 (m, 1H), 3.87-3.94 (m, 1H), 3.99-4.01 (m, 1H), 4.23-4.26 (m, 1H), 4.33-4.37 (m, 1H), 5.03-5.12 (m, 2H), 5.51-5.59 (m, 1H), 5.87-5.90 (m, 1H), 5.95-6.07 (m, 1H), 6.10-6.27 (m, 1H), 7.15-7.23 (m, 3H), 7.31-7.38 (m, 7H), 7.47-7.56 (m, 1H), 11.50 (s, 1H); MS, m/e 577.99 (M + 1) ⁺
53	2-Cl-Ph	H	H	Me	n-Bu	¹ H NMR (400 MHz, DMSO-d ₆) δ 0.81-0.86 (m, 3H), 1.21-1.31 (m, 8H), 1.46-1.52 (m, 2H), 3.84-3.90 (m, 2H), 3.97-4.04 (m, 3H), 4.27-4.41 (m, 2H), 5.53-5.58 (m, 1H), 5.82-5.95 (m, 1H), 5.96-6.10 (m, 1H), 6.27-6.31 (m, 1H), 7.19-7.22 (m, 1H), 7.34 (dd, J = 8.0, 4.0 Hz, 1H), 7.47-7.55 (m, 3H), 11.55 (s, 1H)
54	4-Br-Ph	H	H	Me	i-Pr	¹ H NMR (400 MHz, DMSO-d ₆) δ 1.10-1.14 (m, 6H), 1.20-1.27 (m, 6H), 3.74-3.81 (m, 2H), 3.99-4.01 (m, 1H), 4.21-4.25 (m, 1H), 4.37-4.38 (m, 1H), 4.81-4.85 (m, 1H), 5.58 (dd, J = 8.0, 4.0 Hz, 1H), 5.82-5.95 (m, 1H), 5.96-6.09 (m, 1H), 6.10-6.13 (m, 1H), 7.18 (dd, J = 12.0, 8.0 Hz, 2H), 7.53-7.57 (m, 3H), 11.52 (s, 1H)

-continued



Ex.	R ¹	R ²	R ^{3a}	R ^{3b}	R ⁴	NMR/MS
55	4-F-Ph	H	H	Me	c-Hex	¹ H NMR (DMSO-d ₆) δ 1.20-1.44 (m, 12H), 1.60-1.71 (m, 4H), 3.75-4.02 (m, 2H), 3.94-4.02 (m, 1H), 4.19-4.26 (m, 2H), 4.59-4.61 (m, 1H), 5.57 (t, J = 8.4 Hz, 1H), 5.85-6.06 (m, 3H), 7.17-7.23 (m, 4H), 7.54 (d, J = 8.4 Hz, 1H), 11.50 (s, 1H); MS, m/e 587.92 (M + 1) ⁺
56	4-Br-Ph	H	H	Me	c-Hex	¹ H NMR (400 MHz, DMSO-d ₆): δ = 1.18-1.46 (m, 12H), 1.61-1.69 (m, 4H), 3.75-3.82 (m, 2H), 3.95-4.08 (m, 1H), 4.25-4.28 (m, 1H), 4.38 (s, 1H), 4.60-4.62 (m, 1H), 5.56-5.60 (m, 1H), 5.82-5.95 (m, 1H), 6.02-6.20 (m, 2H), 7.09-7.20 (m, 2H), 7.53-7.57 (m, 3H), 11.52 (s, 1H) MS, m/e 650.0 (M + 3) ⁺
57	Ph	H	H	Et	i-Pr	¹ H NMR (400 MHz, DMSO-d ₆): δ = 0.75-0.82 (m, 3H), 1.12-1.26 (m, 9H), 1.52-1.59 (m, 2H), 3.55-3.68 (m, 1H), 3.72-3.83 (m, 1H), 3.95-4.08 (m, 1H), 4.18-4.28 (m, 1H), 4.32-4.41 (m, 1H), 4.83-4.86 (m, 1H), 5.55 (m, J = 7.6 Hz, 1H), 5.99-6.04 (m, 2H), 6.05-6.10 (m, 1H), 7.14-7.21 (m, 3H), 7.33-7.37 (m, 2H), 7.52-7.54 (m, 1H), 11.53 (s, 1H); MS, m/e 566.07 (M + 23) ⁺
58	Ph	H	H	Et	c-Hex	¹ H NMR (400 MHz, DMSO-d ₆): δ 0.75-0.88 (m, 3H), 1.26-1.46 (m, 9H), 1.52-1.69 (m, 6H), 3.60-3.63 (m, 1H), 3.72-3.90 (m, 1H), 4.02-4.03 (m, 1H), 4.24-4.27 (m, 1H), 4.37-4.39 (m, 1H), 4.63-4.65 (m, 1H), 5.55 (dd, J = 8.0 Hz, 4.4 Hz, 1H), 5.80-5.95 (m, 1H), 6.00-6.07 (m, 2H), 7.15-7.22 (m, 3H), 7.34-7.38 (m, 2H), 7.54 (d, J = 8.0 Hz, 1H), 11.55 (s, 1H); MS, m/e 584.01 (M + 1) ⁺ , 606.17 (M + 23) ⁺
59	4-F-Ph	H	H	Et	c-Hex	¹ H NMR (DMSO-d ₆) δ 0.75-0.84 (m, 3H), 1.24 (d, J = 22.8 Hz, 3H), 1.29-1.47 (m, 6H), 1.51-1.70 (m, 6H), 3.59-3.66 (m, 1H), 3.77-3.84 (m, 1H), 3.98-4.04 (m, 1H), 4.21-4.27 (m, 1H), 4.34-4.41 (m, 1H), 4.60-4.65 (m, 1H), 5.56-5.60 (m, 1H), 5.84-5.90 (m, 1H), 6.00-6.08 (m, 2H), 7.20-7.24 (m, 4H), 7.56 (d, J = 8.0 Hz, 1H), 11.49 (s, 1H); MS, m/e 602.00 (M + 1) ⁺
60	Ph	H	H	Me	F—CH ₂ —CH ₂ —	¹ H NMR (DMSO-d ₆) δ 1.18-1.25 (m, 6H), 3.71-3.89 (m, 2H), 3.92-3.99 (m, 1H), 4.19-4.27 (m, 4H), 4.48-4.61 (m, 2H), 3.94-3.98 (m, 2H), 4.11-4.23 (m, 4H), 5.47-5.52 (m, 1H), 6.01-6.11 (m, 1H), 5.90-6.14 (m, 2H), 7.15-7.21 (m, 3H), 7.32-7.36 (m, 2H), 7.46-7.57 (m, 1H), 11.49 (s, 1H); MS, m/e 533.86 (M + 1) ⁺
61	Ph	H	H	Me	F ₂ CH—CH ₂ —	¹ H NMR (DMSO-d ₆) δ 1.17-1.24 (m, 6H), 3.67-3.81 (m, 1H), 3.89-3.98 (m, 2H), 4.21-4.36 (m, 4H), 5.48-5.53 (m, 1H), 5.82-6.05 (m, 2H), 6.18-6.22 (m, 2H), 7.15-7.20 (m, 3H), 7.32-7.36 (m, 2H), 7.51 (s, 1H), 11.50 (s, 1H); MS, m/e 551.92 (M + 1) ⁺
62	Ph	H	H	Me	(CF ₃) ₂ —CH—	¹ H NMR (DMSO-d ₆) δ 1.13-1.29 (m, 6H), 3.67-3.81 (m, 1H), 3.94-4.32 (m, 4H), 5.47 (t, J = 8 Hz 1H), 5.82-6.01 (m, 2H), 6.33-6.36 (m, 1H), 6.70-6.78 (m, 1H), 7.09-7.15 (m, 3H), 7.28-7.32 (m, 2H), 7.43-7.46 (m, 1H) 11.44 (s, 1H); MS, m/e 637.90 (M + 1) ⁺
63	Ph	H	H	Me	(CH ₂ F) ₂ —CH—	¹ H NMR (DMSO-d ₆) δ 1.20-1.29 (m, 6H), 3.70-3.90 (m, 1H), 3.91-4.12 (m, 2H), 4.20-4.33 (m, 1H), 4.35-4.48 (m, 1H), 4.52-4.55 (m, 2H), 4.63-4.67 (m, 2H), 5.20-5.35 (m, 1H), 5.56 (t, J = 8.4 Hz, 1H), 5.80-5.95 (m, 1H), 5.95-6.10 (m, 1H), 6.18-6.21 (m, 1H), 7.18-7.23 (m, 3H), 7.35-7.39 (m, 2H), 7.54 (s, 1H), 11.55 (s, 1H); MS, m/e 565.98 (M + 1) ⁺
64	Ph	H	H	Me	c-Pr-CH ₂	¹ H NMR (DMSO-d ₆) δ 0.20-0.24 (m, 2H), 0.47-0.48 (m, 2H), 0.76-0.84 (m, 3H), 1.03-1.05 (m, 1H), 1.23 (dd, J = 22.4 6.8 Hz 3H), 1.55-1.60 (m, 2H), 3.61-3.68 (m, 1H), 3.81-3.89 (m, 3H), 3.98-4.03 (m, 1H), 4.23-4.29 (m, 1H), 4.35-4.41 (m, 1H), 5.56-6.00 (m, 1H), 5.88-5.91 (m, 1H), 6.04-6.10 (m, 2H), 7.20-7.24 (m, 4H), 7.55 (d, J = 7.6 Hz 1H), 11.53 (s, 1H); MS, m/e 573.17 (M + 1) ⁺
65	Ph	H	H	Et	c-Pen	¹ H NMR (DMSO-d ₆) δ 0.75-0.83 (m, 3H), 1.20-1.28 (m, 3H), 1.49-1.63 (m, 8H), 1.76-1.80 (m, 2H), 3.58-3.60 (m, 1H), 3.70-3.82 (m, 1H), 3.98-4.05 (m, 1H), 4.24-4.26 (m, 1H), 4.37-4.42 (m, 1H), 5.03 (s, 1H), 5.54-5.57 (m, 1H), 5.90-6.00 (m, 1H), 6.02-6.07 (m, 2H), 7.15-7.22 (m, 3H), 7.35-7.39 (m, 2H) 7.55 (d, J = 8.0 Hz, 1H), 11.55 (s, 1H); MS, m/e 370.03 (M + 1) ⁺

*R² and R^{3b} together are —(CH₂)₃— as derived from L-proline

The Purification Procedure by Prep-HPLC:

[0865] Crude products were dissolved in methanol. Injection volumes of these solutions were 5 mL.

[0866] The preparative HPLC system including 2 sets of Gilson 306 pumps, a Gilson 156 UV/Vis detector, a Gilson 215 injector & fraction collector, with Unipoint control software. A Ymc 25×30×2 mm column was used. The mobile phase was HPLC grade water (A), and HPLC grade acetonitrile (B). Fractions were collected into 100×15 mm glass tubes.

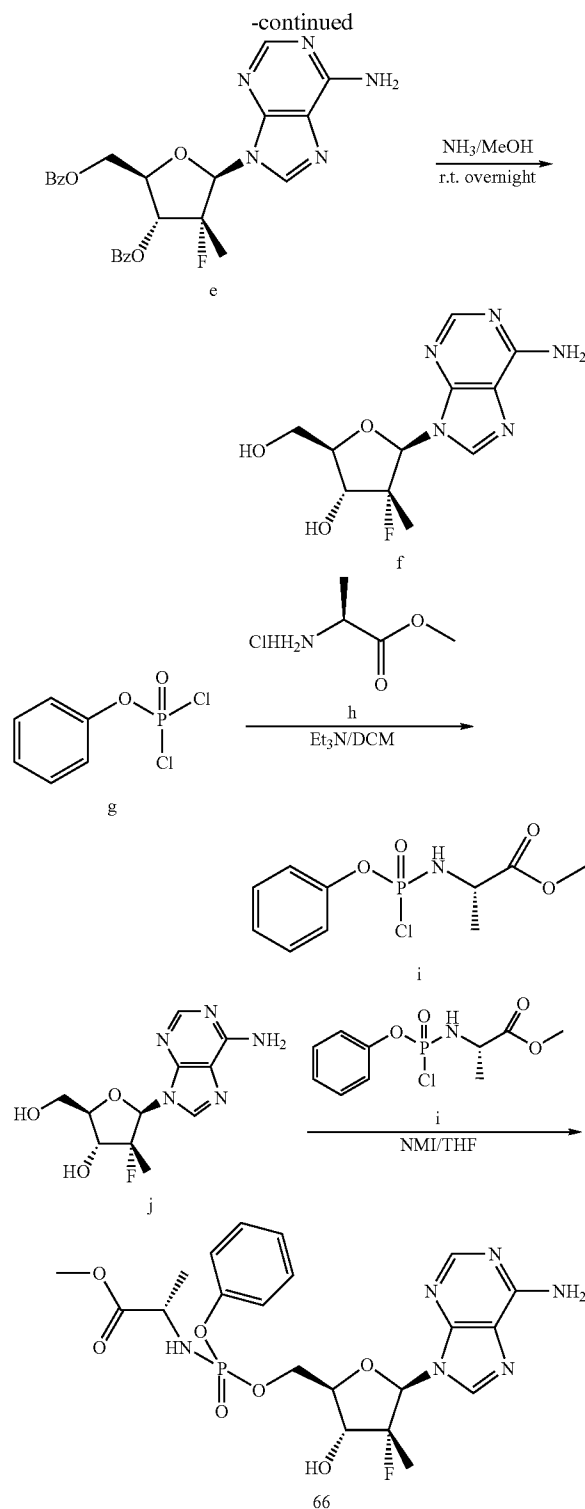
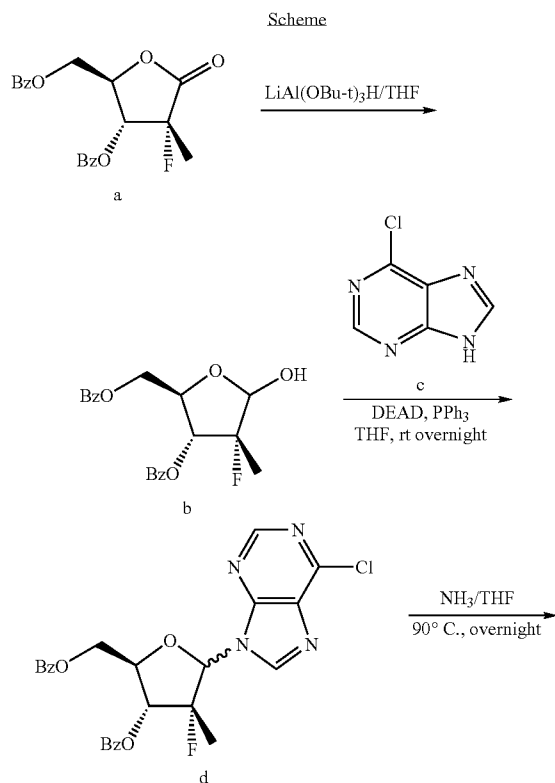
[0867] HPLC gradient is shown in Table 1. Once the gradient was selected, acetonitrile solution was injected into HPLC system, and then fractions collected according to UV peaks. After the separation, each glass tubes were run MS test to collect the desired compounds. The fractions with target MS were combined in a well-weighted flask. Most of acetonitrile was removed under reduce pressure and the remaining solution was freeze-dried to give desired compound.

TABLE 1

Preparative HPLC gradient			
Time (min)	Flow rate (mL/min)	% A	% B
0	15	90	10
30	15	60	40

Preparation of Example 66

[0868]



Preparation of Compound (b)

[0869] To a solution of compound a (1 g, 2.69 mmol) in anhydrous THF (30 mL) was added dropwise 1 M solution of LiAl(OBu-t)₃H in THF (2.69 mL, 2.69 mmol) at -20°C. The

reaction mixture was stirred for 2-3 h at the same temperature. EtOAc (100 mL) was added followed by saturated NH_4Cl solution (10 mL) and reaction mixture was slowly brought to room temperature. Reaction mixture was extracted with EtOAc and washed with 1N HCl and water. Combined organic phase was evaporated to give 0.8 g of crude compound b as transparent oil, which was used directly for next reaction.

Preparation of Compound (d)

[0870] To a solution of compound b (0.8 g, 2.1 mmol), compound c (0.45 g, 2.5 mmol) and Ph_3P (0.56 g, 2.1 mmol) in anhydrous THF (20 mL) under nitrogen atmosphere was added DEAD (1.8 mL). The reaction mixture was stirred at room temperature overnight. The reaction solution was concentrated under reduced pressure. The residue was separated by preparative layer chromatography (hexanes:EtOAc=3:1) to give crude compound d (0.8 g). The crude compound d was used to the next step without further purification.

Preparation of Compound (e)

[0871] Compound d (0.8 g, 1.57 mmol) was dissolved in THF (2 mL) and THF saturated with ammonia (5 mL) was then added to this solution. The reaction mixture was heated to 90°C . overnight. After 18 hours, the solution was cooled to room temperature by ice water, then the solvent was removed under reduced pressure and the residue was purified by column to give compound e (0.75 g) for the next step.

Preparation of Compound (f)

[0872] Compound e (0.5 g, 1.01 mmol) was dissolved in methanol (2 mL) and methanol was saturated with ammonia (5 mL) was then added to this solution. The reaction mixture was stirred at room temperature overnight. After 18 hours, the

solvent was removed under reduced pressure and the residue was purified by column to give crude compound f (0.15 g) for the next step.

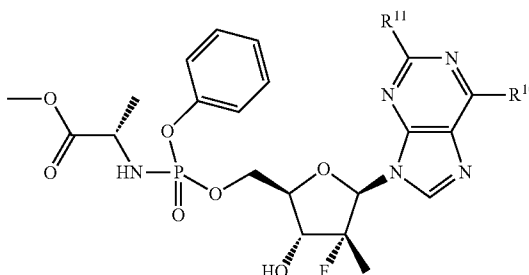
Preparation of Compound (i)

[0873] A solution of triethylamine (1.07 g, 10.6 mmol) in anhydrous dichloromethane (15 mL) was added dropwise to a solution of compound g (1.16 g, 5.3 mmol) and compound h (1.31 g, 5.3 mmol) in dichloromethane (10 mL) with vigorous stirring at -78°C . over a period of 2 hours. After completion of addition, the reaction temperature was allowed to warm to room temperature gradually and stirred over night. Then the solvent was removed under vacuum and anhydrous ether 20 mL was added and the precipitated salt was filtered and the precipitate was washed with ether. The combined organic phase was concentrated to give the colorless oil of compound i (10 g).

Preparation of Compound 66

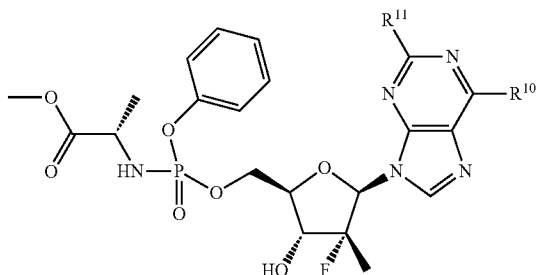
[0874] To a solution of compound j (0.1 g, 0.35 mmol) dissolved in 10 mL of anhydrous THF, stirred and added 0.4 g NMI till the solution became clear, added compound i (0.8 g, 2.89 mmol) in 10 mL THF dropwise, stirred at r.t. overnight. Compound purity and identification was confirmed by LCMS. The solvent was evaporated and purified by Prep-HPLC to afford 66. (25 mg, Yield: 13.6%). ^1H NM(DMSO- d_6) δ 1.08 (d, $J=22.8$ Hz, 3H), 1.17-1.24 (m, 3H), 3.50-3.52 (m, 3H), 3.78-3.83 (m, 1H), 4.10-4.13 (m, 1H), 4.24-4.44 (m, 2H), 5.85-5.92 (m, 1H), 6.01-6.11 (m, 1H), 6.2-6.27 (m, 1H), 7.08-7.19 (m, 4H), 7.31-7.38 (m, 3H), 8.15 (s, 1H), 8.26 (s, 1H); MS, m/e 525 ($M+1$) $^+$.

[0875] Example numbers 67-74, identified below, were prepared using similar procedures disclosed for Example 66, above.



Example	R ¹¹	R ¹⁰	NMR/MS
67	OH	NH ₂	^1H NMR (DMSO- d_6) δ 1.06-1.13 (m, 3H), 1.20-1.24 (m, 3H), 3.27-3.33 (m, 3H), 3.56 (s, 1H), 3.82-3.88 (m, 1H), 4.07-4.13 (m, 1H), 4.25-4.40 (m, 2H), 5.85-5.87 (m, 1H), 5.98-6.09 (m, 2H), 6.59 (s, 32H), 7.14-7.37 (m, 3H), 7.35-7.37 (m, 2H), 7.79 (d, $J = 7.2$ Hz, 1H), 10.69 (s, 1H); MS, m/e 541 ($M+1$) $^+$;
68	NH ₂	NH ₂	^1H NMR (DMSO- d_6) δ 1.07 (d, $J = 22.8$ Hz, 3H), 1.19 (d, $J = 7.2$ Hz, 3H), 3.51 (s, 3H), 3.62 (s, 1H), 3.75-3.81 (m, 1H), 4.05-4.11 (m, 1H), 4.27-4.42 (m, 2H), 5.79-5.83 (m, 1H), 5.92 (s, 2H), 6.00-6.09 (m, 2H), 6.75 (s, 2H), 7.08-7.17 (m, 3H), 7.31-7.35 (m, 2H), 7.78 (s, 1H); MS, m/e 540 ($M+1$) $^+$;
69	NH ₂	c-Pentyl-NH—	^1H NMR (DMSO- d_6) δ 1.05 (d, $J = 22.8$ Hz, 3H), 1.09-1.19 (m, 3H), 1.48 (s, 4H), 1.66 (s, 1H), 1.86 (s, 1H), 3.54 (d, $J = 14$ Hz, 3H), 3.65 (s, 1H), 4.25-4.43 (m, 4H), 5.71-5.82 (m, 1H), 5.94-6.04 (m, 4H), 7.11-7.24 (m, 3H), 7.26-7.34 (m, 2H), 7.77 (d, $J = 3.6$ Hz, 1H); MS, m/e 608 ($M+1$) $^+$

-continued



Example	R ¹¹	R ¹⁰	NMR/MS
70	NH ₂		¹ H NMR (DMSO-d ₆) δ 1.07 (d, J = 22.4 Hz, 3H), 2.35-2.38 (m, 2H), 3.54 (d, J = 9.2 Hz, 3H), 3.59-3.62 (m, 2H), 3.65 (s, 1H), 3.75-3.82 (m, 1H), 4.01-4.13 (m, 2H), 4.22-4.40 (m, 6H), 5.75-5.85 (m, 1H), 6.00-6.07 (m, 4H), 7.15-7.21 (m, 3H), 7.32-7.35 (m, 2H), 7.79 (d, J = 4.0 Hz, 1H); MS, m/e 580 (M + 1) ⁺
71	NH ₂	Et ₂ N—	¹ H NMR (DMSO-d ₆) δ 1.06-1.28 (m, 12H), 3.55 (d, J = 4.8 Hz, 3H), 3.79-3.87 (m, 4H), 4.07-4.12 (m, 2H), 4.29-4.42 (m, 3H), 5.75-5.82 (m, 1H), 5.94 (s, 2H), 6.04-6.10 (m, 2H), 7.14-7.22 (m, 3H), 7.31-7.37 (m, 2H), 7.82 (d, J = 4.4 Hz, 1H); MS, m/e 596 (M + 1) ⁺
72	NH ₂	n-Propyl-NH—	¹ H NMR (DMSO-d ₆) δ 0.84 (t, J = 7.2 Hz, 3H), 1.01-1.01 (m, 3H), 1.09-1.12 (m, 3H), 1.51-1.56 (m, 2H), 3.48 (d, J = 15.2 Hz, 3H), 3.79-3.82 (m, 1H), 4.04-4.05 (m, 1H), 4.27-4.38 (m, 3H), 5.72-5.79 (m, 1H), 5.98-6.04 (m, 4H), 7.13-7.20 (m, 3H), 7.26-7.32 (m, 2H), 7.76 (d, J = 5.2 Hz, 1H); MS, m/e 582 (M + 1) ⁺
73	NH ₂	c-Butyl-NH—	¹ H NMR (DMSO-d ₆) δ 1.02-1.08 (m, 3H), 1.18 (d, J = 4.8 Hz, 3H), 1.44-1.61 (m, 2H), 2.02-2.17 (m, 4H), 3.51 (d, J = 10.8 Hz, 3H), 3.78-3.83 (m, 1H), 4.03-4.06 (m, 1H), 4.27-4.38 (m, 2H), 4.53-4.62 (m, 1H), 5.68-5.79 (m, 1H), 5.95-6.04 (m, 4H), 7.11-7.18 (m, 3H), 7.29-7.35 (m, 2H), 7.51-7.58 (m, 1H), 7.78 (d, J = 5.2 Hz, 1H); MS, m/e 594 (M + 1) ⁺
74	NH ₂		¹ H NMR (DMSO-d ₆) δ 0.97-1.20 (m, 6H), 2.18 (s, 3H), 2.19 (s, 4H), 3.43-3.47 (m, 3H), 3.75 (s, 1H), 4.01-4.06 (m, 4H), 4.22-4.35 (m, 3H), 5.69-5.75 (m, 1H), 5.98-6.05 (m, 3H), 7.09-7.15 (m, 3H), 7.25-7.29 (m, 2H), 7.77 (d, J = 3.6 Hz, 1H); MS, m/e 623 (M + 1) ⁺

[0876] Example numbers 75-80 are prepared using similar procedures disclosed for Example 66, above.

Example	R ¹¹	R ¹⁰
75	H	n-propyl-NH—
76	H	c-Butyl-NH—
77	H	c-Pentyl-NH—

-continued

78	H	
79	H	
80	H	

Example 81

[0877] Certain exemplified compounds were obtained as mixture of diastereomers because of the chirality at phosphorous. The diastereomers were separated on a Chiralpak-AS-H (2×25 cm) column under Supercritical Fluid Chromatogra-

phy (SFC) conditions using 20% methanol in carbon dioxide as solvent. The absolute stereochemistry of the P-chiral center of the diastereomers were not determined. However, chromatographic resolution of these two diastereomers provides for isomers that are characterized as fast eluting and slow eluting isomers. Some examples are shown below.

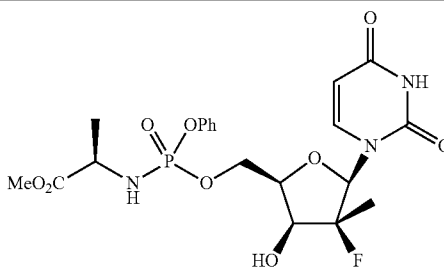
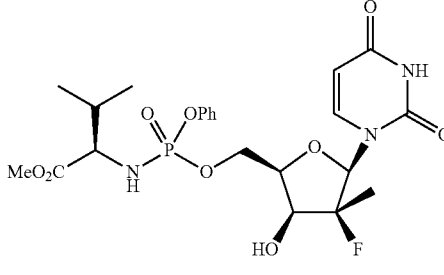
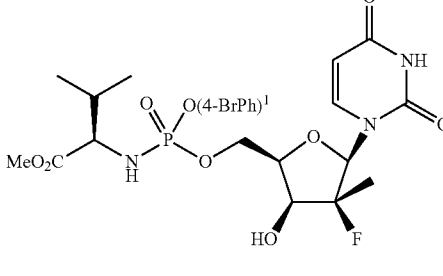
Compound	EC90 (uM)
Example 15 (Diastereomeric mixture)	0.86
Fast Moving isomer of Example 15	1.35
Slow Moving isomer of Example 15	0.26
Example 39 (Diastereomeric mixture)	0.47
Fast Moving isomer of Example 39	0.78
Slow Moving isomer of Example 39	0.02
Example 49 (Diastereomeric mixture)	0.126
Fast Moving isomer of Example 49	0.03
Slow Moving isomer of Example 49	5.78

Example 82

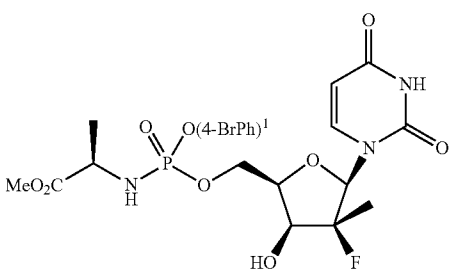
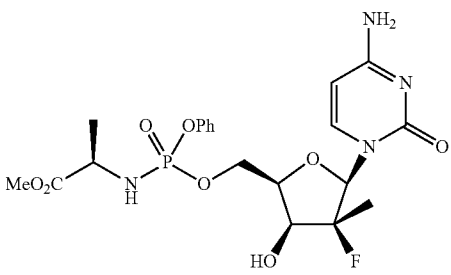
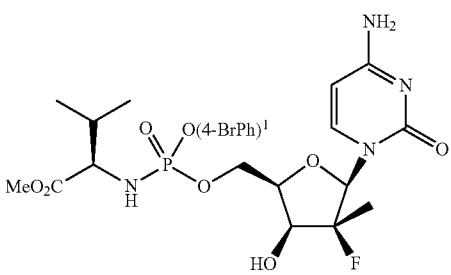
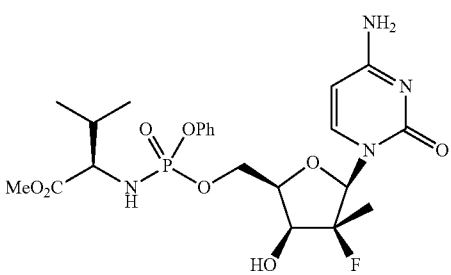
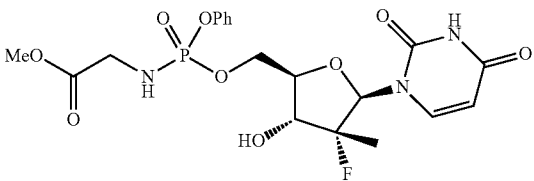
[0878] HCV replicon assay. HCV replicon RNA-containing Huh7 cells (clone A cells; Apath, LLC, St. Louis, Mo.) were kept at exponential growth in Dulbecco's modified Eagle's medium (high glucose) containing 10% fetal bovine serum, 4 mM L-glutamine and 1 mM sodium pyruvate,

1xnonessential amino acids, and G418 (1,000 µg/ml). Antiviral assays were performed in the same medium without G418. Cells were seeded in a 96-well plate at 1,500 cells per well, and test compounds were added immediately after seeding. Incubation time 4 days. At the end of the incubation step, total cellular RNA was isolated (RNeasy 96 kit; Qiagen). Replicon RNA and an internal control (TaqMan rRNA control reagents; Applied Biosystems) were amplified in a single-step multiplex RT-PCR protocol as recommended by the manufacturer. The HCV primers and probe were designed with Primer Express software (Applied Biosystems) and covered highly conserved 5'-untranslated region (UTR) sequences (sense, 5'-AGCCATGGCGTTAGTA(T)GAGTGT-3', and antisense, 5'-TTCCGCAGACCAC-TATGG-3'-probe, 5'-FAM-CCTCCAGGAC-CCCCCTCCC-TAMRA-3').

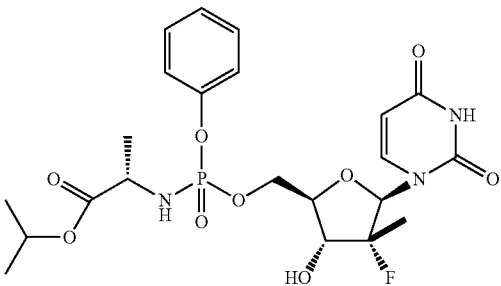
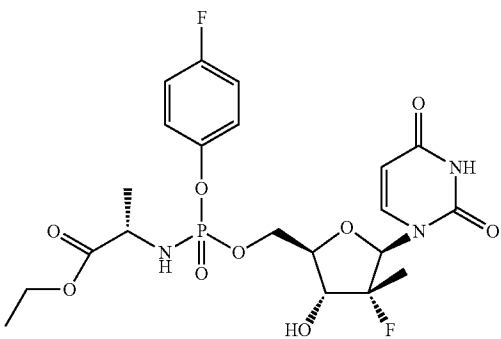
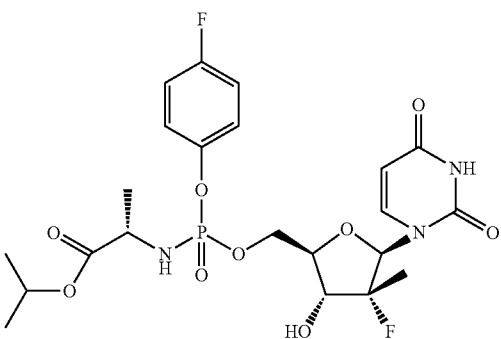
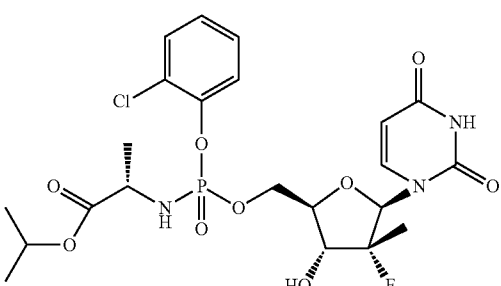
[0879] To express the antiviral effectiveness of a compound, the threshold RT-PCR cycle of the test compound was subtracted from the average threshold RT-PCR cycle of the no-drug control ($\Delta C_{t_{HCV}}$). A ΔC_t of 3.3 equals a 1-log 10 reduction (equal to the 90% effective concentration [EC_{90}]) in replicon RNA levels. The cytotoxicity of the test compound could also be expressed by calculating the $\Delta C_{t_{rRNA}}$ values. The $\Delta \Delta C_t$ specificity parameter could then be introduced ($\Delta C_{t_{HCV}} - \Delta C_{t_{rRNA}}$), in which the levels of HCV RNA are normalized for the rRNA levels and calibrated against the no-drug control.

Ex #	Compound	Log10 Reduction at 50 µM	EC90 (µM)
5		-1.21	3.0
6		-0.45	ND
7		0.31	ND

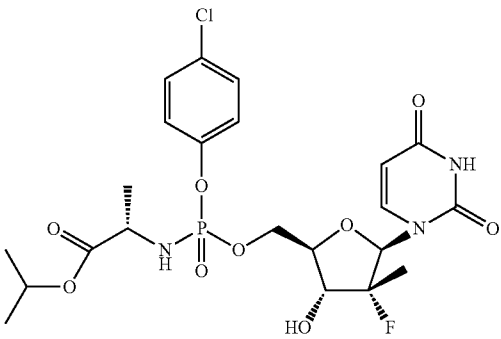
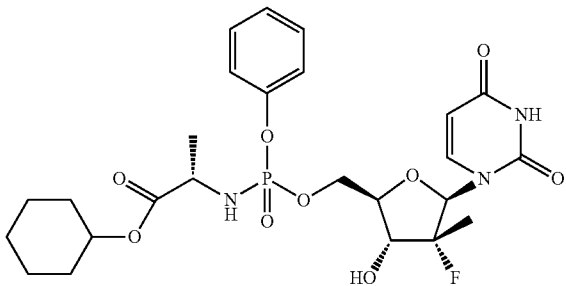
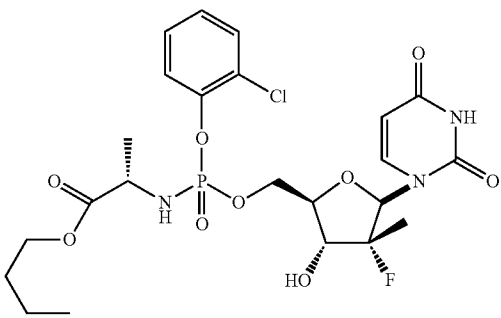
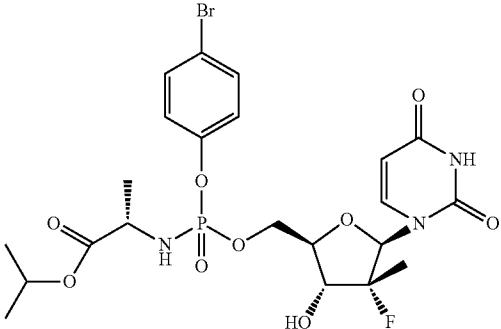
-continued

Ex #	Compound	Log10 Reduction at EC90	
		50 μ M	(μ M)
8		-1.48	2.11
10		-1.25	19.15
11		-0.55	ND
12		0.31	ND
15		ND	0.86

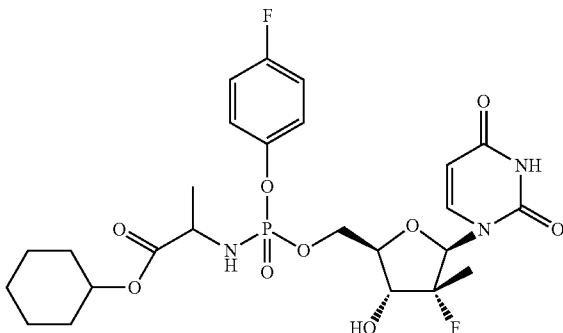
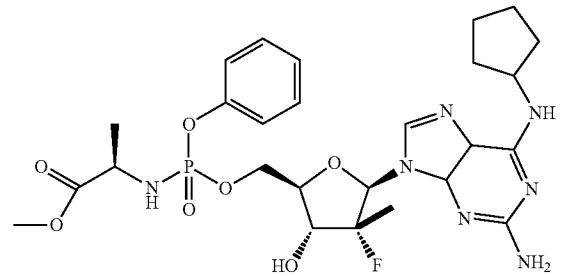
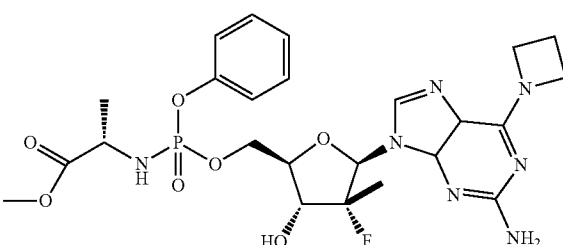
-continued

Ex #	Compound	Log10 Reduction at EC90	
		50 μ M	(μ M)
25		-2.22	0.39
27		-2.25	0.66
28		-2.16	0.75
36		-1.64	21.9

-continued

Ex #	Compound	Log10 Reduction at EC90	
		50 μ M	(μ M)
39		-1.78	0.47
49		-2.69	0.126
53		-1.33	<0.3
54		-1.55	0.57

-continued

Ex #	Compound	Log10 Reduction at EC90	
		50 μ M	(μ M)
55		-2.38	<0.3
69		-2.25	<0.3
70		-2.25	<0.3

¹(4-BrPh): 4-bromo-phenyl.

[0880] The entire contents of U.S. Provisional Application Nos. 60/909,315, filed Mar. 30, 2007, and 60/982,309, filed Oct. 24, 2007, are hereby incorporated by reference in the present application so far as needed to supplement the present disclosure and/or rectify any errors. Moreover, the patent and non-patent references disclosed herein are incorporated by reference. In the event that the incorporated patent and non-

patent reference contains a term that conflicts with a term disclosed in either one of the two Provisional Applications or the present application text, the meaning of the term contained in the present application text and the two Provisional Applications controls provided that the overall meaning of the incorporated subject matter is not lost.

LENGTHY TABLES

The patent application contains a lengthy table section. A copy of the table is available in electronic form from the USPTO web site (<http://seqdata.uspto.gov/?pageRequest=docDetail&DocID=US20100016251A1>). An electronic copy of the table will also be available from the USPTO upon request and payment of the fee set forth in 37 CFR 1.19(b)(3).

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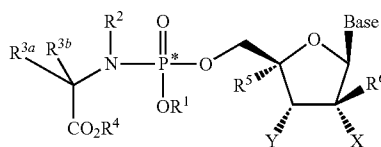
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<400> SEQUENCE: 3

cctccaggac cccccctccc 20

We claim:

1. A compound, its stereoisomer, salt, hydrate, solvate, or crystalline form thereof, represented by formula I:



wherein

- (a) R^1 is hydrogen, n-alkyl; branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-6} alkoxy, F, Cl, Br, I, nitro, cyano, C_{1-6} haloalkyl, $-N(R^1)_2$, C_{1-6} acylamino, $-NHSO_2C_{1-6}$ alkyl, $-SO_2N(R^1)_2$, COR^1 , and $-SO_2C_{1-6}$ alkyl; R^1 is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, $R^{1'}$ is $-OR^1$ or $-N(R^1)_2$;
- (b) R^2 is hydrogen, C_{1-10} alkyl, R^{3a} or R^{3b} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms, $C(O)CR^{3a}R^{3b}NHR^1$, where n is 2 to 4 and R^1 , R^{3a} , and R^{3b} ;
- (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C_{1-10} alkyl, cycloalkyl, $-(CH_2)_c(NR^3)_2$, C_{1-6}

hydroxyalkyl, $-CH_2SH$, $-(CH_2)_2S(O)_dMe$, $-(CH_2)_3NHC(=NH)NH_2$, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, $-(CH_2)_eCOR^{3'}$, aryl and aryl C_{1-3} alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C_{1-10} alkyl, C_{1-6} alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C_{1-6} alkyl; (iii) R^{3a} and R^{3b} together are $(CH_2)_f$ so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where $R^{3'}$ is independently hydrogen or C_{1-6} alkyl and $R^{3''}$ is $-OR^1$ or $-N(R^3)_2$; (vi) R^{3a} is H and R^{3b} is H, CH_3 , CH_2CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl; or (viii) R^{3a} is CH_3 , $-CH_2CH_3$, $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl and R^{3b} is H, where R^3 is independently hydrogen or alkyl,

6 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(phenyl methoxy-valyl phosphate)((S)-2-{[(2R,3R,4R,5R)-5-(2,4-

- Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-3-methyl-butyric acid methyl ester;
- 7 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-valyl phosphate((S)-2-((4-bromophenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 8 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-alanyl phosphate(((S)-2-((4-bromophenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester);
- 9 N⁴-(N,N-dimethylformamidinyl)-2'-deoxy-2'-fluoro-2'-C-methylcytidine;
- 10 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(phenyl methoxy-alanyl phosphate(((S)-2-((2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester);
- 11 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(4-bromophenyl methoxy-valyl phosphate(2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(4-bromophenyl methoxy-valyl phosphate((S)-2-((4-bromophenoxy)-[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 12 2'-deoxy-2'-fluoro-2'-C-methylcytidine 5'-(phenyl methoxy-valyl phosphate((S)-2-((2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-3-methyl-butyric acid methyl ester;
- 13 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino)-propionic acid ethyl ester;
- 14 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-naphthalen-1-yloxy)-phosphorylamino)-propionic acid benzyl ester;
- 15 (((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino)-acetic acid methyl ester;
- 16 (S)-2-((2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino)-propionic acid methyl ester;
- 17 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-naphthalen-1-yloxy)-phosphorylamino)-propionic acid methyl ester;
- 18 (S)-1-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphoryl)-pyrrolidine-2-carboxylic acid methyl ester;
- 19 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino)-propionic acid butyl ester;
- 20 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino)-propionic acid benzyl ester;
- 21 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy)-(4-fluoro-phenoxy)-phosphorylamino)-propionic acid methyl ester;
- 22 (S)-2-((4-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino)-propionic acid methyl ester;
- 23 (S)-2-((3,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino)-propionic acid methyl ester;
- 24 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy)-phosphorylamino)-propionic acid sec-butyl ester;
- 25 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy)-phosphorylamino)-propionic acid isopropyl ester;
- 26 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy)-(4-methoxy-phenoxy)-phosphorylamino)-propionic acid butyl ester;
- 27 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy)-(4-fluoro-phenoxy)-phosphorylamino)-propionic acid ethyl ester;
- 28 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy)-(4-fluoro-phenoxy)-phosphorylamino)-propionic acid ethyl ester;
- 29 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy)-(4-fluoro-phenoxy)-phosphorylamino)-propionic acid benzyl ester;
- 30 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy)-(4-methoxy-phenoxy)-phosphorylamino)-propionic acid isopropyl ester;
- 31 (S)-2-((2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino)-propionic acid benzyl ester;
- 32 (S)-2-((2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino)-propionic acid butyl ester;
- 33 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy)-p-tolylloxy-phosphorylamino)-propionic acid isopropyl ester;
- 34 (S)-2-(((2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy)-(4-fluoro-phenoxy)-phosphorylamino)-propionic acid butyl ester;
- 35 (S)-2-((3,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino)-propionic acid ethyl ester;

- [illegible]

67 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-hydroxy-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;

68 (S)-2-[(R)-[(2R,3R,4R,5R)-5-(2,6-Diamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;

69 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-cyclopentylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;

70 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-azetidin-1-yl-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;

71 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-diethylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;

72 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-propylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;

73 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-cyclobutylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;

74 (S)-2-[[[(2R,3R,4R,5R)-5-[2-Amino-6-(4-methyl-piperazin-1-yl)-purin-9-yl]-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid methyl ester;

75 (2S)-methyl 2-((((2R,3R,4R,5R)-4-fluoro-3-hydroxy-4-methyl-5-(6-(propylamino)-9H-purin-9-yl)tetrahydro-furan-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;

76 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(cyclobutylamino)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;

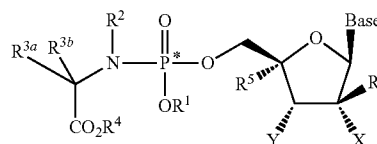
77 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6(cyclopentylamino)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;

78 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(aziridin-1-yl)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;

79 (2S)-methyl 2-((((2R,3R,4R,5R)-4-fluoro-3-hydroxy-4-methyl-5-(6-(piperidin-1-yl)-9H-purin-9-yl)tetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate; and

80 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(azetidin-1-yl)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate.

3. A composition for the treatment and/or prophylaxis of any of the viral agents disclosed herein said composition comprising a pharmaceutically acceptable medium selected from among an excipient, carrier, diluent, and equivalent medium and a compound, its stereoisomer, salt, hydrate, solvate, or crystalline form thereof, represented by formula I:



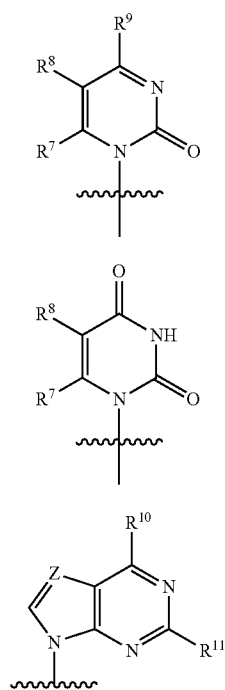
wherein

- (a) R^1 is hydrogen, n-alkyl; branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-6} alkoxy, F, Cl, Br, I, nitro, cyano, C_{1-6} haloalkyl, $-N(R^1)_2$, C_{1-6} acylamino, $-NHSO_2C_{1-6}$ alkyl, $-SO_2N(R^1)_2$, $COR^{1'}$, and $-SO_2C_{1-6}$ alkyl; ($R^{1'}$ is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, $R^{1'}$ is $-OR'$ or $-N(R^{1'})_2$);
- (b) R^2 is hydrogen, C_{1-10} alkyl, R^{3a} or R^{3b} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms, $C(O)CR^{3a}R^{3b}NHR^1$, where n is 2 to 4 and R^1 , R^{3a} , and R^{3b} ;
- (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C_{1-10} alkyl, cycloalkyl, $-(CH_2)_c(NR^{3'})_2$, C_{1-6} hydroxyalkyl, $-CH_2SH$, $-(CH_2)_2S(O)_dMe$, $-(CH_2)_3NHC(=NH)NH_2$, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, $-(CH_2)_eCOR^{3''}$, aryl and aryl C_{1-3} alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C_{1-10} alkyl, C_{1-6} alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C_{1-6} alkyl; (iii) R^{3a} and R^{3b} together are $(CH_2)_n$ so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where $R^{3'}$ is independently hydrogen or C_{1-6} alkyl and $R^{3''}$ is $-OR'$ or $-N(R^{3'})_2$; (vi) R^{3a} is H and R^{3b} is H, CH_3 , CH_2CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl; or (viii) R^{3a} is CH_3 , $-CH_2CH_3$, $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl and R^{3b} is H where $R^{3'}$ is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, $R^{3''}$ is $-OR'$ or $-N(R^{3'})_2$);
- (d) R^4 is hydrogen, C_{1-10} alkyl, C_{1-10} alkyl optionally substituted with a lower alkyl, alkoxy, di(lower alkyl)-amino, or halogen, C_{1-10} haloalkyl, C_{3-10} cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;
- (e) R^5 is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., $-(CH_2)_pOH$, where p is 1-6,

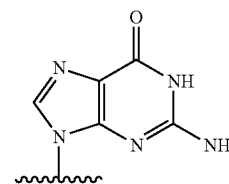
including hydroxyl methyl (CH_2OH), CH_2F , N_3 , CH_2CN , CH_2NH_2 , CH_2NHCH_3 , $\text{CH}_2\text{N}(\text{CH}_3)_2$, alkylne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R^6 is H, R^5 cannot be N_3 and when X is OH, R^6 is CH_3 or CH_2F and B is a purine base, R^5 cannot be H;

- (f) R^6 H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;
 (g) X is H, OH, F, OMe, halogen, NH_2 , or N_3 ;
 (h) Y is OH, H, C_{1-4} alkyl, C_{2-4} alkenyl, C_{2-4} alkynyl, vinyl, N_3 , CN, Cl, Br, F, I, NO_2 , $\text{OC}(\text{O})\text{O}(\text{C}_{1-4}\text{alkyl})$, $\text{OC}(\text{O})\text{O}(\text{C}_{1-4}\text{alkyl})$, $\text{OC}(\text{O})\text{O}(\text{C}_{2-4}\text{alkenyl})$, $\text{OC}(\text{O})\text{O}(\text{C}_{2-4}\text{alkynyl})$, $\text{OC}(\text{O})\text{O}(\text{C}_{2-4}\text{alkenyl})$, OC_{1-10} haloalkyl, O(aminoacyl), $\text{O}(\text{C}_{1-10}\text{acyl})$, $\text{O}(\text{C}_{1-4}\text{alkyl})$, $\text{O}(\text{C}_{2-4}\text{alkenyl})$, $\text{S}(\text{C}_{1-4}\text{acyl})$, $\text{S}(\text{C}_{1-4}\text{alkyl})$, $\text{S}(\text{C}_{2-4}\text{alkynyl})$, $\text{S}(\text{C}_{2-4}\text{alkenyl})$, $\text{SO}(\text{C}_{1-4}\text{acyl})$, $\text{SO}(\text{C}_{1-4}\text{alkyl})$, $\text{SO}(\text{C}_{2-4}\text{alkynyl})$, $\text{SO}(\text{C}_{2-4}\text{alkenyl})$, $\text{SO}_2(\text{C}_{1-4}\text{acyl})$, $\text{SO}_2(\text{C}_{1-4}\text{alkyl})$, $\text{SO}_2(\text{C}_{2-4}\text{alkynyl})$, $\text{SO}_2(\text{C}_{2-4}\text{alkenyl})$, $\text{OS}(\text{O})_2(\text{C}_{1-4}\text{acyl})$, $\text{OS}(\text{O})_2(\text{C}_{1-4}\text{alkyl})$, $\text{OS}(\text{O})_2(\text{C}_{2-4}\text{alkynyl})$, $\text{OS}(\text{O})_2(\text{C}_{2-4}\text{alkenyl})$, NH_2 , $\text{NH}(\text{C}_{1-4}\text{alkyl})$, $\text{NH}(\text{C}_{2-4}\text{alkenyl})$, $\text{NH}(\text{C}_{2-4}\text{alkynyl})$, $\text{NH}(\text{C}_{1-4}\text{acyl})$, $\text{N}(\text{C}_{1-4}\text{alkyl})_2$, $\text{N}(\text{C}_{1-18}\text{acyl})_2$, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N_3 , CN, one to three halogen (Cl, Br, F, I), NO_2 , $\text{C}(\text{O})\text{O}(\text{C}_{1-4}\text{alkyl})$, $\text{C}(\text{O})\text{O}(\text{C}_{1-4}\text{alkyl})$, $\text{C}(\text{O})\text{O}(\text{C}_{2-4}\text{alkynyl})$, $\text{C}(\text{O})\text{O}(\text{C}_{2-4}\text{alkenyl})$, $\text{O}(\text{C}_{1-4}\text{acyl})$, $\text{O}(\text{C}_{1-4}\text{alkyl})$, $\text{O}(\text{C}_{2-4}\text{alkenyl})$, $\text{S}(\text{C}_{1-4}\text{acyl})$, $\text{S}(\text{C}_{1-4}\text{alkyl})$, $\text{S}(\text{C}_{2-4}\text{alkynyl})$, $\text{S}(\text{C}_{2-4}\text{alkenyl})$, $\text{SO}(\text{C}_{1-4}\text{acyl})$, $\text{SO}(\text{C}_{1-4}\text{alkyl})$, $\text{SO}(\text{C}_{2-4}\text{alkynyl})$, $\text{SO}(\text{C}_{2-4}\text{alkenyl})$, $\text{SO}_2(\text{C}_{1-4}\text{acyl})$, $\text{SO}_2(\text{C}_{2-4}\text{alkynyl})$, $\text{SO}_2(\text{C}_{2-4}\text{alkenyl})$, $\text{OS}(\text{O})_2(\text{C}_{1-4}\text{acyl})$, $\text{OS}(\text{O})_2(\text{C}_{1-4}\text{alkyl})$, $\text{OS}(\text{O})_2(\text{C}_{2-4}\text{alkenyl})$, NH_2 , $\text{NH}(\text{C}_{1-4}\text{alkyl})$, $\text{NH}(\text{C}_{2-4}\text{alkenyl})$, $\text{NH}(\text{C}_{2-4}\text{alkynyl})$, $\text{NH}(\text{C}_{1-4}\text{acyl})$, $\text{N}(\text{C}_{1-4}\text{alkyl})_2$, $\text{N}(\text{C}_{1-4}\text{acyl})_2$;

the base is a naturally occurring or modified purine or pyrimidine base represented by the following structures:



-continued



d

wherein

Z is N or CR^{12} ;

R^7 , R^8 , R^9 , R^{10} , and R^{11} are independently H, F, Cl, Br, I, OH, OR', SH, SR', NH_2 , NHR' , NR'_2 , lower alkyl of $\text{C}_1\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkyl of $\text{C}_1\text{-C}_6$, lower alkenyl of $\text{C}_2\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkenyl of $\text{C}_2\text{-C}_6$, lower alkynyl of $\text{C}_2\text{-C}_6$ such as $\text{C}\equiv\text{CH}$, halogenated (F, Cl, Br, I) lower alkynyl of $\text{C}_2\text{-C}_6$, lower alkoxy of $\text{C}_1\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkoxy of $\text{C}_1\text{-C}_6$, CO_2H , $\text{CO}_2\text{R}'$, CONH_2 , CONHR' , CONR'_2 , $\text{C}=\text{CHCO}_2\text{H}$, or $\text{CH}=\text{CHCO}_2\text{R}'$,

wherein R' is an optionally substituted alkyl, which includes, but is not limited to, an optionally substituted C_{1-20} alkyl, an optionally substituted C_{1-10} alkyl, an optionally substituted lower alkyl; an optionally substituted cycloalkyl; an optionally substituted alkynyl of $\text{C}_2\text{-C}_6$, an optionally substituted lower alkenyl of $\text{C}_2\text{-C}_6$, or optionally substituted acyl, which includes but is not limited to $\text{C}(\text{O})$ alkyl, $\text{C}(\text{O})(\text{C}_{1-20}\text{alkyl})$, $\text{C}(\text{O})(\text{C}_{1-10}\text{alkyl})$, or $\text{C}(\text{O})(\text{lower alkyl})$ or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

R^{12} is an H, halogen (including F, Cl, Br, I), OH, OR', SH, SR', NH_2 , NHR' , NR'_2 , NO_2 lower alkyl of $\text{C}_1\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkyl of $\text{C}_1\text{-C}_6$, lower alkenyl of $\text{C}_2\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkenyl of $\text{C}_2\text{-C}_6$, lower alkynyl of $\text{C}_2\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkynyl of $\text{C}_2\text{-C}_6$, lower alkoxy of $\text{C}_1\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkoxy of $\text{C}_1\text{-C}_6$, CO_2H , $\text{CO}_2\text{R}'$, CONH_2 , CONHR' , CONR'_2 , $\text{CH}=\text{CHCO}_2\text{H}$, or $\text{CH}=\text{CHCO}_2\text{R}'$; with the proviso that when base is represented by the structure C with R^{11} being hydrogen, R^{12} is not a: (i) $\text{C}\equiv\text{C}-\text{H}$, (ii) $\text{C}=\text{CH}_2$, or (iii) NO_2 .

4. A composition for the treatment and/or prophylaxis of any of the viral agents disclosed herein said composition comprising a pharmaceutically acceptable medium selected from among an excipient, carrier, diluent, and equivalent medium and a compound, its stereoisomer, salt, hydrate, solvate, or crystalline form thereof, selected from among

5 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-phenyl methoxy-alanyl phosphate((S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester);

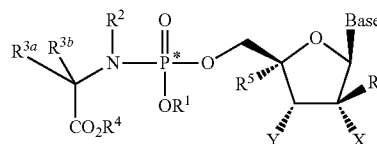
6 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(phenyl methoxy-valyl phosphate((S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-3-methyl-butyric acid methyl ester);

- 7 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-valyl phosphate((S)-2-{(4-bromophenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 8 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-alanyl phosphate(((S)-2-{(4-bromophenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester);
- 9 N⁴-(N,N-dimethylformamidinyl)-2'-deoxy-2'-fluoro-2'-C-methylcytidine;
- 10 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(phenyl methoxy-alanyl phosphate(((S)-2-{[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester);
- 11 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(4-bromophenyl methoxy-valyl phosphate(2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-valyl phosphate((S)-2-{(4-bromophenoxy)-[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 12 2'-deoxy-2'-fluoro-2'-C-methylcytidine 5-(phenyl methoxy-valyl phosphate((S)-2-{[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 13 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid ethyl ester;
- 14 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(naphthalen-1-yloxy)-phosphorylamino}-propionic acid benzyl ester;
- 15 {[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-acetic acid methyl ester;
- 16 (S)-2-{(2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-ethyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
- 17 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(naphthalen-1-yloxy)-phosphorylamino}-propionic acid methyl ester;
- 18 (S)-1-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphoryl}-pyrrolidine-2-carboxylic acid methyl ester;
- 19 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid butyl ester;
- 20 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid benzyl ester;
- 21 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino}-propionic acid methyl ester;
- 22 (S)-2-{(4-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
- 23 (S)-2-{(3,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
- 24 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid sec-butyl ester;
- 25 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid isopropyl ester;
- 26 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-methoxy-phenoxy)-phosphorylamino}-propionic acid butyl ester;
- 27 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino}-propionic acid ethyl ester;
- 28 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino}-propionic acid ethyl ester;
- 29 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino}-propionic acid benzyl ester;
- 30 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-methoxy-phenoxy)-phosphorylamino}-propionic acid isopropyl ester;
- 31 (S)-2-{(2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid benzyl ester;
- 32 (S)-2-{(2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid butyl ester;
- 33 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-p-tolyloxy-phosphorylamino}-propionic acid isopropyl ester;
- 34 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino}-propionic acid butyl ester;
- 35 (S)-2-{(3,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid ethyl ester;
- 36 (S)-2-{(2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid isopropyl ester;

- 68 (S)-2-[(R)-[(2R,3R,4R,5R)-5-(2,6-Diamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 69 (S)-2-[(2R,3R,4R,5R)-5-(2-Amino-6-cyclopentylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 70 (S)-2-[(2R,3R,4R,5R)-5-(2-Amino-6-azetidin-1-yl-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 71 (S)-2-[(2R,3R,4R,5R)-5-(2-Amino-6-diethylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 72 (S)-2-[(2R,3R,4R,5R)-5-(2-Amino-6-propylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 73 (S)-2-[(2R,3R,4R,5R)-5-(2-Amino-6-cyclobutylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 74 (S)-2-[(2R,3R,4R,5R)-5-[2-Amino-6-(4-methyl-piperazin-1-yl)-purin-9-yl]-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 75 (2S)-methyl 2-((((2R,3R,4R,5R)-4-fluoro-3-hydroxy-4-methyl-5-(6-(propylamino)-9H-purin-9-yl)tetrahydro-furan-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 76 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(cyclobutylamino)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 77 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6(cyclopentylamino)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 78 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(aziridin-1-yl)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 79 (2S)-methyl 2-((((2R,3R,4R,5R)-4-fluoro-3-hydroxy-4-methyl-5-(6-(piperidin-1-yl)-9H-purin-9-yl)tetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate; and
- 80 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(azetidin-1-yl)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate.

5. A use of the compound represented by formula I in the manufacture of a medicament for the treatment of any condition the result of an infection by hepatitis C virus, West Nile virus, yellow fever virus, dengue virus, rhinovirus, polio virus, hepatitis A virus, bovine viral diarrhea virus or Japanese encephalitis virus.

wherein the compound, its stereoisomer, salt, hydrate, solvate, or crystalline form thereof, are represented by formula I:



wherein

- (a) R¹ is hydrogen, n-alkyl; branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C₁₋₆ alkyl, C₂₋₆ alkenyl, C₁₋₆ alkoxy, F, Cl, Br, I, nitro, cyano, C₁₋₆ haloalkyl, —N(R^{1'})₂, C₁₋₆ acylamino, —NH—SO₂—C₁₋₆ alkyl, —SO₂—N(R^{1'})₂, COR^{1''}, and —SO₂—C₁₋₆ alkyl; (R^{1'} is independently hydrogen or alkyl, which includes, but is not limited to, C₁₋₂₀ alkyl, C₁₋₁₀ alkyl, or C₁₋₆ alkyl, R^{1''} is —OR' or —N(R^{1'})₂);
- (b) R² is hydrogen, C₁₋₁₀ alkyl, R^{3a} or R^{3b} and R² together are (CH₂)_n so as to form a cyclic ring that includes the adjoining N and C atoms, C(O)CR^{3a}R^{3b}NHR, where n is 2 to 4 and R¹, R^{3a}, and R^{3b};
- (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C₁₋₁₀ alkyl, cycloalkyl, —(CH₂)_c(NR^{3'})₂, C₁₋₆ hydroxyalkyl, —CH₂SH, —(CH₂)₂S(O)_dMe, —(CH₂)₃NHC(=NH)NH₂, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, —(CH₂)_e, COR^{3''}, aryl and aryl C₁₋₃ alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C₁₋₁₀ alkyl, C₁₋₆ alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C₁₋₆ alkyl; (iii) R^{3a} and R^{3b} together are (CH₂)_f so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R² together are (CH₂)_n so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where R^{3'} is independently hydrogen or C₁₋₆ alkyl and R^{3''} is —OR' or —N(R^{3'})₂; (v) R^{3a} is H and R^{3b} is H, CH₃, CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl; or (viii) R^{3a} is CH₃, —CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl and R^{3b} is H, where R^{3'} is independently hydrogen or alkyl, which includes, but is not limited to, C₁₋₂₀ alkyl, C₁₋₁₀ alkyl, or C₁₋₆ alkyl, R^{3''} is —OR' or —N(R^{3'})₂);
- (d) R⁴ is hydrogen, C₁₋₁₀ alkyl, C₁₋₁₀ alkyl optionally substituted with a lower alkyl, alkoxy, di(lower alkyl)-amino, or halogen, C₁₋₁₀ haloalkyl, C₃₋₁₀ cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;
- (e) R⁵ is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., —(CH₂)_pOH, where p is 1-6,

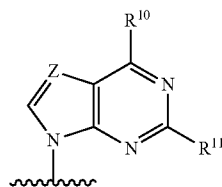
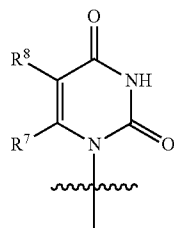
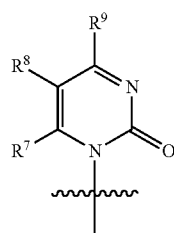
including hydroxyl methyl (CH_2OH), CH_2F , N_3 , CH_2CN , CH_2NH_2 , CH_2NHCH_3 , $\text{CH}_2\text{N}(\text{CH}_3)_2$, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R^6 is H, R^5 cannot be N_3 and when X is OH, R^6 is CH_3 or CH_2F and B is a purine base, R^5 cannot be H;

(f) R^6 is H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;

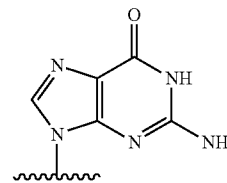
(g) X is H, OH, F, OMe, halogen, NH_2 , or N_3 ;

(h) Y is OH, H, C_{1-4} alkyl, C_{2-4} alkenyl, C_{2-4} alkynyl, vinyl, N_3 , CN, Cl, Br, F, I, NO_2 , $\text{OC}(\text{O})\text{O}(\text{C}_{1-4}$ alkyl), $\text{OC}(\text{O})\text{O}(\text{C}_{1-4}$ alkyl), $\text{OC}(\text{O})\text{O}(\text{C}_{2-4}$ alkynyl), $\text{OC}(\text{O})\text{O}(\text{C}_{2-4}$ alkenyl), OC_{1-10} haloalkyl, O(aminoacyl), $\text{O}(\text{C}_{1-10}$ acyl), $\text{O}(\text{C}_{1-4}$ alkyl), $\text{O}(\text{C}_{2-4}$ alkenyl), $\text{S}(\text{C}_{1-4}$ acyl), $\text{S}(\text{C}_{1-4}$ alkyl), $\text{S}(\text{C}_{2-4}$ alkynyl), $\text{S}(\text{C}_{2-4}$ alkenyl), $\text{SO}(\text{C}_{1-4}$ acyl), $\text{SO}(\text{C}_{1-4}$ alkyl), $\text{SO}(\text{C}_{2-4}$ alkynyl), $\text{SO}(\text{C}_{2-4}$ alkenyl), $\text{SO}_2(\text{C}_{1-4}$ acyl), $\text{SO}_2(\text{C}_{1-4}$ alkyl), $\text{SO}_2(\text{C}_{2-4}$ alkynyl), $\text{SO}_2(\text{C}_{2-4}$ alkenyl), $\text{OS}(\text{O})_2(\text{C}_{1-4}$ acyl), $\text{OS}(\text{O})_2(\text{C}_{1-4}$ alkyl), $\text{OS}(\text{O})_2(\text{C}_{2-4}$ alkynyl), NH_2 , $\text{NH}(\text{C}_{1-4}$ alkyl), $\text{NH}(\text{C}_{2-4}$ alkenyl), $\text{NH}(\text{C}_{2-4}$ alkynyl), $\text{NH}(\text{C}_{1-4}$ acyl), $\text{N}(\text{C}_{1-4}$ alkyl) $_2$, $\text{N}(\text{C}_{1-18}$ acyl) $_2$, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N_3 , CN, one to three halogen (Cl, Br, F, I), NO_2 , $\text{C}(\text{O})\text{O}(\text{C}_{1-4}$ alkyl), $\text{C}(\text{O})\text{O}(\text{C}_{1-4}$ alkyl), $\text{C}(\text{O})\text{O}(\text{C}_{2-4}$ alkynyl), $\text{C}(\text{O})\text{O}(\text{C}_{2-4}$ alkenyl), $\text{O}(\text{C}_{1-4}$ acyl), $\text{O}(\text{C}_{1-4}$ alkyl), $\text{O}(\text{C}_{2-4}$ alkynyl), $\text{S}(\text{C}_{1-4}$ acyl), $\text{S}(\text{C}_{1-4}$ alkyl), $\text{S}(\text{C}_{2-4}$ alkynyl), $\text{S}(\text{C}_{2-4}$ alkenyl), $\text{SO}(\text{C}_{1-4}$ acyl), $\text{SO}(\text{C}_{1-4}$ alkyl), $\text{SO}(\text{C}_{2-4}$ alkynyl), $\text{SO}(\text{C}_{2-4}$ alkenyl), $\text{SO}_2(\text{C}_{1-4}$ acyl), $\text{SO}_2(\text{C}_{1-4}$ alkyl), $\text{SO}_2(\text{C}_{2-4}$ alkynyl), $\text{SO}_2(\text{C}_{2-4}$ alkenyl), $\text{OS}(\text{O})_2(\text{C}_{1-4}$ acyl), $\text{OS}(\text{O})_2(\text{C}_{1-4}$ alkyl), $\text{OS}(\text{O})_2(\text{C}_{2-4}$ alkynyl), NH_2 , $\text{NH}(\text{C}_{1-4}$ alkyl), $\text{NH}(\text{C}_{2-4}$ alkenyl), $\text{NH}(\text{C}_{2-4}$ alkynyl), $\text{NH}(\text{C}_{1-4}$ acyl), $\text{N}(\text{C}_{1-4}$ alkyl) $_2$, $\text{N}(\text{C}_{1-4}$ acyl) $_2$;

the base is a naturally occurring or modified purine or pyrimidine base represented by the following structures:



-continued



wherein

Z is N or CR^{12} ;

R^7 , R^8 , R^9 , R^{10} , and R^{11} are independently H, F, Cl, Br, I, OH, OR' , SH, SR' , NH_2 , NHR' , NR'_2 , lower alkyl of C_1 - C_6 , halogenated (F, Cl, Br, I) lower alkyl of C_1 - C_6 , lower alkenyl of C_2 - C_6 , halogenated (F, Cl, Br, I) lower alkenyl of C_2 - C_6 , lower alkynyl of C_2 - C_6 such as $\text{C}\equiv\text{CH}$, halogenated (F, Cl, Br, I) lower alkynyl of C_2 - C_6 , lower alkoxy of C_1 - C_6 , halogenated (F, Cl, Br, I) lower alkoxy of C_1 - C_6 , CO_2H , $\text{CO}_2\text{R}'$, CONH_2 , CONHR' , CONR'_2 , $\text{CH}=\text{CHCO}_2\text{H}$, or $\text{CH}=\text{CHCO}_2\text{R}'$,

wherein R' is an optionally substituted alkyl, which includes, but is not limited to, an optionally substituted C_{1-20} alkyl, an optionally substituted C_{1-10} alkyl, an optionally substituted lower alkyl; an optionally substituted cycloalkyl; an optionally substituted alkynyl of C_2 - C_6 , an optionally substituted lower alkenyl of C_2 - C_6 , or optionally substituted acyl, which includes but is not limited to $\text{C}(\text{O})$ alkyl, $\text{C}(\text{O})(\text{C}_{1-20}$ alkyl), $\text{C}(\text{O})(\text{C}_{1-10}$ alkyl), or $\text{C}(\text{O})(\text{lower alkyl})$ or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

R^{12} is an H, halogen (including F, Cl, Br, I), OH, OR' , SH, SR' , NH_2 , NHR' , NR'_2 , NO_2 lower alkyl of C_1 - C_6 , halogenated (F, Cl, Br, I) lower alkyl of C_1 - C_6 , lower alkenyl of C_2 - C_6 , halogenated (F, Cl, Br, I) lower alkenyl of C_2 - C_6 , lower alkynyl of C_2 - C_6 , halogenated (F, Cl, Br, I) lower alkynyl of C_2 - C_6 , lower alkoxy of C_1 - C_6 , halogenated (F, Cl, Br, I) lower alkoxy of C_1 - C_6 , CO_2H , $\text{CO}_2\text{R}'$, CONH_2 , CONHR' , CONR'_2 , $\text{CH}=\text{CHCO}_2\text{H}$, or $\text{CH}=\text{CHCO}_2\text{R}'$; with the proviso that when base is represented by the structure c with R^{11} being hydrogen, R^{12} is not a: (i) $-\text{C}\equiv\text{C}-\text{H}$, (ii) $-\text{C}=\text{CH}_2$, or (iii) $-\text{NO}_2$.

6. A use of

a compound, its stereoisomer, salt, hydrate, solvate, or crystalline form thereof, selected from among

- 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-phenyl methoxy-alanyl phosphate((S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester);
- 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(phenyl methoxy-valyl phosphate((S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-valyl phosphate((S)-2-[(4-bromophenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-

- pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 8 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-alanyl phosphate(((S)-2-((4-bromophenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester);
- 9 N⁴-(N,N-dimethylformamidinyl)-2'-deoxy-2'-fluoro-2'-C-methylcytidine;
- 10 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(phenyl methoxy-alanyl phosphate(((S)-2-[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester);
- 11 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(4-bromophenyl methoxy-valyl phosphate(2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-valyl phosphate((S)-2-((4-bromo-phenoxy)-[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 12 2'-deoxy-2'-fluoro-2'-C-methylcytidine 5-(phenyl methoxy-valyl phosphate((S)-2-[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-3-methyl-butyric acid methyl ester;
- 13 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid ethyl ester;
- 14 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-naphthalen-1-yloxy)-phosphorylamino]-propionic acid benzyl ester;
- 15 [(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-acetic acid methyl ester;
- 16 (S)-2-((2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-ethyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
- 17 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-naphthalen-1-yloxy)-phosphorylamino]-propionic acid methyl ester;
- 18 (S)-1-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphoryl]-pyrrolidine-2-carboxylic acid methyl ester;
- 19 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid butyl ester;
- 20 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino)-propionic acid benzyl ester;
- 21 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino]-propionic acid methyl ester;
- 22 (S)-2-((4-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
- 23 (S)-2-((3,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
- 24 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid sec-butyl ester;
- 25 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino)-propionic acid isopropyl ester;
- 26 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-methoxy-phenoxy)-phosphorylamino]-propionic acid butyl ester;
- 27 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino]-propionic acid ethyl ester;
- 28 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino]-propionic acid ethyl ester;
- 29 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino]-propionic acid benzyl ester;
- 30 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-methoxy-phenoxy)-phosphorylamino]-propionic acid isopropyl ester;
- 31 (S)-2-((2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino)-propionic acid benzyl ester;
- 32 (S)-2-((2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino)-propionic acid butyl ester;
- 33 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-p-tolyloxy)-phosphorylamino]-propionic acid isopropyl ester;
- 34 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino]-propionic acid butyl ester;
- 35 (S)-2-((3,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino)-propionic acid ethyl ester;
- 36 (S)-2-((2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino)-propionic acid isopropyl ester;
- 37 (S)-2-([(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-methoxy-phenoxy)-phosphorylamino]-propionic acid benzyl ester;

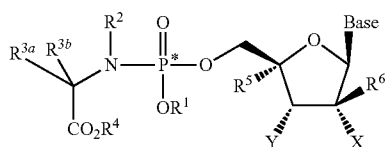
- 70 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-azetidin-1-yl-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 71 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-diethylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 72 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-propylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 73 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-cyclobutylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 74 (S)-2-[[[(2R,3R,4R,5R)-5-[2-Amino-6-(4-methyl-piperazin-1-yl)-purin-9-yl]-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid methyl ester;
- 75 (2S)-methyl 2-((((2R,3R,4R,5R)-4-fluoro-3-hydroxy-4-methyl-5-(6-(propylamino)-9H-purin-9-yl)tetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 76 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(cyclobutylamino)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 77 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6(cyclopentylamino)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 78 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(aziridin-1-yl)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 79 (2S)-methyl 2-((((2R,3R,4R,5R)-4-fluoro-3-hydroxy-4-methyl-5-(6-(piperidin-1-yl)-9H-purin-9-yl)tetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate; and
- 80 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(azetidin-1-yl)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate

in the manufacture of a medicament for the treatment of any condition the result of an infection by hepatitis C virus, West Nile virus, yellow fever virus, dengue virus, rhinovirus, polio virus, hepatitis A virus, bovine viral diarrhea virus or Japanese encephalitis virus.

7. A method of treatment in a subject in need thereof, which comprises:

administering a therapeutically effective amount of the compound represented by formula I to the subject;

wherein the compound or its stereoisomer, salt, hydrate, solvate, or crystalline form thereof represented by formula I:

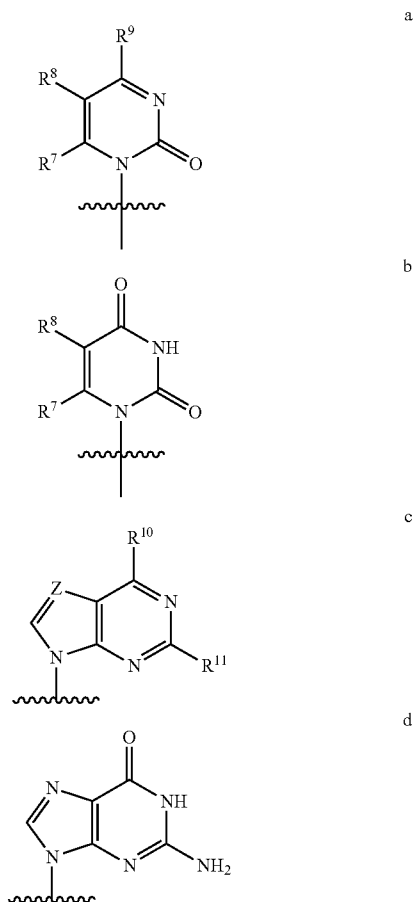


I

wherein

- (a) R^1 is hydrogen, n-alkyl; branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C_{1-6} alkyl, C_{2-6} alkenyl, C_{2-6} alkynyl, C_{1-6} alkoxy, F, Cl, Br, I, nitro, cyano, C_{1-6} haloalkyl, $-N(R^1)_2$, C_{1-6} acylamino, $-NHSO_2C_{1-6}$ alkyl, $-SO_2N(R^1)_2$, $COR^{1'}$, and $-SO_2C_{1-6}$ alkyl; (R^1 is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, $R^{1'}$ is $-OR'$ or $-N(R^{1'})_2$);
- (b) R^2 is hydrogen, C_{1-10} alkyl, R^{3a} or R^{3b} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms, $C(O)CR^{3a}R^{3b}NHR^1$, where n is 2 to 4 and R^1 , R^{3a} , and R^{3b} ;
- (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C_{1-10} alkyl, cycloalkyl, $-(CH_2)_c(NR^3)_2$, C_{1-6} hydroxyalkyl, $-CH_2SH$, $-(CH_2)_2S(O)_2Me$, $-(CH_2)_3NHC(=NH)NH_2$, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, $-(CH_2)_eCOR^{3'}$, aryl and aryl C_{1-3} alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C_{1-10} alkyl, C_{1-6} alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C_{1-6} alkyl; (iii) R^{3a} and R^{3b} together are $(CH_2)_f$ so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R^2 together are $(CH_2)_n$ so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where $R^{3'}$ is independently hydrogen or C_{1-6} alkyl and $R^{3''}$ is $-OR'$ or $-N(R^{3'})_2$; (vi) R^{3a} is H and R^{3b} is H, CH_3 , CH_2CH_3 , $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl; or (viii) R^{3a} is CH_3 , $-CH_2CH_3$, $CH(CH_3)_2$, $CH_2CH(CH_3)_2$, $CH(CH_3)CH_2CH_3$, CH_2Ph , CH_2 -indol-3-yl, $-CH_2CH_2SCH_3$, CH_2CO_2H , $CH_2C(O)NH_2$, CH_2CH_2COOH , $CH_2CH_2C(O)NH_2$, $CH_2CH_2CH_2CH_2NH_2$, $-CH_2CH_2CH_2NHC(NH)NH_2$, CH_2 -imidazol-4-yl, CH_2OH , $CH(OH)CH_3$, $CH_2((4'-OH)-Ph)$, CH_2SH , or lower cycloalkyl and R^{3b} is H where $R^{3'}$ is independently hydrogen or alkyl, which includes, but is not limited to, C_{1-20} alkyl, C_{1-10} alkyl, or C_{1-6} alkyl, $R^{3''}$ is $-OR'$ or $-N(R^{3'})_2$);
- (d) R^4 is hydrogen, C_{1-10} alkyl, C_{1-10} alkyl optionally substituted with a lower alkyl, alkoxy, di(lower alkyl)-amino, or halogen, C_{1-10} haloalkyl, C_{3-10} cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;
- (e) R^5 is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., $-(CH_2)_pOH$, where p is 1-6, including hydroxyl methyl (CH_2OH), CH_2F , N_3 , CH_2CN , CH_2NH_2 , CH_2NHCH_3 , $CH_2N(CH_3)_2$, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R^6 is H, R^5 cannot be N_3 and when X is OH, R^6 is CH_3 or CH_2F and B is a purine base, R^5 cannot be H;
- (f) R^6 H, CH_3 , CH_2F , CHF_2 , CF_3 , F, or CN;
- (g) X is H, OH, F, OMe, halogen, NH_2 , or N_3 ;

(h) Y is OH, H, C₁₋₄ alkyl, C₂₋₄ alkenyl, C₂₋₄ alkynyl, vinyl, N₃, CN, Cl, Br, F, I, NO₂, OC(O)O(C₁₋₄ alkyl), OC(O)O(C₁₋₄ alkyl), OC(O)O(C₂₋₄ alkynyl), OC(O)O(C₂₋₄ alkenyl), OC₁₋₁₀ haloalkyl, O(aminoacyl), O(C₁₋₁₀ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₄ alkyl), S(C₂₋₄ alkynyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl), SO(C₁₋₄ alkyl), SO(C₂₋₄ alkynyl), SO(C₂₋₄ alkenyl), SO₂(C₁₋₄ acyl), SO₂(C₁₋₄ alkyl), SO₂(C₂₋₄ alkynyl), SO₂(C₂₋₄ alkenyl), OS(O)₂(C₁₋₄ acyl), OS(O)₂(C₁₋₄ alkyl), OS(O)₂(C₂₋₄ alkynyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkenyl), NH(C₂₋₄ alkynyl), NH(C₁₋₄ acyl), N(C₁₋₄ alkyl)₂, N(C₁₋₁₈ acyl)₂, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N₃, CN, one to three halogen (Cl, Br, F, I), NO₂, C(O)O(C₁₋₄ alkyl), C(O)O(C₁₋₄ alkyl), C(O)O(C₂₋₄ alkynyl), C(O)O(C₂₋₄ alkenyl), O(C₁₋₄ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₄ alkyl), S(C₂₋₄ alkynyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl), SO(C₁₋₄ alkyl), SO(C₂₋₄ alkynyl), SO(C₂₋₄ alkenyl), SO₂(C₁₋₄ acyl), SO₂(C₁₋₄ alkyl), SO₂(C₂₋₄ alkynyl), SO₂(C₂₋₄ alkenyl), OS(O)₂(C₁₋₄ acyl), OS(O)₂(C₁₋₄ alkyl), OS(O)₂(C₂₋₄ alkynyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkenyl), NH(C₂₋₄ alkynyl), NH(C₁₋₄ acyl), N(C₁₋₄ alkyl)₂, N(C₁₋₄ acyl)₂; the base is a naturally occurring or modified purine or pyrimidine base represented by the following structures:



wherein

Z is N or CR¹²;

R⁷, R⁸, R⁹, R¹⁰, and R¹¹ are independently H, F, Cl, Br, I, OH, OR', SH, SR', NH₂, NHR', NR'₂, lower alkyl of

C₁₋₆, halogenated (F, Cl, Br, I) lower alkyl of C₁₋₆, lower alkenyl of C₂₋₆, halogenated (F, Cl, Br, I) lower alkenyl of C₂₋₆, lower alkynyl of C₂₋₆ such as C≡CH, halogenated (F, Cl, Br, I) lower alkynyl of C₂₋₆, lower alkoxy of C₁₋₆, halogenated (F, Cl, Br, I) lower alkoxy of C₁₋₆, CO₂H, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R',

wherein R' is an optionally substituted alkyl, which includes, but is not limited to, an optionally substituted C₁₋₂₀ alkyl, an optionally substituted C₁₋₁₀ alkyl, an optionally substituted lower alkyl; an optionally substituted cycloalkyl; an optionally substituted alkynyl of C₂₋₆, an optionally substituted lower alkenyl of C₂₋₆, or optionally substituted acyl, which includes but is not limited to C(O) alkyl, C(O)(C₁₋₂₀ alkyl), C(O)(C₁₋₁₀ alkyl), or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

R¹² is an H, halogen (including F, Cl, Br, I), OH, OR', SH, SR', NH₂, NHR', NR'₂, NO₂ lower alkyl of C₁₋₆, halogenated (F, Cl, Br, I) lower alkyl of C₁₋₆, lower alkenyl of C₂₋₆, halogenated (F, Cl, Br, I) lower alkenyl of C₂₋₆, lower alkynyl of C₂₋₆, halogenated (F, Cl, Br, I) lower alkynyl of C₂₋₆, lower alkoxy of C₁₋₆, halogenated (F, Cl, Br, I) lower alkoxy of C₁₋₆, CO₂H, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R'; with the proviso that when base is represented by the structure c with R¹¹ being hydrogen, R¹² is not a: (i) —C≡C—H, (ii) —C≡CH₂, or (iii) —NO₂.

8. A method of treatment in a subject in need thereof, which comprises:

administering a therapeutically effective amount of a compound, its stereoisomer, salt, hydrate, solvate, or crystalline form thereof, selected from among

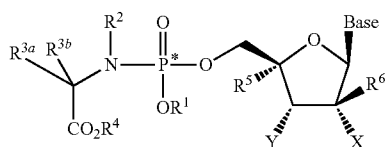
- 5 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-phenyl methoxy-alanyl phosphate((S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester);
- 6 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(phenyl methoxy-valyl phosphate((S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 7 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-valyl phosphate((S)-2-[(4-bromo-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 8 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-alanyl phosphate(((S)-2-[(4-bromo-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester);
- 9 N⁴-(N,N-dimethylformamidinyl)-2'-deoxy-2'-fluoro-2'-C-methylcytidine;
- 10 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(phenyl methoxy-alanyl phosphate(((S)-2-[(2R,3R,4R,5R)-5-

- (2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester);
- 11 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(4-bromophenyl methoxy-valyl phosphate(2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-valyl phosphate((S)-2-[(4-bromo-phenoxy)-[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 12 2'-deoxy-2'-fluoro-2'-C-methylcytidine 5-(phenyl methoxy-valyl phosphate((S)-2-[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-3-methyl-butyric acid methyl ester;
- 13 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid ethyl ester;
- 14 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[naphthalen-1-yloxy]-phosphorylamino}-propionic acid benzyl ester;
- 15 [(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-acetic acid methyl ester;
- 16 (S)-2-[(2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-ethyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
- 17 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[naphthalen-1-yloxy]-phosphorylamino}-propionic acid methyl ester;
- 18 (S)-1-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphoryl]-pyrrolidine-2-carboxylic acid methyl ester;
- 19 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid butyl ester;
- 20 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid benzyl ester;
- 21 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-fluoro-phenoxy]-phosphorylamino}-propionic acid methyl ester;
- 22 (S)-2-[(4-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
- 23 (S)-2-[(3,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
- 24 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid sec-butyl ester;
- 25 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid isopropyl ester;
- 26 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-y)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-methoxy-phenoxy]-phosphorylamino}-propionic acid butyl ester;
- 27 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-fluoro-phenoxy]-phosphorylamino}-propionic acid ethyl ester;
- 28 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-fluoro-phenoxy]-phosphorylamino}-propionic acid ethyl ester;
- 29 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-fluoro-phenoxy]-phosphorylamino}-propionic acid benzyl ester;
- 30 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-y)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-methoxy-phenoxy]-phosphorylamino}-propionic acid isopropyl ester;
- 31 (S)-2-[(2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid benzyl ester;
- 32 (S)-2-[(2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid butyl ester;
- 33 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-p-tolyl-oxy-phosphorylamino}-propionic acid isopropyl ester;
- 34 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-fluoro-phenoxy]-phosphorylamino}-propionic acid butyl ester;
- 35 (S)-2-[(3,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid ethyl ester;
- 36 (S)-2-[(2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid isopropyl ester;
- 37 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-methoxy-phenoxy]-phosphorylamino}-propionic acid benzyl ester;
- 38 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid pentyl ester;
- 39 (S)-2-[(4-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid isopropyl ester;
- 40 (S)-2-[(4-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid butyl ester;

- 41 (S)-2-[(4-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino]-propionic acid ethyl ester;
- 42 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-p-tolyloxy-phosphorylamino]-propionic acid butyl ester;
- 43 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-p-tolyloxy-phosphorylamino]-propionic acid benzyl ester;
- 44 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-butyric acid methyl ester;
- 45 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid 4-fluorobenzyl ester;
- 46 (S)-2-[(2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino]-propionic acid butyl ester;
- 47 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid 3-methyl-butyl ester;
- 48 (S)-2-[(2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino]-propionic acid ethyl ester;
- 49 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid cyclohexyl ester;
- 50 (R)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid butyl ester;
- 51 (R)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid isopropyl ester;
- 52 (R)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid benzyl ester;
- 53 (S)-2-[(2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino]-propionic acid butyl ester;
- 54 (S)-2-[(4-Bromo-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino]-propionic acid isopropyl ester;
- 55 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino]-propionic acid cyclohexyl ester;
- 56 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-bromo-phenoxy)-phosphorylamino]-propionic acid cyclohexyl ester;
- 57 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-butyric acid isopropyl ester;
- 58 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-butyric acid cyclohexyl ester;
- 59 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino]-butyric acid cyclohexyl ester;
- 60 (S)-2-[(4-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino]-butyric acid isopropyl ester;
- 61 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid 2,2-difluoro-ethyl ester;
- 62 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid 2,2,2-trifluoro-1-trifluoromethyl-ethyl ester;
- 63 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid 2-fluoro-1-fluoromethyl-ethyl ester;
- 64 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid cyclopropyl methyl ester;
- 65 (S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-butyric acid cyclopentyl ester;
- 66 (S)-2-[(2R,3R,4R,5R)-5-(6-Amino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid methyl ester;
- 67 (S)-2-[(2R,3R,4R,5R)-5-(2-Amino-6-hydroxy-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid methyl ester;
- 68 (S)-2-[(R)-[(2R,3R,4R,5R)-5-(2,6-Diamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid methyl ester;
- 69 (S)-2-[(2R,3R,4R,5R)-5-(2-Amino-6-cyclopentylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid methyl ester;
- 70 (S)-2-[(2R,3R,4R,5R)-5-(2-Amino-6-azetidin-1-yl-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid methyl ester;
- 71 (S)-2-[(2R,3R,4R,5R)-5-(2-Amino-6-diethylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid methyl ester;

- 72 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-propylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid methyl ester;
- 73 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-cyclobutylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid methyl ester;
- 74 (S)-2-[[[(2R,3R,4R,5R)-5-[2-Amino-6-(4-methyl-piperazin-1-yl)-purin-9-yl]-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid methyl ester;
- 75 (2S)-methyl 2-((((2R,3R,4R,5R)-4-fluoro-3-hydroxy-4-methyl-5-(6-(propylamino)-9H-purin-9-yl)tetrahydro-furan-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 76 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(cyclobutylamino)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 77 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6(cyclopentylamino)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 78 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(aziridin-1-yl)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 79 (2S)-methyl 2-((((2R,3R,4R,5R)-4-fluoro-3-hydroxy-4-methyl-5-(6-(piperidin-1-yl)-9H-purin-9-yl)tetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate; and
- 80 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(azetidin-1-yl)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate.

9. A process for preparing a compound, its stereoisomer, salt, hydrate, solvate, or crystalline form thereof, represented by formula I:

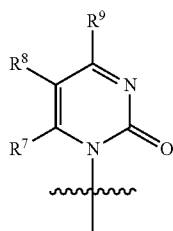


wherein

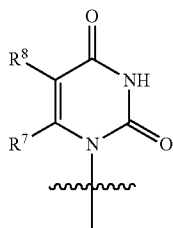
- (a) R¹ is hydrogen, n-alkyl; branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C₁-₆ alkyl, C₂-₆ alkenyl, C₁-₆ alkoxy, F, Cl, Br, I, nitro, cyano, C₁-₆ haloalkyl, —N(R¹)₂, C₁-₆ acylamino, —NHSO₂C₁-₆ alkyl, —SO₂N(R¹)₂, COR¹, and —SO₂C₁-₆ alkyl; (R¹ is independently hydrogen or alkyl, which includes, but is not limited to, C₁-₂₀ alkyl, C₁-₁₀ alkyl, or C₁-₆ alkyl, R¹ is —OR¹ or —N(R¹)₂);
- (b) R² is hydrogen, C₁-₁₀ alkyl, R³ᵃ or R³ᵇ and R² together are (CH₂)ₙ so as to form a cyclic ring that includes the adjoining N and C atoms, C(O)CR³ᵃR³ᵇNHR, where n is 2 to 4 and R¹, R³ᵃ, and R³ᵇ,

- (c) R³ᵃ and R³ᵇ are (i) independently selected from hydrogen, C₁-₁₀ alkyl, cycloalkyl, —(CH₂)ₑ(NR³')₂, C₁-₆ hydroxyalkyl, —CH₂SH, —(CH₂)₂S(O)ₐMe, —(CH₂)₃NHC(=NH)NH₂, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, —(CH₂)ₑCOR³'', aryl and aryl C₁-₃ alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C₁-₁₀ alkyl, C₁-₆ alkoxy, halogen, nitro and cyano; (ii) R³ᵃ and R³ᵇ both are C₁-₆ alkyl; (iii) R³ᵃ and R³ᵇ together are (CH₂)ₑ so as to form a spiro ring; (iv) R³ᵃ is hydrogen and R³ᵇ and R² together are (CH₂)ₙ so as to form a cyclic ring that includes the adjoining N and C atoms (v) R³ᵇ is hydrogen and R³ᵃ and R² together are (CH₂)ₙ so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where R³' is independently hydrogen or C₁-₆ alkyl and R³'' is —OR' or —N(R³')₂; (vi) R³ᵃ is H and R³ᵇ is H, CH₃, CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl; or (viii) R³ᵃ is CH₃, —CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl and R³ᵇ is H where R³' is independently hydrogen or alkyl, which includes, but is not limited to, C₁-₂₀ alkyl, C₁-₁₀ alkyl, or C₁-₆ alkyl, R³'' is —OR' or —N(R³')₂);
- (d) R⁴ is hydrogen, C₁-₁₀ alkyl, C₁-₁₀ alkyl optionally substituted with a lower alkyl, alkoxy, di(lower alkyl)-amino, or halogen, C₁-₁₀ haloalkyl, C₃-₁₀ cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;
- (e) R⁵ is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., —(CH₂)ₚOH, where p is 1-6, including hydroxyl methyl (CH₂OH), CH₂F, N₃, CH₂CN, CH₂NH₂, CH₂NHCH₃, CH₂N(CH₃)₂, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R⁶ is H, R⁵ cannot be N₃ and when X is OH, R⁶ is CH₃ or CH₂F and B is a purine base, R⁵ cannot be H;
- (f) R⁶ H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;
- (g) X is H, OH, F, OMe, halogen, NH₂, or N₃;
- (h) Y is OH, H, C₁-₄ alkyl, C₂-₄ alkenyl, C₂-₄ alkynyl, vinyl, N₃, CN, Cl, Br, F, I, NO₂, OC(O)O(C₁-₄ alkyl), OC(O)O(C₁-₄ alkyl), OC(O)O(C₂-₄ alkynyl), OC(O)O(C₂-₄ alkenyl), OC₁-₁₀ haloalkyl, O(aminoacyl), O(C₁-₁₀ acyl), O(C₁-₄ alkyl), O(C₂-₄ alkenyl), S(C₁-₄ alkyl), S(C₂-₄ alkynyl), S(C₂-₄ alkenyl), SO(C₁-₄ acyl), SO(C₁-₄ alkyl), SO(C₂-₄ alkynyl), SO(C₂-₄ alkenyl), SO₂(C₁-₄ acyl), SO₂(C₁-₄ alkyl), SO₂(C₂-₄ alkynyl), SO₂(C₂-₄ alkenyl), OS(O)₂(C₁-₄ acyl), OS(O)₂(C₁-₄ alkyl), OS(O)₂(C₂-₄ alkenyl), NH₂, NH(C₁-₄ alkyl), NH(C₂-₄ alkenyl), NH(C₂-₄ alkynyl), NH(C₁-₄ acyl), N(C₁-₄ alkyl)₂, N(C₁-₁₈ acyl)₂, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N₃, CN, one to three halogen (Cl, Br, F, I), NO₂, C(O)O(C₁-₄ alkyl), C(O)O(C₁-₄ alkyl), C(O)O(C₂-₄ alkynyl),

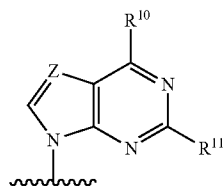
C(O)O(C₂₋₄ alkenyl), O(C₁₋₄ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₄ alkyl), S(C₂₋₄ alkenyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl), SO(C₁₋₄ alkyl), SO(C₂₋₄ alkenyl), SO(C₂₋₄ alkenyl), SO₂(C₁₋₄ acyl), SO₂(C₁₋₄ alkyl), SO₂(C₂₋₄ alkenyl), SO₂(C₂₋₄ alkenyl), OS(O)₂(C₁₋₄ acyl), OS(O)₂(C₁₋₄ alkyl), OS(O)₂(C₂₋₄ alkenyl), NH₂, NH(C₁₋₄ alkyl), NH(C₂₋₄ alkenyl), NH(C₂₋₄ alkenyl), NH(C₁₋₄ acyl), N(C₁₋₄ alkyl)₂, N(C₁₋₄ acyl)₂; the base is a naturally occurring or modified purine or pyrimidine base represented by the following structures;



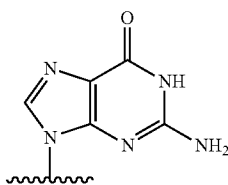
a



b



c



d

wherein

Z is N or CR¹²;

R⁷, R⁸, R⁹, R¹⁰, and R¹¹ are independently H, F, Cl, Br, I, OH, OR', SH, SR', NH₂, NHR', NR'₂, lower alkyl of C₁-C₆, halogenated (F, Cl, Br, I) lower alkyl of C₁-C₆, lower alkenyl of C₂-C₆, halogenated (F, Cl, Br, I) lower alkenyl of C₂-C₆, lower alkynyl of C₂-C₆ such as C≡CH, halogenated (F, Cl, Br, I) lower alkynyl of C₂-C₆, lower alkoxy of C₁-C₆, halogenated (F, Cl, Br, I) lower alkoxy of C₁-C₆, CO₂H, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H, or CH=CHCO₂R',

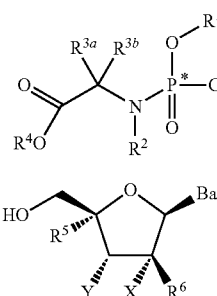
wherein R' is an optionally substituted alkyl, which includes, but is not limited to, an optionally substituted C₁₋₂₀ alkyl, an optionally substituted C₁₋₁₀ alkyl, an optionally substituted lower alkyl; an optionally substituted cycloalkyl; an optionally substituted alkynyl of

C₂-C₆, an optionally substituted lower alkenyl of C₂-C₆, or optionally substituted acyl, which includes but is not limited to C(O) alkyl, C(O)(C₁₋₂₀ alkyl), C(O)(C₁₋₁₀ alkyl), or C(O)(lower alkyl) or alternatively, in the instance of NR'₂, each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

R¹² is an H, halogen (including F, Cl, Br, I), OH, OR', SH, SR', NH₂, NHR', NR'₂, NO₂ lower alkyl of C₁-C₆, halogenated (F, Cl, Br, I) lower alkyl of C₁-C₆, lower alkenyl of C₂-C₆, halogenated (F, Cl, Br, I) lower alkenyl of C₂-C₆, lower alkynyl of C₂-C₆, halogenated (F, Cl, Br, I) lower alkynyl of C₂-C₆, lower alkoxy of C₁-C₆, halogenated (F, Cl, Br, I) lower alkoxy of C₁-C₆, CO₂H, CO₂R', CONH₂, CONHR', CONR'₂, CH=CHCO₂H or CH=CHCO₂R'; with the proviso that when base is represented by the structure c with R¹¹ being hydrogen, R¹² is not a: (i) —C≡C—H, (ii) —C=CH₂, or (iii) —NO₂;

said process comprising:

reacting a substituted phosphochloridate compound 4 with a nucleoside analog 5



4

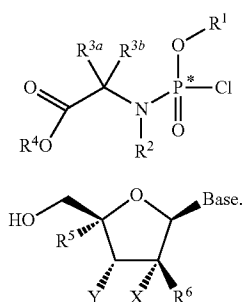
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10. A process for preparing a compound, its stereoisomer, salt, hydrate, solvate, or crystalline form thereof, selected from among

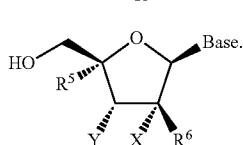
- 5 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-phenyl methoxy-alanyl phosphate((S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester);
- 6 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(phenyl methoxy-valyl phosphate((S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 7 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-valyl phosphate((S)-2-[(4-bromophenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 8 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-alanyl phosphate((S)-2-[(4-bromophenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester);

- 9 N⁴-(N,N-dimethylformamidinyl)-2'-deoxy-2'-fluoro-2'-C-methylcytidine;
- 10 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(phenyl methoxy-alanyl phosphate(((S)-2-{[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester);
- 11 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(4-bromophenyl methoxy-valyl phosphate(2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-valyl phosphate((S)-2-{(4-bromo-phenoxy)-[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 12 2'-deoxy-2'-fluoro-2'-C-methylcytidine 5'-(phenyl methoxy-valyl phosphate((S)-2-{[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-3-methyl-butyric acid methyl ester;
- 13 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid ethyl ester;
- 14 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(naphthalen-1-yloxy)-phosphorylamino}-propionic acid benzyl ester;
- 15 {[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-acetic acid methyl ester;
- 16 (S)-2-{(2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
- 17 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(naphthalen-1-yloxy)-phosphorylamino}-propionic acid methyl ester;
- 18 (S)-1-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphoryl}-pyrrolidine-2-carboxylic acid methyl ester;
- 19 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid butyl ester;
- 20 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid benzyl ester;
- 21 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino}-propionic acid methyl ester;
- 22 (S)-2-{(4-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
- 23 (S)-2-{(3,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
- 24 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid sec-butyl ester;
- 25 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid isopropyl ester;
- 26 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-methoxy-phenoxy)-phosphorylamino}-propionic acid butyl ester;
- 27 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino}-propionic acid ethyl ester;
- 28 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino}-propionic acid ethyl ester;
- 29 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino}-propionic acid benzyl ester;
- 30 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-methoxy-phenoxy)-phosphorylamino}-propionic acid isopropyl ester;
- 31 (S)-2-{(2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid benzyl ester;
- 32 (S)-2-{(2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid butyl ester;
- 33 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-p-tolylloxy-phosphorylamino}-propionic acid isopropyl ester;
- 34 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-fluoro-phenoxy)-phosphorylamino}-propionic acid butyl ester;
- 35 (S)-2-{(3,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid ethyl ester;
- 36 (S)-2-{(2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid isopropyl ester;
- 37 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-(4-methoxy-phenoxy)-phosphorylamino}-propionic acid benzyl ester;
- 38 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid pentyl ester;
- 39 (S)-2-{(4-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid isopropyl ester;

- tetrahydro-furan-2-ylmethoxy}-phenoxy-phosphorylamino)-propionic acid methyl ester;
- 75 (2S)-methyl 2-((((2R,3R,4R,5R)-4-fluoro-3-hydroxy-4-methyl-5-(6-(propylamino)-9H-purin-9-yl)tetrahydro-furan-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 76 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(cyclobutylamino)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 77 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6(cyclopentylamino)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 78 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(aziridin-1-yl)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 79 (2S)-methyl 2-((((2R,3R,4R,5R)-4-fluoro-3-hydroxy-4-methyl-5-(6-(piperidin-1-yl)-9H-purin-9-yl)tetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate; and
- 80 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(azetidin-1-yl)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate
- said process comprising:
- reacting a substituted phosphochloridate compound 4 with a nucleoside analog 5

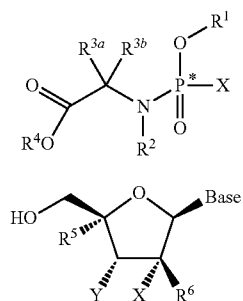


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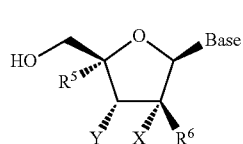


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11. A product its stereoisomer, salt; hydrate, solvate or crystalline form thereof, prepared a process comprising:
- reacting a substituted phosphochloridate compound 4 with a nucleoside analog 5



4



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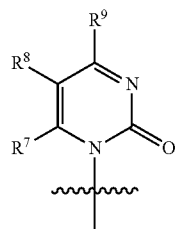
wherein

- (a) R¹ is hydrogen, n-alkyl; branched alkyl; cycloalkyl; or aryl, which includes, but is not limited to, phenyl or naphthyl, where phenyl or naphthyl are optionally substituted with at least one of C₁₋₆ alkyl, C₂₋₆ alkenyl, C₂₋₆ alkynyl, C₁₋₆ alkoxy, F, Cl, Br, I, nitro, cyano, C₁₋₆ haloalkyl, —N(R^{1'})₂, C₁₋₆ acylamino, —NH—SO₂C₁₋₆

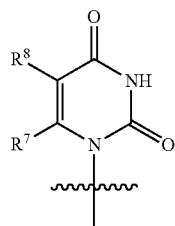
alkyl, —SO₂N(R^{1'})₂, COR^{1'}, and —SO₂C₁₋₆ alkyl; (R^{1'} is independently hydrogen or alkyl, which includes, but is not limited to, C₁₋₂₀ alkyl, C₁₋₁₀ alkyl, or C₁₋₆ alkyl, R^{1'} is —OR' or —N(R^{1'})₂);

- (b) R² is hydrogen, C₁₋₁₀ alkyl, R^{3a} or R^{3b} and R² together are (CH₂)_n so as to form a cyclic ring that includes the adjoining N and C atoms, C(O)CR^{3a}R^{3b}NHR¹, where n is 2 to 4 and R¹, R^{3a}, and R^{3b};
- (c) R^{3a} and R^{3b} are (i) independently selected from hydrogen, C₁₋₁₀ alkyl, cycloalkyl, —(CH₂)_c(NR^{3'})₂, C₁₋₆ hydroxyalkyl, —CH₂SH, —(CH₂)₂S(O)_aMe, —(CH₂)₃NHC(=NH)NH₂, (1H-indol-3-yl)methyl, (1H-imidazol-4-yl)methyl, —(CH₂)_eCOR^{3''}, aryl and aryl C₁₋₃ alkyl, said aryl groups optionally substituted with a group selected from hydroxyl, C₁₋₁₀ alkyl, C₁₋₆ alkoxy, halogen, nitro and cyano; (ii) R^{3a} and R^{3b} both are C₁₋₆ alkyl; (iii) R^{3a} and R^{3b} together are (CH₂)_f so as to form a spiro ring; (iv) R^{3a} is hydrogen and R^{3b} and R² together are (CH₂)_n so as to form a cyclic ring that includes the adjoining N and C atoms (v) R^{3b} is hydrogen and R^{3a} and R² together are (CH₂)_n so as to form a cyclic ring that includes the adjoining N and C atoms, where c is 1 to 6, d is 0 to 2, e is 0 to 3, f is 2 to 5, n is 2 to 4, and where R^{3'} is independently hydrogen or C₁₋₆ alkyl and R^{3''} is —OR' or —N(R^{3'})₂; (vi) R^{3a} is H and R^{3b} is H, CH₃, CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl; or (viii) R^{3a} is CH₃, —CH₂CH₃, CH(CH₃)₂, CH₂CH(CH₃)₂, CH(CH₃)CH₂CH₃, CH₂Ph, CH₂-indol-3-yl, —CH₂CH₂SCH₃, CH₂CO₂H, CH₂C(O)NH₂, CH₂CH₂COOH, CH₂CH₂C(O)NH₂, CH₂CH₂CH₂NH₂, —CH₂CH₂CH₂NHC(NH)NH₂, CH₂-imidazol-4-yl, CH₂OH, CH(OH)CH₃, CH₂((4'-OH)-Ph), CH₂SH, or lower cycloalkyl and R^{3b} is H where R^{3'} is independently hydrogen or alkyl, which includes, but is not limited to, C₁₋₂₀ alkyl, C₁₋₁₀ alkyl, or C₁₋₆ alkyl, R^{3''} is —OR' or —N(R^{3'})₂);
- (d) R⁴ is hydrogen, C₁₋₁₀ alkyl, C₁₋₁₀ alkyl optionally substituted with a lower alkyl, alkoxy, di(lower alkyl)-amino, or halogen, C₁₋₁₀ haloalkyl, C₃₋₁₀ cycloalkyl, cycloalkyl alkyl, cycloheteroalkyl, aminoacyl, aryl, such as phenyl, heteroaryl, such as, pyridinyl, substituted aryl, or substituted heteroaryl;
- (e) R⁵ is H, a lower alkyl, CN, vinyl, O-(lower alkyl), hydroxyl lower alkyl, i.e., —(CH₂)_pOH, where p is 1-6, including hydroxyl methyl (CH₂OH), CH₂F, N₃, CH₂CN, CH₂NH₂, CH₂NHCH₃, CH₂N(CH₃)₂, alkyne (optionally substituted), or halogen, including F, Cl, Br, or I, with the provisos that when X is OH, base is cytosine and R⁶ is H, R⁵ cannot be N₃ and when X is OH, R⁶ is CH₃ or CH₂F and B is a purine base, R⁵ cannot be H;
- (f) R⁶ is H, CH₃, CH₂F, CHF₂, CF₃, F, or CN;
- (g) X is H, OH, F, OMe, halogen, NH₂, or N₃;
- (h) Y is OH, H, C₁₋₄ alkyl, C₂₋₄ alkenyl, C₂₋₄ alkynyl, vinyl, N₃, CN, Cl, Br, F, I, NO₂, OC(O)O(C₁₋₄ alkyl), OC(O)O(C₁₋₄ alkyl), OC(O)O(C₂₋₄ alkynyl), OC(O)O(C₂₋₄ alkenyl), OC₁₋₁₀ haloalkyl, O(aminoacyl), O(C₁₋₁₀ acyl), O(C₁₋₄ alkyl), O(C₂₋₄ alkenyl), S(C₁₋₄ acyl), S(C₁₋₄ alkyl), S(C₂₋₄ alkynyl), S(C₂₋₄ alkenyl), SO(C₁₋₄ acyl), SO(C₁₋₄ alkyl), SO(C₂₋₄ alkynyl), SO(C₂₋₄ alk-

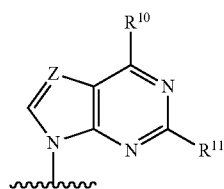
enyl), $\text{SO}_2(\text{C}_{1-4} \text{ acyl})$, $\text{SO}_2(\text{C}_{1-4} \text{ alkyl})$, $\text{SO}_2(\text{C}_{2-4} \text{ alkynyl})$, $\text{SO}_2(\text{C}_{2-4} \text{ alkenyl})$, $\text{OS}(\text{O})_2(\text{C}_{1-4} \text{ acyl})$, $\text{OS}(\text{O})_2(\text{C}_{1-4} \text{ alkyl})$, $\text{OS}(\text{O})_2(\text{C}_{2-4} \text{ alkynyl})$, NH_2 , $\text{NH}(\text{C}_{1-4} \text{ alkyl})$, $\text{NH}(\text{C}_{2-4} \text{ alkenyl})$, $\text{NH}(\text{C}_{2-4} \text{ alkynyl})$, $\text{NH}(\text{C}_{1-4} \text{ acyl})$, $\text{N}(\text{C}_{1-4} \text{ alkyl})_2$, $\text{N}(\text{C}_{1-18} \text{ acyl})_2$, wherein alkyl, alkynyl, alkenyl and vinyl are optionally substituted by N_3 , CN , one to three halogen (Cl, Br, F, I), NO_2 , $\text{C}(\text{O})\text{O}(\text{C}_{1-4} \text{ alkyl})$, $\text{C}(\text{O})\text{O}(\text{C}_{1-4} \text{ alkyl})$, $\text{C}(\text{O})\text{O}(\text{C}_{2-4} \text{ alkynyl})$, $\text{C}(\text{O})\text{O}(\text{C}_{2-4} \text{ alkenyl})$, $\text{O}(\text{C}_{1-4} \text{ acyl})$, $\text{O}(\text{C}_{1-4} \text{ alkyl})$, $\text{O}(\text{C}_{2-4} \text{ alkenyl})$, $\text{S}(\text{C}_{1-4} \text{ acyl})$, $\text{S}(\text{C}_{1-4} \text{ alkyl})$, $\text{S}(\text{C}_{2-4} \text{ alkynyl})$, $\text{S}(\text{C}_{2-4} \text{ alkenyl})$, $\text{SO}(\text{C}_{1-4} \text{ acyl})$, $\text{SO}(\text{C}_{1-4} \text{ alkyl})$, $\text{SO}(\text{C}_{2-4} \text{ alkynyl})$, $\text{SO}(\text{C}_{2-4} \text{ alkenyl})$, $\text{SO}_2(\text{C}_{1-4} \text{ acyl})$, $\text{SO}_2(\text{C}_{1-4} \text{ alkyl})$, $\text{SO}_2(\text{C}_{2-4} \text{ alkynyl})$, $\text{SO}_2(\text{C}_{2-4} \text{ alkenyl})$, $\text{OS}(\text{O})_2(\text{C}_{1-4} \text{ acyl})$, $\text{OS}(\text{O})_2(\text{C}_{1-4} \text{ alkyl})$, $\text{OS}(\text{O})_2(\text{C}_{2-4} \text{ alkynyl})$, NH_2 , $\text{NH}(\text{C}_{1-4} \text{ alkyl})$, $\text{NH}(\text{C}_{2-4} \text{ alkenyl})$, $\text{NH}(\text{C}_{2-4} \text{ alkynyl})$, $\text{NH}(\text{C}_{1-4} \text{ acyl})$, $\text{N}(\text{C}_{1-4} \text{ alkyl})_2$, $\text{N}(\text{C}_{1-4} \text{ acyl})_2$; the base is a naturally occurring or modified purine or pyrimidine base represented by the following structures:



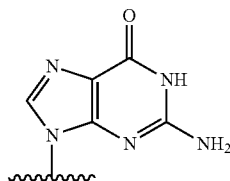
a



b



c



d

wherein

Z is N or CR^{12} ;

R^7 , R^8 , R^{10} , and R^{11} are independently H, F, Cl, Br, I, OH, OR' , SH, SR' , NHR' , NR'_2 , lower alkyl of $\text{C}_1\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkyl of $\text{C}_1\text{-C}_6$, lower alkenyl of $\text{C}_2\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkenyl of $\text{C}_2\text{-C}_6$, lower alkynyl of $\text{C}_2\text{-C}_6$ such as $\text{C}\equiv\text{CH}$, halogenated (F, Cl, Br, I) lower alkynyl of $\text{C}_2\text{-C}_6$, lower alkoxy of $\text{C}_1\text{-C}_6$, halogenated (F, Cl, Br, I)

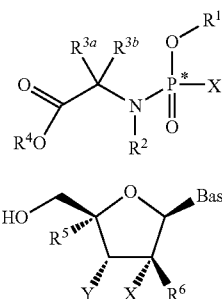
lower alkoxy of $\text{C}_1\text{-C}_6$, CO_2H , $\text{CO}_2\text{R}'$, CONH_2 , CONHR' , CONR'_2 , $\text{CH}=\text{CHCO}_2\text{H}$, or $\text{CH}=\text{CHCO}_2\text{R}'$;

wherein R' is an optionally substituted alkyl, which includes, but is not limited to, an optionally substituted C_{1-20} alkyl, an optionally substituted C_{1-10} alkyl, an optionally substituted lower alkyl; an optionally substituted cycloalkyl; an optionally substituted alkynyl of $\text{C}_2\text{-C}_6$, an optionally substituted lower alkenyl of $\text{C}_2\text{-C}_6$, or optionally substituted acyl, which includes but is not limited to $\text{C}(\text{O})$ alkyl, $\text{C}(\text{O})(\text{C}_{1-20} \text{ alkyl})$, $\text{C}(\text{O})(\text{C}_{1-10} \text{ alkyl})$, or $\text{C}(\text{O})(\text{lower alkyl})$ or alternatively, in the instance of NR'_2 , each R' comprise at least one C atom that are joined to form a heterocycle comprising at least two carbon atoms; and

R^{12} is an H, halogen (including F, Cl, Br, I), OH, OR' , SH, SR' , NH_2 , NHR' , NR'_2 , NO_2 lower alkyl of $\text{C}_1\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkyl of $\text{C}_1\text{-C}_6$, lower alkenyl of $\text{C}_2\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkenyl of $\text{C}_2\text{-C}_6$, lower alkynyl of $\text{C}_2\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkynyl of $\text{C}_2\text{-C}_6$, lower alkoxy of $\text{C}_1\text{-C}_6$, halogenated (F, Cl, Br, I) lower alkoxy of $\text{C}_1\text{-C}_6$, CO_2H , $\text{CO}_2\text{R}'$, CONH_2 , CONHR' , CONR'_2 , $\text{CH}=\text{CHCO}_2\text{H}$, or $\text{CH}=\text{CHCO}_2\text{R}'$; with the proviso that when base is represented by the structure c with R^{11} being hydrogen, R^{12} is not at: (i) $\text{—C}\equiv\text{C—H}$, (ii) —C=CH_2 , or (iii) —NO_2 and

wherein X' is a leaving group, such as, Cl.

12. A product its stereoisomer, salt, hydrate, solvate or crystalline form thereof, prepared a process comprising: reacting a substituted phosphochloridate compound 4 with a nucleoside analog 5



4

5

wherein the product comprises a compound, its stereoisomer, salt, hydrate, solvate, or crystalline form thereof, selected from among

- 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-phenyl methoxy-alanyl phosphate((S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester);
- 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(phenyl methoxy-valyl phosphate((S)-2-[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-valyl phosphate((S)-2-[(4-bromophenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-

- tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-3-methyl-butyric acid methyl ester);
- 8 2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-alanyl phosphate(((S)-2-{(4-bromo-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester);
 - 9 N⁴-(N,N-dimethylformamidinyl)-2'-deoxy-2'-fluoro-2'-C-methylcytidine;
 - 10 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(phenyl methoxy-alanyl phosphate(((S)-2-{[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester);
 - 11 2'-Deoxy-2'-fluoro-2'-C-methylcytidine 5'-(4-bromophenyl methoxy-valyl phosphate(2'-Deoxy-2'-fluoro-2'-C-methyluridine 5'-(4-bromophenyl methoxy-valyl phosphate((S)-2-{(4-bromo-phenoxy)-[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-3-methyl-butyric acid methyl ester);
 - 12 2'-deoxy-2'-fluoro-2'-C-methylcytidine 5-(phenyl methoxy-valyl phosphate((S)-2-{[(2R,3R,4R,5R)-5-(2-oxo-3-amino-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-3-methyl-butyric acid methyl ester;
 - 13 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid ethyl ester;
 - 14 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[naphthalen-1-yloxy]-phosphorylamino}-propionic acid benzyl ester;
 - 15 {[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-acetic acid methyl ester;
 - 16 (S)-2-{(2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-ethyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
 - 17 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[naphthalen-1-yloxy]-phosphorylamino}-propionic acid methyl ester;
 - 18 (S)-1-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphoryl}-pyrrolidine-2-carboxylic acid methyl ester;
 - 19 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid butyl ester;
 - 20 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid benzyl ester;
 - 21 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-fluoro-phenoxy]-phosphorylamino}-propionic acid methyl ester;
 - 22 (S)-2-{(4-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
 - 23 (S)-2-{(3,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid methyl ester;
 - 24 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid sec-butyl ester;
 - 25 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid isopropyl ester;
 - 27 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-fluoro-phenoxy]-phosphorylamino}-propionic acid ethyl ester;
 - 28 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-fluoro-phenoxy]-phosphorylamino}-propionic acid ethyl ester;
 - 29 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-fluoro-phenoxy]-phosphorylamino}-propionic acid benzyl ester;
 - 30 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-methoxy-phenoxy]-phosphorylamino}-propionic acid isopropyl ester;
 - 31 (S)-2-{(2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid benzyl ester;
 - 32 (S)-2-{(2,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid butyl ester;
 - 33 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-p-tolyloxy-phosphorylamino}-propionic acid isopropyl ester;
 - 34 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-fluoro-phenoxy]-phosphorylamino}-propionic acid butyl ester;
 - 35 (S)-2-{(3,4-Dichloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid ethyl ester;
 - 36 (S)-2-{(2-Chloro-phenoxy)-[(2R,3R,4R,5R)-5-(2,4-dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phosphorylamino}-propionic acid isopropyl ester;
 - 37 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-[4-methoxy-phenoxy]-phosphorylamino}-propionic acid benzyl ester;
 - 38 (S)-2-{[(2R,3R,4R,5R)-5-(2,4-Dioxo-3,4-dihydro-2H-pyrimidin-1-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid pentyl ester;

- [illegible]

- 70 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-azetidin-1-yl-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 71 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-diethylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 72 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-propylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 73 (S)-2-[[[(2R,3R,4R,5R)-5-(2-Amino-6-cyclobutylamino-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-ylmethoxy]-phenoxy-phosphorylamino}-propionic acid methyl ester;
- 74 (S)-2-[[[(2R,3R,4R,5R)-5-[2-Amino-6-(4-methyl-piperazin-1-yl)-purin-9-yl]-4-fluoro-3-hydroxy-4-methyl-tetrahydrofuran-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid methyl ester;
- 75 (2S)-methyl 2-((((2R,3R,4R,5R)-4-fluoro-3-hydroxy-4-methyl-5-(6-(propylamino)-9H-purin-9-yl)tetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 76 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(cyclobutylamino)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 77 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6(cyclopentylamino)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 78 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(aziridin-1-yl)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate;
- 79 (2S)-methyl 2-((((2R,3R,4R,5R)-4-fluoro-3-hydroxy-4-methyl-5-(6-(piperidin-1-yl)-9H-purin-9-yl)tetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate; and
- 80 (2S)-methyl 2-((((2R,3R,4R,5R)-5-(6-(azetidin-1-yl)-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphorylamino)propanoate
- wherein Base' is as recited in each of compounds 5-74; and wherein X' is a leaving group, such as, Cl.
- * * * * *

(19) **United States**(12) **Patent Application Publication**
CHANG et al.(10) **Pub. No.: US 2011/0257121 A1**(43) **Pub. Date: Oct. 20, 2011**(54) **PURINE NUCLEOSIDE PHOSPHORAMIDATE**

filed on Mar. 31, 2010, provisional application No. 61/355,940, filed on Jun. 17, 2010.

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C07H 19/207 (2006.01)

(52) **U.S. Cl.** **514/48**; 536/26.7; 378/73(57) **ABSTRACT**

Disclosed herein is a compound represented by formula 1 or its hydrate thereof in crystalline or crystal-like form.

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PRINCETON, NJ (US)

(21) Appl. No.: **13/076,718**(22) Filed: **Mar. 31, 2011****Related U.S. Application Data**

(60) Provisional application No. 61/319,513, filed on Mar. 31, 2010, provisional application No. 61/319,548,

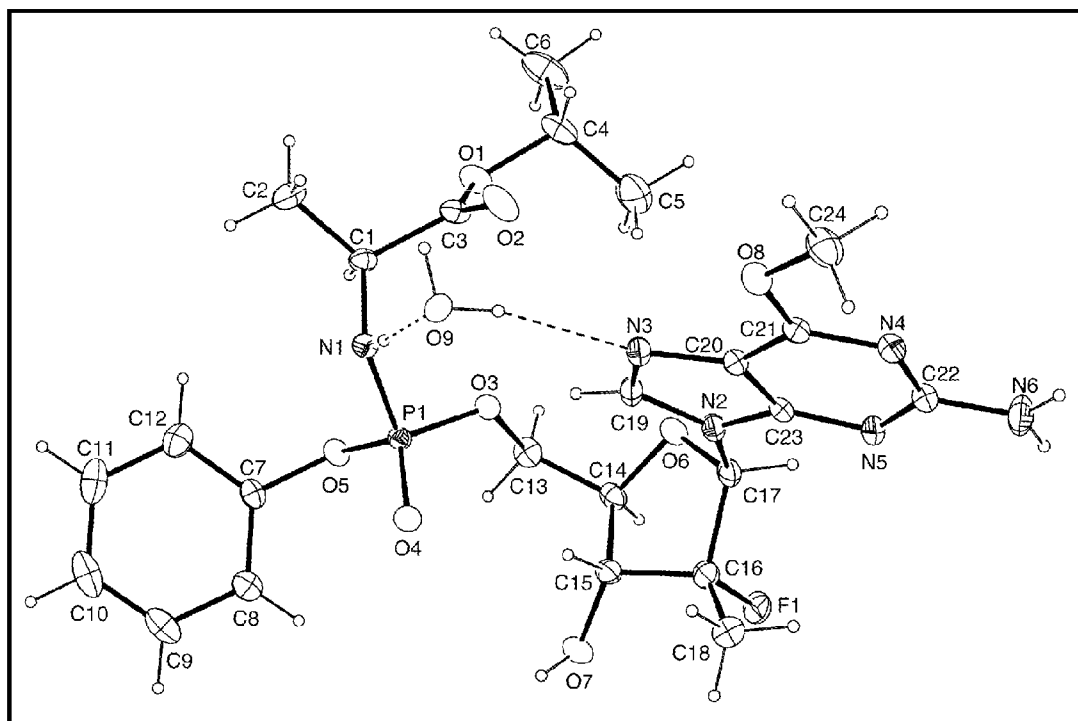
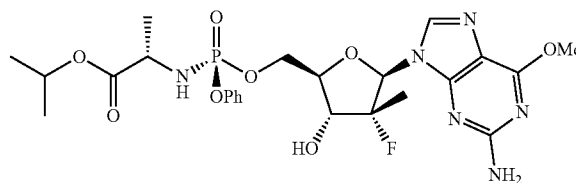
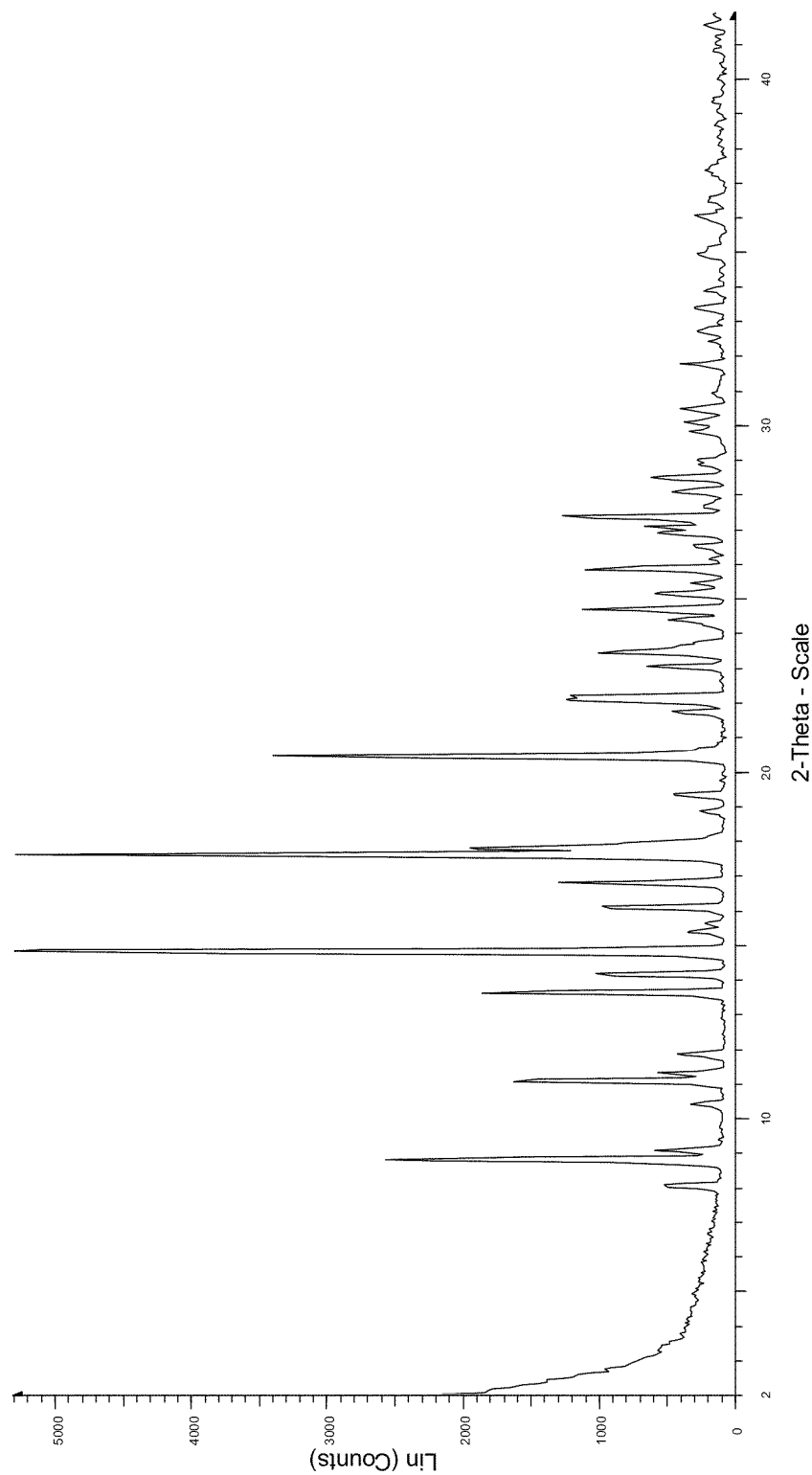
ORTEP drawing of **1·H₂O** with 30% probability thermal ellipsoids

Figure 1. High resolution XRPD of $1 \cdot H_2O$ 

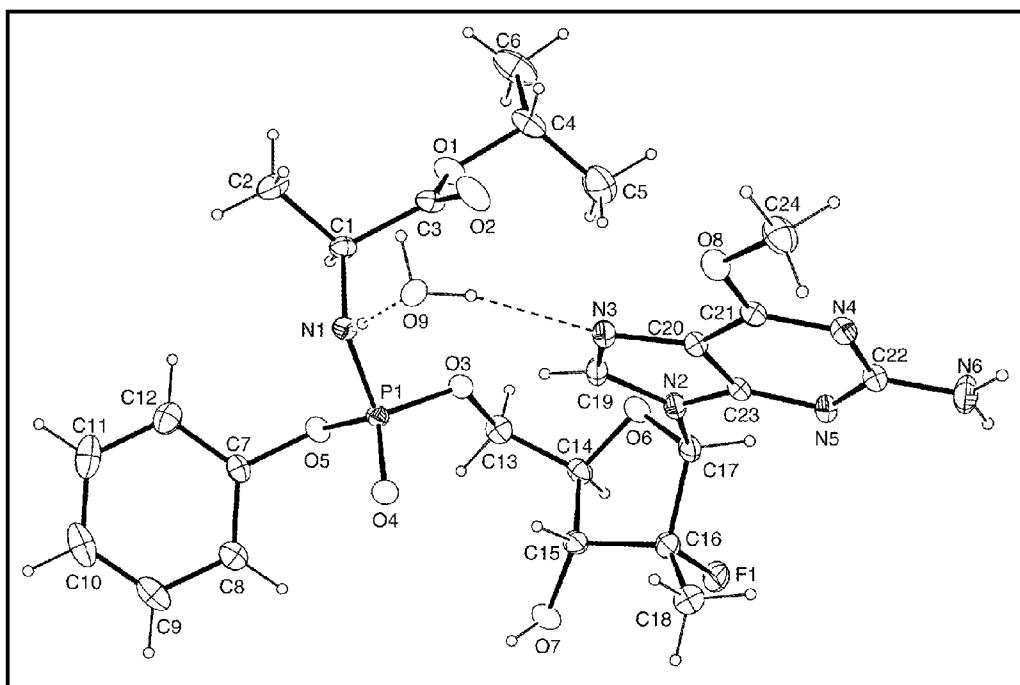


Fig 2. ORTEP drawing of 1·H₂O with 30% probability thermal ellipsoids

PURINE NUCLEOSIDE PHOSPHORAMIDATE**PRIORITY**

[0001] This application claims priority to U.S. 61/319,513, filed on Mar. 31, 2010; U.S. 61/319,548, filed on Mar. 31, 2010; and U.S. 61/355,940, filed on Jun. 17, 2010, the subject matter of which is incorporated by reference in its entirety.

FIELD OF THE INVENTION

[0002] Disclosed herein are nucleoside phosphoramidates and their use as agents for treating viral diseases. These compounds are inhibitors of RNA-dependent RNA viral replication and are useful as inhibitors of HCV NS5B polymerase, as inhibitors of HCV replication and for treatment of hepatitis C infection in mammals.

BACKGROUND

[0003] Hepatitis C virus (HCV) infection is a major health problem that leads to chronic liver disease, such as cirrhosis and hepatocellular carcinoma, in a substantial number of infected individuals, estimated to be 2-15% of the world's population. There are an estimated 4.5 million infected people in the United States alone, according to the U.S. Center for Disease Control. According to the World Health Organization, there are more than 200 million infected individuals worldwide, with at least 3 to 4 million people being infected each year. Once infected, about 20% of people clear the virus, but the rest can harbor HCV the rest of their lives. Ten to twenty percent of chronically infected individuals eventually develop liver-destroying cirrhosis or cancer. The viral disease is transmitted parenterally by contaminated blood and blood products, contaminated needles, or sexually and vertically from infected mothers or carrier mothers to their offspring. Current treatments for HCV infection, which are restricted to immunotherapy with recombinant interferon- α alone or in combination with the nucleoside analog ribavirin, are of limited clinical benefit. Moreover, there is no established vaccine for HCV. Consequently, there is an urgent need for improved therapeutic agents that effectively combat chronic HCV infection.

[0004] The HCV virion is an enveloped positive-strand RNA virus with a single oligoribonucleotide genomic sequence of about 9600 bases which encodes a polyprotein of about 3,010 amino acids. The protein products of the HCV gene consist of the structural proteins C, E1, and E2, and the non-structural proteins NS2, NS3, NS4A and NS4B, and NS5A and NS5B. The nonstructural (NS) proteins are believed to provide the catalytic machinery for viral replication. The NS3 protease releases NS5B, the RNA-dependent RNA polymerase from the polyprotein chain. HCV NS5B polymerase is required for the synthesis of a double-stranded RNA from a single-stranded viral RNA that serves as a template in the replication cycle of HCV. Therefore, NS5B polymerase is considered to be an essential component in the HCV replication complex (K. Ishi, et al, *Heptology*, 1999, 29: 1227-1235; V. Lohmann, et al., *Virology*, 1998, 249: 108-118). Inhibition of HCV NS5B polymerase prevents formation of the double-stranded HCV RNA and therefore constitutes an attractive approach to the development of HCV-specific antiviral therapies.

[0005] HCV belongs to a much larger family of viruses that share many common features.

Flaviviridae Viruses

[0006] The Flaviviridae family of viruses comprises at least three distinct genera: pestiviruses, which cause disease in cattle and pigs; flaviviruses, which are the primary cause of diseases such as dengue fever and yellow fever; and hepaciviruses, whose sole member is HCV. The flavivirus genus includes more than 68 members separated into groups on the basis of serological relatedness (Calisher et al., *J. Gen. Virol.*, 1993, 70, 37-43). Clinical symptoms vary and include fever, encephalitis and hemorrhagic fever (Fields Virology, Editors: Fields, B. N., Knipe, D. M., and Howley, P. M., Lippincott-Raven Publishers, Philadelphia, Pa., 1996, Chapter 31, 931-959). Flaviviruses of global concern that are associated with human disease include the Dengue Hemorrhagic Fever viruses (DHF), yellow fever virus, shock syndrome and Japanese encephalitis virus (Halstead, S. B., *Rev. Infect. Dis.*, 1984, 6, 251-264; Halstead, S. B., *Science*, 239:476-481, 1988; Monath, T. P., *New Eng. J. Med.*, 1988, 319, 641-643).

[0007] The pestivirus genus includes bovine viral diarrhea virus (BVDV), classical swine fever virus (CSFV, also called hog cholera virus) and border disease virus (BDV) of sheep (Moennig, V. et al. *Adv. Vir. Res.* 1992, 41, 53-98). Pestivirus infections of domesticated livestock (cattle, pigs and sheep) cause significant economic losses worldwide. BVDV causes mucosal disease in cattle and is of significant economic importance to the livestock industry (Meyers, G. and Thiel, H. J., *Advances in Virus Research*, 1996, 47, 53-118; Moennig V., et al, *Adv. Vir. Res.* 1992, 41, 53-98). Human pestiviruses have not been as extensively characterized as the animal pestiviruses. However, serological surveys indicate considerable pestivirus exposure in humans.

[0008] Pestiviruses and hepaciviruses are closely related virus groups within the Flaviviridae family. Other closely related viruses in this family include the GB virus A, GB virus A-like agents, GB virus-B and GB virus-C (also called hepatitis G virus, HGV). The hepacivirus group (hepatitis C virus; HCV) consists of a number of closely related but genotypically distinguishable viruses that infect humans. There are at least 6 HCV genotypes and more than 50 subtypes. Due to the similarities between pestiviruses and hepaciviruses, combined with the poor ability of hepaciviruses to grow efficiently in cell culture, bovine viral diarrhea virus (BVDV) is often used as a surrogate to study the HCV virus.

[0009] The genetic organization of pestiviruses and hepaciviruses is very similar. These positive stranded RNA viruses possess a single large open reading frame (ORF) encoding all the viral proteins necessary for virus replication. These proteins are expressed as a polyprotein that is co- and post-translationally processed by both cellular and virus-encoded proteinases to yield the mature viral proteins. The viral proteins responsible for the replication of the viral genome RNA are located within approximately the carboxy-terminal. Two-thirds of the ORF are termed nonstructural (NS) proteins. The genetic organization and polyprotein processing of the non-structural protein portion of the ORF for pestiviruses and hepaciviruses is very similar. For both the pestiviruses and hepaciviruses, the mature nonstructural (NS) proteins, in sequential order from the amino-terminus of the nonstructural protein coding region to the carboxy-terminus of the ORF, consist of p7, NS2, NS3, NS4A, NS4B, NS5A, and NS5B.

[0010] The NS proteins of pestiviruses and hepaciviruses share sequence domains that are characteristic of specific protein functions. For example, the NS3 proteins of viruses in both groups possess amino acid sequence motifs characteristic of serine proteinases and of helicases (Gorbalenya et al., *Nature*, 1988, 333, 22; Bazan and Fletterick *Virology*, 1989, 171, 637-639; Gorbalenya et al., *Nucleic Acid Res.*, 1989, 17, 3889-3897). Similarly, the NS5B proteins of pestiviruses and hepaciviruses have the motifs characteristic of RNA-directed RNA polymerases (Koonin, E. V. and Dolja, V. V., *Crit. Rev. Biochem. Molec. Biol.* 1993, 28, 375-430).

[0011] The actual roles and functions of the NS proteins of pestiviruses and hepaciviruses in the lifecycle of the viruses are directly analogous. In both cases, the NS3 serine proteinase is responsible for all proteolytic processing of polyprotein precursors downstream of its position in the ORF (Wiskerchen and Collett, *Virology*, 1991, 184, 341-350; Bartenschlager et al., *J. Virol.* 1993, 67, 3835-3844; Eckart et al. *Biochem. Biophys. Res. Comm.* 1993, 192, 399-406; Grakoui et al., *J. Virol.* 1993, 67, 2832-2843; Grakoui et al., *Proc. Natl. Acad. Sci. USA* 1993, 90, 10583-10587; Hijikata et al., *J. Virol.* 1993, 67, 4665-4675; Tome et al., *J. Virol.*, 1993, 67, 4017-4026). The NS4A protein, in both cases, acts as a cofactor with the NS3 serine protease (Bartenschlager et al., *J. Virol.* 1994, 68, 5045-5055; Failla et al., *J. Virol.* 1994, 68, 3753-3760; Xu et al., *J. Virol.*, 1997, 71:53 12-5322). The NS3 protein of both viruses also functions as a helicase (Kim et al., *Biochem. Biophys. Res. Comm.*, 1995, 215, 160-166; Jin and Peterson, *Arch. Biochem. Biophys.*, 1995, 323, 47-53; Warrenner and Collett, *J. Virol.* 1995, 69, 1720-1726). Finally, the NS5B proteins of pestiviruses and hepaciviruses have the predicted RNA-directed RNA polymerases activity (Behrens et al., *EMBO*, 1996, 15, 12-22; Lechmann et al., *J. Virol.*, 1997, 71, 8416-8428; Yuan et al., *Biochem. Biophys. Res. Comm.* 1997, 232, 231-235; Hagedorn, PCT WO 97/12033; Zhong et al., *J. Virol.*, 1998, 72, 9365-9369).

[0012] Currently, there are limited treatment options for individuals infected with hepatitis C virus. The current approved therapeutic option is the use of immunotherapy with recombinant interferon- α alone or in combination with the nucleoside analog ribavirin. This therapy is limited in its clinical effectiveness and only 50% of treated patients respond to therapy. Therefore, there is significant need for more effective and novel therapies to address the unmet medical need posed by HCV infection.

[0013] A number of potential molecular targets for drug development of direct acting antivirals as anti-HCV therapeutics have now been identified including, but not limited to, the NS2-NS3 autoprotease, the N3 protease, the N3 helicase and the NS5B polymerase. The RNA-dependent RNA polymerase is absolutely essential for replication of the single-stranded, positive sense, RNA genome and this enzyme has elicited significant interest among medicinal chemists.

[0014] Inhibitors of HCV NS5B as potential therapies for HCV infection have been reviewed: Tan, S.-L., et al., *Nature Rev. Drug Discov.*, 2002, 1, 867-881; Walker, M. P. et al., *Exp. Opin. Investigational Drugs*, 2003, 12, 1269-1280; Ni, Z.-J., et al., *Current Opinion in Drug Discovery and Development*, 2004, 7, 446-459; Beaulieu, P. L., et al., *Current Opinion in Investigational Drugs*, 2004, 5, 838-850; Wu, J., et al., *Current Drug Targets-Infectious Disorders*, 2003, 3, 207-219; Griffith, R. C., et al., *Annual Reports in Medicinal Chemistry*, 2004, 39, 223-237; Carrol, S., et al., *Infectious Disorders-Drug Targets*, 2006, 6, 17-29. The potential for the emergence

of resistant HCV strains and the need to identify agents with broad genotype coverage supports the need for continuing efforts to identify novel and more effective nucleosides as HCV NS5B inhibitors.

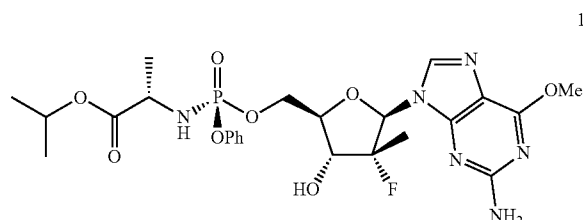
[0015] Nucleoside inhibitors of NS5B polymerase can act either as a non-natural substrate that results in chain termination or as a competitive inhibitor which competes with nucleotide binding to the polymerase. To function as a chain terminator the nucleoside analog must be taken up by the cell and converted in vivo to a triphosphate to compete for the polymerase nucleotide binding site. This conversion to the triphosphate is commonly mediated by cellular kinases which imparts additional structural requirements on a potential nucleoside polymerase inhibitor. Unfortunately, this limits the direct evaluation of nucleosides as inhibitors of HCV replication to cell-based assays capable of in situ phosphorylation.

[0016] In some cases, the biological activity of a nucleoside is hampered by its poor substrate characteristics for one or more of the kinases needed to convert it to the active triphosphate form. Formation of the monophosphate by a nucleoside kinase is generally viewed as the rate limiting step of the three phosphorylation events. To circumvent the need for the initial phosphorylation step in the metabolism of a nucleoside to the active triphosphate analog, the preparation of stable phosphate prodrugs has been reported. Nucleoside phosphoramidate prodrugs have been shown to be precursors of the active nucleoside triphosphate and to inhibit viral replication when administered to viral infected whole cells (McGuigan, C., et al., *J. Med. Chem.*, 1996, 39, 1748-1753; Valette, G., et al., *J. Med. Chem.*, 1996, 39, 1981-1990; Balzarini, J., et al., *Proc. National Acad Sci USA*, 1996, 93, 7295-7299; Siddiqui, A. Q., et al., *J. Med. Chem.*, 1999, 42, 4122-4128; Eisenberg, E. J., et al., *Nucleosides, Nucleotides and Nucleic Acids*, 2001, 20, 1091-1098; Lee, W. A., et al., *Antimicrobial Agents and Chemotherapy*, 2005, 49, 1898; US 2006/0241064; and WO 2007/095269.

[0017] Also limiting the utility of nucleosides as viable therapeutic agents is their sometimes poor physicochemical and pharmacokinetic properties. These poor properties can limit the intestinal absorption of an agent and limit uptake into the target tissue or cell. To improve on their properties prodrugs of nucleosides have been employed. It has been demonstrated that preparation of nucleoside phosphoramidates improves the systemic absorption of a nucleoside and furthermore, the phosphoramidate moiety of these "pronucleotides" is masked with neutral lipophilic groups to obtain a suitable partition coefficient to optimize uptake and transport into the cell dramatically enhancing the intracellular concentration of the nucleoside monophosphate analog relative to administering the parent nucleoside alone. Enzyme-mediated hydrolysis of the phosphate ester moiety produces a nucleoside monophosphate wherein the rate limiting initial phosphorylation is unnecessary. To this end, U.S. patent application Ser. No. 12/053,015, which corresponds to WO 2008/121634 and US 2010/0016251, discloses a number of phosphoramidate nucleoside prodrugs, many of which show activity in an HCV assay. Several compounds disclosed in US 2010/0016251 were tested as a potential clinical candidate for approval by the FDA.

SUMMARY OF THE INVENTION

[0018] Disclosed herein is a compound represented by formula 1 its hydrate or solvate thereof in crystalline or crystal-like form.



BRIEF DESCRIPTION OF THE DRAWINGS

[0019] FIG. 1. High resolution XRD diffractogram of 1.H₂O.

[0020] FIG. 2. X-Ray Crystal Structure for 1.H₂O.

DETAILED DESCRIPTION OF THE INVENTION

Definitions

[0021] The phrase “a” or “an” entity as used herein refers to one or more of that entity; for example, a compound refers to one or more compounds or at least one compound. As such, the terms “a” (or “an”), “one or more”, and “at least one” can be used interchangeably herein.

[0022] The terms “optional” or “optionally” as used herein means that a subsequently described event or circumstance may but need not occur, and that the description includes instances where the event or circumstance occurs and instances in which it does not. For example, “optional bond” means that the bond may or may not be present, and that the description includes single, double, or triple bonds.

[0023] The term “P*” (where used) means that the phosphorus atom is chiral and that it has a corresponding Cahn-Ingold-Prelog designation of “R” or “S” which have their accepted plain meanings. Due to the chirality at phosphorus, Compound 1, as used herein, is sometime referred to the S_P-isomer. Its diastereomeric analog is some times referred to as the R_P-isomer. Mixtures of the S_P-isomer and R_P-isomer are sometimes referred to as a mixture containing 1 and the R_P-isomer.

[0024] The term “purified,” as described herein, refers to the purity of a given compound. For example, a compound is “purified” when the given compound is a major component of the composition, i.e., at least 50% w/w pure. Thus, “purified” embraces at least 50% w/w purity, at least 60% w/w purity, at least 70% purity, at least 80% purity, at least 85% purity, at least 90% purity, at least 92% purity, at least 94% purity, at least 96% purity, at least 97% purity, at least 98% purity, at least 99% purity, at least 99.5% purity, and at least 99.9% purity, wherein “substantially pure” embraces at least 97% purity, at least 98% purity, at least 99% purity, at least 99.5% purity, and at least 99.9% purity

[0025] The term “about” (also represented by ~) means that the recited numerical value is part of a range that varies within standard experimental error.

[0026] The expression “substantially as shown in . . .” a specified XRPD pattern means that the peak positions shown in the XRPD pattern are substantially the same, within visual

inspection or resort to selected peak listings ($\pm 0.2^\circ 2\theta$). One of ordinary skill understands that the intensities can vary depending on the sample.

[0027] The term “substantially anhydrous” means that a substance contains at most 10% by weight of water, preferably at most 1% by weight of water, more preferably at most 0.5% by weight of water, and most preferably at most 0.1% by weight of water.

[0028] A solvent or anti-solvent (as used in reactions, crystallization, etc. or lattice and/or adsorbed solvents) includes at least one of a C₁ to C₈ alcohol, a C₂ to C₈ ether, a C₃ to C₇ ketone, a C₃ to C₇ ester, a C₁ to C₂ chlorocarbon, a C₂ to C₇ nitrile, a miscellaneous solvent, a C₅ to C₁₂ saturated hydrocarbon, and a C₆ to C₁₂ aromatic hydrocarbon.

[0029] The C₁ to C₈ alcohol refers to a straight/branched and/or cyclic/acyclic alcohol having such number of carbons. The C₁ to C₈ alcohol includes, but is not limited to, methanol, ethanol, n-propanol, isopropanol, isobutanol, hexanol, and cyclohexanol.

[0030] The C₂ to C₈ ether refers to a straight/branched and/or cyclic/acyclic ether having such number of carbons. The C₂ to C₈ ether includes, but is not limited to, dimethyl ether, diethyl ether, di-isopropyl ether, di-n-butyl ether, methyl-t-butyl ether (MTBE), tetrahydrofuran, and dioxane

[0031] The C₃ to C₇ ketone refers to a straight/branched and/or cyclic/acyclic ketone having such number of carbons. The C₃ to C₇ ketone includes, but is not limited to, acetone, methyl ethyl ketone, propanone, butanone, methyl isobutyl ketone, methyl butyl ketone, and cyclohexanone.

[0032] The C₃ to C₇ ester refers to a straight/branched and/or cyclic/acyclic ester having such number of carbons. The C₃ to C₇ ester includes, but is not limited to, ethyl acetate, propyl acetate, i-propyl acetate, n-butyl acetate, etc.

[0033] The C₁ to C₂ chlorocarbon refers to a chlorocarbon having such number of carbons. The C₁ to C₂ chlorocarbon includes, but is not limited to, chloroform, methylene chloride (DCM), carbon tetrachloride, 1,2-dichloroethane, and tetrachloroethane.

[0034] A C₂ to C₇ nitrile refers to a nitrile have such number of carbons. The C₂ to C₇ nitrile includes, but is not limited to, acetonitrile, propionitrile, etc.

[0035] A miscellaneous solvent refers to a solvent commonly employed in organic chemistry, which includes, but is not limited to, diethylene glycol, diglyme (diethylene glycol dimethyl ether), 1,2-dimethoxy-ethane, dimethylformamide, dimethylsulfoxide, ethylene glycol, glycerin, hexamethylphosphoramide, hexamethylphosphorous triame, N-methyl-2-pyrrolidinone, nitromethane, pyridine, triethyl amine, and acetic acid.

[0036] The term C₅ to C₁₂ saturated hydrocarbon refers to a straight/branched and/or cyclic/acyclic hydrocarbon. The C₅ to C₁₂ saturated hydrocarbon includes, but is not limited to, n-pentane, petroleum ether (ligroine), n-hexane, n-heptane, cyclohexane, and cycloheptane.

[0037] The term C₆ to C₁₂ aromatic refers to substituted and unsubstituted hydrocarbons having a phenyl group as their backbone. Preferred hydrocarbons include benzene, xylene, toluene, chlorobenzene, o-xylene, m-xylene, p-xylene, xylenes, and anisole.

[0038] The term “co-crystallates” include co-crystallates of 1 in combination with salts, which embraces pharmaceutically acceptable salts.

[0039] The term “salts,” as described herein, refers to a compound comprising a cation and an anion, which can pro-

duced by the protonation of a proton-accepting moiety and/or deprotonation of a proton-donating moiety. It should be noted that protonation of the proton-accepting moiety results in the formation of a cationic species in which the charge is balanced by the presence of a physiological anion, whereas deprotonation of the proton-donating moiety results in the formation of an anionic species in which the charge is balanced by the presence of a physiological cation. This term is meant to embrace pharmaceutically acceptable salts.

[0040] The phrase “pharmaceutically acceptable salt” means a salt that is pharmaceutically acceptable. Examples of pharmaceutically acceptable salts include, but are not limited to: (1) acid addition salts, formed with inorganic acids such as hydrochloric acid, hydrobromic acid, sulfuric acid, nitric acid, phosphoric acid, and the like; or formed with organic acids such as glycolic acid, pyruvic acid, lactic acid, malonic acid, malic acid, maleic acid, fumaric acid, tartaric acid, citric acid, 3-(4-hydroxybenzoyl)benzoic acid, cinnamic acid, mandelic acid, methanesulfonic acid, ethanesulfonic acid, 1,2-ethane-disulfonic acid, 2-hydroxyethanesulfonic acid, benzenesulfonic acid, 4-chlorobenzenesulfonic acid, 2-naphthalenesulfonic acid, 4-toluenesulfonic acid, camphorsulfonic acid, lauryl sulfuric acid, gluconic acid, glutamic acid, salicylic acid, muconic acid, and the like or (2) basic addition salts formed with the conjugate bases of any of the inorganic acids listed above, wherein the conjugate bases comprise a cationic component selected from among Na^+ , Mg^{2+} , Ca^{2+} , NH_4^+ , R^g , in which R^g is a C_{1-3} alkyl and g is a number selected from among 0, 1, 2, 3, or 4. It should be understood that all references to pharmaceutically acceptable salts include solvent addition forms (solvates) or crystal forms (polymorphs) as defined herein, of the same acid addition salt.

[0041] The term “preparation” or “dosage form” is intended to include both solid and liquid formulations of the active compound and one skilled in the art will appreciate that an active ingredient can exist in different preparations depending on the desired dose and pharmacokinetic parameters.

[0042] The term “excipient” as used herein refers to a compound that is used to prepare a pharmaceutical composition, and is generally safe, non-toxic and neither biologically nor otherwise undesirable, and includes excipients that are acceptable for veterinary use as well as human pharmaceutical use.

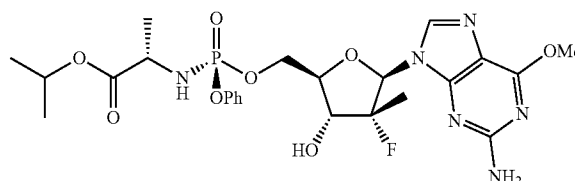
[0043] The term “crystalline” refers to a situation where a solid sample of 1 has crystalline characteristics when determined by X-ray powder diffraction or a single crystal X-ray technique.

[0044] The term “crystal-like” refers to a situation where a solid sample of 1 has crystalline characteristics when determined by one means, e.g., visually or by optical or polarizing microscopy, but does not have crystalline characteristics when determined by another means, e.g., x-ray powder diffraction. Methods of visually determining the crystallinity of a solid sample by visual or by optical or by polarizing microscopy are disclosed in U.S. Pat. Nos. <695> and <776>, both of which are incorporated by reference. A solid sample of 1 that is “crystal-like” may be crystalline under certain conditions but may become non-crystalline when subjected to other conditions.

[0045] The term “amorphous” refers to a situation where a solid sample of 1 is neither crystalline nor crystal-like.

Embodiments

[0046] A first embodiment is directed to a compound represented by a compound represented by formula 1 its hydrate or solvate thereof in crystalline or crystal-like form.



[0047] The compound represented by formula 1 as its hydrate in crystalline or crystal-like form is designated as $1.m\text{H}_2\text{O}$, where m varies in an integer or non-integer amount from about 0 to about 5. The compound represented by formula 1 as its solvate in crystalline or crystal-like form is designated as $1.n\text{S}$, where n varies in an integer or non-integer amount from about 0 to about 3. The compound represented by formula 1 its hydrate or solvate might have a certain advantageous amount of adsorbed solvent (S) or water. In which case, the amount of S or water can vary from about 0 wt. % to about 10 wt. % based on the weight of the compound represented by formula 1 or its hydrate in crystalline or crystal-like form.

[0048] A second embodiment is directed to crystalline or crystal-like 1.

[0049] A third embodiment is directed to crystalline or crystal-like $1.m\text{H}_2\text{O}$, where m varies in an integer or non-integer amount from about 0 to about 5.

[0050] A first aspect of the third embodiment is directed to crystalline or crystal-like $1.\text{H}_2\text{O}$.

[0051] A second aspect of the third embodiment is directed to crystalline or crystal-like $1.1/2\text{H}_2\text{O}$.

[0052] A fourth embodiment is directed to crystalline $1.\text{H}_2\text{O}$.

[0053] A first aspect of the fourth embodiment is directed to an orthorhombic crystalline $1.\text{H}_2\text{O}$, preferably having the following unit cell parameters $a \sim 10.99 \text{ \AA}$, $b \sim 13.09 \text{ \AA}$, and $c \sim 20.36 \text{ \AA}$.

[0054] A second aspect of the fourth embodiment is directed to a crystalline $1.\text{H}_2\text{O}$ having an XRPD 2θ -reflection ($^\circ$) at about: 14.8.

[0055] A third aspect of the fourth embodiment is directed to a crystalline $1.\text{H}_2\text{O}$ having XRPD 2θ -reflections ($^\circ$) at about: 14.8 and 17.6

[0056] A fourth aspect of the fourth embodiment is directed to a crystalline $1.\text{H}_2\text{O}$ having XRPD 2θ -reflections ($^\circ$) at about: 14.8, 17.6, and 20.4

[0057] A fifth aspect of the fourth embodiment is directed to a crystalline $1.\text{H}_2\text{O}$ having XRPD 2θ -reflections ($^\circ$) at about: 8.7, 14.8, 17.6, and 20.4.

[0058] A sixth aspect of the fourth embodiment is directed to a crystalline $1.\text{H}_2\text{O}$ having XRPD 2θ -reflections ($^\circ$) at about: 8.7, 13.6, 14.8, 17.6, and 20.4.

[0059] A seventh aspect of the fourth embodiment is directed to a crystalline 1.H₂O having XRPD 2θ-reflections (°) at about: 8.7, 11.1, 13.6, 14.8, 17.6, and 20.4.

[0060] An eighth aspect of the fourth embodiment is directed to a crystalline 1.H₂O having an XRPD diffraction pattern substantially as that shown in FIG. 1.

[0061] A fifth embodiment is directed to substantially pure crystalline 1.H₂O.

[0062] A sixth embodiment is directed to crystal-like 1.H₂O.

Dosage, Administration, and Use

[0063] A seventh embodiment is directed to a composition for the treatment and/or prophylaxis of any of the viral agents using crystalline or crystal-like 1.mH₂O or 1.nS. Possible viral agents include, but are not limited to: hepatitis C virus, hepatitis B virus, Hepatitis A virus, West Nile virus, yellow fever virus, dengue virus, rhinovirus, polio virus, bovine viral diarrhea virus, Japanese encephalitis virus, or those viruses belonging to the groups of Pestiviruses, hepaciviruses, or flavaviruses.

[0064] An aspect of this embodiment is directed to a composition for the treatment of any of the viral agents disclosed herein said composition comprising a pharmaceutically acceptable medium selected from among an excipient, carrier, diluent, and equivalent medium and crystalline or crystal-like 1.mH₂O or 1.nS, that is intended to include its hydrates, solvates, and any crystalline forms of crystalline or crystal-like 1.mH₂O or 1.nS.

[0065] The crystalline or crystal-like 1.mH₂O or 1.nS may be independently formulated in a wide variety of oral administration dosage forms and carriers. Oral administration can be in the form of tablets, coated tablets, hard and soft gelatin capsules, solutions, emulsions, syrups, or suspensions. The crystalline or crystal-like 1.mH₂O or 1.nS is efficacious when administered by suppository administration, among other routes of administration. The most convenient manner of administration is generally oral using a convenient daily dosing regimen which can be adjusted according to the severity of the disease and the patient's response to the antiviral medication.

[0066] The crystalline or crystal-like 1.mH₂O or 1.nS together with one or more conventional excipients, carriers, or diluents, may be placed into the form of pharmaceutical compositions and unit dosages. The pharmaceutical compositions and unit dosage forms may be comprised of conventional ingredients in conventional proportions, with or without additional active compounds and the unit dosage forms may contain any suitable effective amount of the active ingredient commensurate with the intended daily dosage range to be employed. The pharmaceutical compositions may be employed as solids, such as tablets or filled capsules, semi-solids, powders, sustained release formulations, or liquids such as suspensions, emulsions, or filled capsules for oral use; or in the form of suppositories for rectal or vaginal administration. A typical preparation will contain from about 5% to about 95% active compound or compounds (w/w).

[0067] The crystalline or crystal-like 1.mH₂O or 1.nS can be administered alone but will generally be administered in admixture with one or more suitable pharmaceutical excipients, diluents or carriers selected with regard to the intended route of administration and standard pharmaceutical practice.

[0068] Solid form preparations include, for example, powders, tablets, pills, capsules, suppositories, and dispersible

granules. A solid carrier may be one or more substances which may also act as diluents, flavoring agents, solubilizers, lubricants, suspending agents, binders, preservatives, tablet disintegrating agents, or an encapsulating material. In powders, the carrier generally is a finely divided solid which is a mixture with the finely divided active component. In tablets, the active component generally is mixed with the carrier having the necessary binding capacity in suitable proportions and compacted in the shape and size desired. Suitable carriers include but are not limited to magnesium carbonate, magnesium stearate, talc, sugar, lactose, pectin, dextrin, starch, gelatin, tragacanth, methylcellulose, sodium carboxymethylcellulose, a low melting wax, cocoa butter, and the like. Solid form preparations may contain, in addition to the active component, colorants, flavors, stabilizers, buffers, artificial and natural sweeteners, dispersants, thickeners, solubilizing agents, and the like. Examples of solid formulations are exemplified in EP 0524579; US 2002/0142050; US 2004/0224917; US 2005/0048116; US 2005/0058710; US 2006/0034937; US 2006/0057196; US 2006/0188570; US 2007/0026073; US 2007/0059360; US 2007/0077295; US 2007/0099902; US 2008/0014228; U.S. Pat. No. 6,267,985; U.S. Pat. No. 6,294,192; U.S. Pat. No. 6,383,471; U.S. Pat. No. 6,395,300; U.S. Pat. No. 6,569,463; U.S. Pat. No. 6,635,278; U.S. Pat. No. 6,645,528; U.S. Pat. No. 6,923,988; U.S. Pat. No. 6,932,983; U.S. Pat. No. 7,060,294; and U.S. Pat. No. 7,462,608, each of which is incorporated by reference.

[0069] Liquid formulations also are suitable for oral administration include liquid formulation including emulsions, syrups, elixirs and aqueous suspensions. These include solid form preparations which are intended to be converted to liquid form preparations shortly before use. Examples of liquid formulation are exemplified in U.S. Pat. Nos. 3,994,974; 5,695,784; and 6,977,257. Emulsions may be prepared in solutions, for example, in aqueous propylene glycol solutions or may contain emulsifying agents such as lecithin, sorbitan monooleate, or acacia. Aqueous suspensions can be prepared by dispersing the finely divided active component in water with viscous material, such as natural or synthetic gums, resins, methylcellulose, sodium carboxymethylcellulose, and other well known suspending agents.

[0070] The crystalline or crystal-like 1.mH₂O or 1.nS may be independently formulated for administration as suppositories. A low melting wax, such as a mixture of fatty acid glycerides or cocoa butter is first melted and the active component is dispersed homogeneously, for example, by stirring. The molten homogeneous mixture is then poured into convenient sized molds, allowed to cool, and to solidify.

[0071] The crystalline or crystal-like 1.mH₂O or 1.nS may be independently formulated for vaginal administration. Pessaries, tampons, creams, gels, pastes, foams or sprays containing in addition to the active ingredient such carriers as are known in the art to be appropriate. Certain of these formulations may also be used in conjunction with a condom with or without a spermicidal agent.

[0072] Suitable formulations along with pharmaceutical carriers, diluents and excipients are described in *Remington: The Science and Practice of Pharmacy* 1995, edited by E. W. Martin, Mack Publishing Company, 19th edition, Easton, Pa., which is hereby incorporated by reference. A skilled formulation scientist may modify the formulations within the teachings of the specification to provide numerous formulations for a particular route of administration without rendering

compositions containing the compounds contemplated herein unstable or compromising their therapeutic activity.

[0073] Additionally, the purified crystalline or crystal-like 1.mH₂O or 1.nS may be independently formulated in conjunction with liposomes or micelles. As to liposomes, it is contemplated that the purified compounds can be formulated in a manner as disclosed in U.S. Pat. Nos. 4,797,285; 5,013,556; 5,077,056; 5,077,057; 5,154,930; 5,192,549; 5,213,804; 5,225,212; 5,277,914; 5,316,771; 5,376,380; 5,549,910; 5,567,434; 5,736,155; 5,827,533; 5,882,679; 5,891,468; 6,060,080; 6,132,763; 6,143,321; 6,180,134; 6,200,598; 6,214,375; 6,224,903; 6,296,870; 6,653,455; 6,680,068; 6,726,925; 7,060,689; and 7,070,801, each of which is incorporated by reference. As to micelles, it is contemplated that the purified compounds can be formulated in a manner as disclosed in U.S. Pat. Nos. 5,145,684 and 5,091,188, both of which are incorporated by reference.

[0074] The fifth embodiment is directed to a use of crystalline or crystal-like 1.mH₂O or 1.nS in the manufacture of a medicament for the treatment of any condition the result of an infection by any one of the following viral agents: hepatitis C virus, West Nile virus, yellow fever virus, degue virus, rhinovirus, polio virus, hepatitis A virus, bovine viral diarrhea virus and Japanese encephalitis virus.

[0075] The term “medicament” means a substance used in a method of treatment and/or prophylaxis of a subject in need thereof, wherein the substance includes, but is not limited to, a composition, a formulation, a dosage form, and the like, comprising crystalline or crystal-like 1.mH₂O or 1.nS. It is contemplated that crystalline or crystal-like 1.mH₂O or 1.nS in the manufacture of a medicament, for the treatment of any of the antiviral conditions disclosed herein, either alone or in combination with another compound disclosed herein. A medicament includes, but is not limited to, any one of the compositions contemplated by the fourth embodiment disclosed herein.

[0076] An eighth embodiment is directed to a method of treatment and/or prophylaxis in a subject in need thereof said method comprises administering a therapeutically effective amount of crystalline or crystal-like 1.mH₂O or 1.nS to the subject.

[0077] It is intended that a subject in need thereof is one that has any condition the result of an infection by any of the viral agents disclosed herein, which includes, but is not limited to, hepatitis C virus, West Nile virus, yellow fever virus, degue virus, rhinovirus, polio virus, hepatitis A virus, bovine viral diarrhea virus or Japanese encephalitis virus, flaviviridae viruses or pestiviruses or hepaciviruses or a viral agent causing symptoms equivalent or comparable to any of the above-listed viruses.

[0078] The term “subject” means a mammal, which includes, but is not limited to, cattle, pigs, sheep, chicken, turkey, buffalo, llama, ostrich, dogs, cats, and humans, preferably the subject is a human. It is contemplated that in the method of treating a subject thereof of the ninth embodiment can be any of the compounds contemplated herein, either alone or in combination with another compound disclosed herein.

[0079] The term “therapeutically effective amount” as used herein means an amount required to reduce symptoms of the disease in an individual. The dose will be adjusted to the individual requirements in each particular case. That dosage can vary within wide limits depending upon numerous factors such as the severity of the disease to be treated, the age and

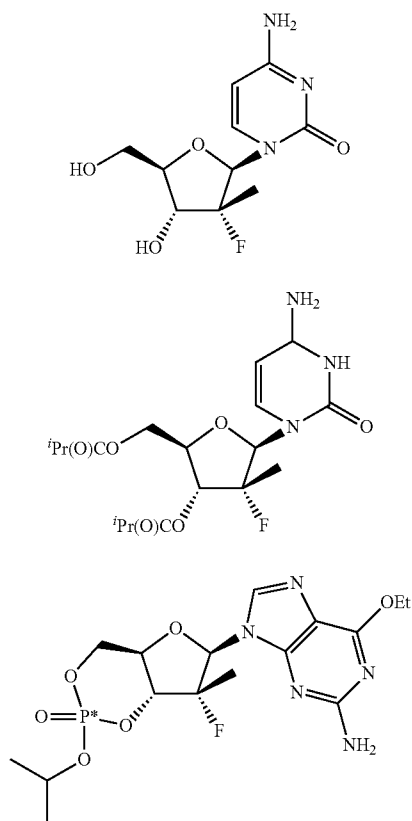
general health condition of the patient, other medicaments with which the patient is being treated, the route and form of administration and the preferences and experience of the medical practitioner involved. For oral administration, a daily dosage of between about 0.001 and about 10 g, including all values in between, such as 0.001, 0.0025, 0.005, 0.0075, 0.01, 0.025, 0.050, 0.075, 0.1, 0.125, 0.150, 0.175, 0.2, 0.25, 0.5, 0.75, 1, 1.5, 2, 2.5, 3, 3.5, 4, 4.5, 5, 5.5, 6, 6.5, 7, 7.5, 8, 8.5, 9, and 9.5, per day should be appropriate in monotherapy and/or in combination therapy. A particular daily dosage is between about 0.01 and about 1 g per day, including all incremental values of 0.01 g (i.e., 10 mg) in between, a preferred daily dosage about 0.01 and about 0.8 g per day, more preferably about 0.01 and about 0.6 g per day, and most preferably about 0.01 and about 0.25 g per day, each of which including all incremental values of 0.01 g in between. Generally, treatment is initiated with a large initial “loading dose” to rapidly reduce or eliminate the virus following by a decreasing the dose to a level sufficient to prevent resurgence of the infection. One of ordinary skill in treating diseases described herein will be able, without undue experimentation and in reliance on knowledge, experience and the disclosures of this application, to ascertain a therapeutically effective amount of the compound disclosed herein for a given disease and patient.

[0080] Therapeutic efficacy can be ascertained from tests of liver function including, but not limited to protein levels such as serum proteins (e.g., albumin, clotting factors, alkaline phosphatase, aminotransferases (e.g., alanine transaminase, aspartate transaminase), 5'-nucleosidase, γ -glutamyl-transpeptidase, etc.), synthesis of bilirubin, synthesis of cholesterol, and synthesis of bile acids; a liver metabolic function, including, but not limited to, carbohydrate metabolism, amino acid and ammonia metabolism. Alternatively the therapeutic effectiveness may be monitored by measuring HCV-RNA. The results of these tests will allow the dose to be optimized.

[0081] A first aspect of the eighth embodiment is directed to a method of treatment and/or prophylaxis in a subject in need thereof said method comprises administering to the subject a therapeutically effective amount of a compound represented by compound 1.mH₂O or 1.nS and a therapeutically effective amount of another antiviral agent; wherein the administration is concurrent or alternative. It is understood that the time between alternative administration can range between 1-24 hours, which includes any sub-range in between including, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, and 23 hours.

[0082] Examples of “another antiviral agent” include, but are not limited to: HCV NS3 protease inhibitors (see EP 1881001, US 2003187018, US 2005267018, WO 2003006490, WO 200364456, WO 2004094452, WO 2005028502, WO 2005037214, WO 2005095403, WO 2007014920, WO 2007014921, WO 2007014922, WO 2007014925, WO 2007014926, WO 2007015824, WO 2008010921, and WO 2008010921); HCV NS5B Inhibitors (see US 2004229840, US 2005154056, US 2005-98125, US 20060194749, US 20060241064, US 20060293306, US 2006040890, US 2006040927, US 2006166964, US 2007275947, U.S. Pat. No. 6,784,166, US20072759300, WO 2002057287, WO 2002057425, WO 2003010141, WO 2003037895, WO 2003105770, WO 2004000858, WO 2004002940, WO 2004002944, WO 2004002977, WO 2004003138, WO 2004041201, WO 2004065367, WO

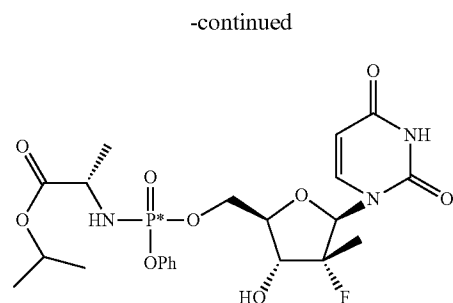
2004096210, WO 2005021568, WO 2005103045, WO 2005123087, WO 2006012078, WO 2006020082, WO 2006065335, WO 2006065590, WO 2006093801, WO 200702602, WO 2007039142, WO 2007039145, WO 2007076034, WO 2007088148, WO 2007092000, and WO2007095269); HCV NS4 Inhibitors (see WO 2005067900 and WO 2007070556); HCV NS5a Inhibitors (see US 2006276511, WO 2006035061, WO 2006100310, WO 2006120251, and WO 2006120252); Toll-like receptor agonists (see WO 2007093901); and other inhibitors (see WO 2000006529, WO 2003101993, WO 2004009020, WO 2004014313, WO 2004014852, and WO 2004035571); compound A (shown below and disclosed in U.S. Pat. No. 7,429, 572); compound B (disclosed in US 2007/0197463); compound C (disclosed in US 2010/0081628, see also compound 19a and 19b disclosed in the same application, which are individual diastereomers of compound C); compound D (disclosed in US 2010/0016251); compound E (disclosed in U.S. Ser. No. 12/783,680, as well as Rp-4 disclosed in the same application); telaprevir (also known as VX-950, which is disclosed in US 2010/0015090); boceprevir (disclosed in US 2006/0276405); BMS-790052 (disclosed in WO 2008/021927); ITMN-191 (disclosed in US 2009/0269305 at Example 62-1); ANA-598 (shown below and identified as compound 31 in F. Ruebasam et al. *Biorg. Med. Chem. Lett.* (2008) 18: 3616-3621; and TMC435 (formerly known as TMC435350)



A

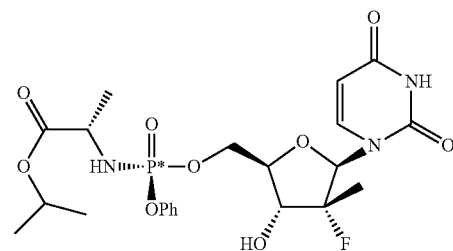
B

C

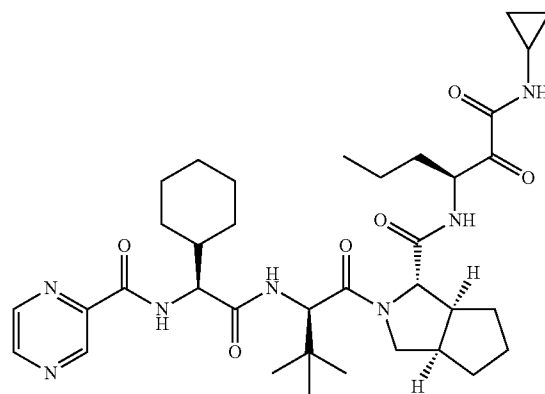


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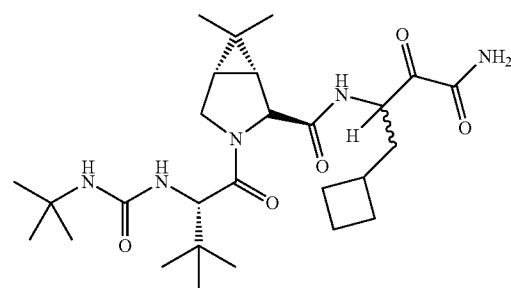
D



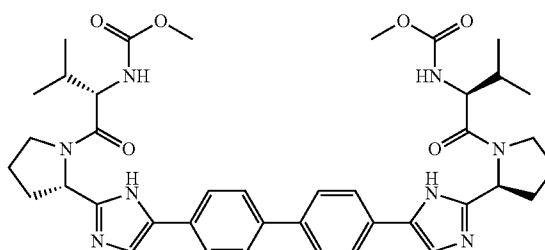
E



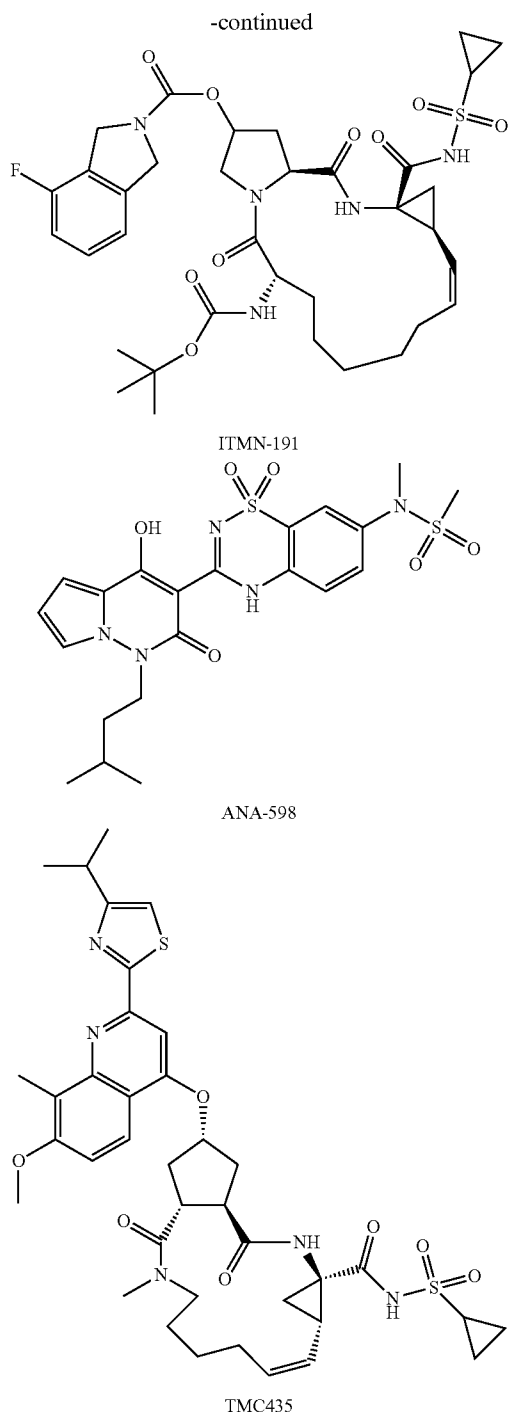
Telaprevir (VX-950)



Boceprevir



BMS-790052



as well as, interferon- α , interferon- β , pegylated interferon- α , ribavirin, levovirin, viramidine, another nucleoside HCV polymerase inhibitor, a HCV non-nucleoside polymerase inhibitor, a HCV protease inhibitor, a HCV helicase inhibitor or a HCV fusion inhibitor.

[0083] When crystalline or crystal-like 1.mH₂O or 1.nS are administered in combination with another antiviral agent the activity may be increased over the parent compound. When the treatment is combination therapy, such administration

may be concurrent or sequential with respect to that of the nucleoside derivatives. "Concurrent administration" as used herein thus includes administration of the agents at the same time or at different times. Administration of two or more agents at the same time can be achieved by a single formulation containing two or more active ingredients or by substantially simultaneous administration of two or more dosage forms with a single active agent.

[0084] It will be understood that references herein to treatment extend to prophylaxis as well as to the treatment of existing conditions. Furthermore, the term "treatment" of a HCV infection, as used herein, also includes treatment or prophylaxis of a disease or a condition associated with or mediated by HCV infection, or the clinical symptoms thereof.

Preparation

[0085] A ninth embodiment is directed to process for preparing crystalline or crystal-like 1.mH₂O or 1.nS, which comprises crystallizing 1.mH₂O or 1.nS, wherein m and n are as defined above.

[0086] A first aspect of the ninth embodiment is directed to a process for preparing crystalline or crystal-like 1.mH₂O or 1.nS, further comprises dissolving or suspending 1 in a solvent or solvent mixture.

[0087] A second aspect of the ninth embodiment is directed to a process for preparing crystalline or crystal-like 1.mH₂O or 1.nS, which further comprises adding seed crystals of 1.mH₂O or 1.nS.

[0088] A third aspect of the ninth embodiment is directed to a process for preparing crystalline or crystal-like 1.mH₂O or 1.nS, which further comprises adding an anti-solvent to the solvent or solvent mixture.

[0089] Any suitable solvent may be used that affords crystalline or crystal-like 1.mH₂O or 1.nS. Specific solvents contemplated include, but are not limited to, anisole, ethyl acetate; xylenes; toluene; isopropanol; acetone; dichloromethane; diethyl ether; isopropyl acetate; t-Butyl methyl ether; or combinations thereof. Specific combinations include, but are not limited to, anisole/ethyl acetate; heptanes/ethyl acetate; xylenes/ethyl acetate/water; anisole/water; ethyl acetate/xylenes; isopropanol/xylenes; acetone/xylenes; dichloromethane/xylenes; dichloromethane/hexanes; ethyl acetate/toluene; diethyl ether/xylenes; isopropyl acetate/xylenes; isopropyl acetate/heptanes; ethyl acetate/water; t-butyl methyl ether/water; t-butyl methyl ether/ethyl ether; or t-butyl methyl ether. For solvent combinations that also include water, it is understood that a sufficient amount of water is required to form 1.mH₂O (when m \neq 0), which is based on an estimation of the amount of 1 used for the crystallization or the amount of 1 contained in a mixture containing 1 and its Rp-isomer. It is also understood that commercially available solvents contain a certain amount of water that might be sufficient, alone, to provide sufficient water for the formation of crystalline or crystal-like 1.mH₂O (when m \neq 0).

[0090] A tenth embodiment is directed to a method for determining the crystallinity of crystalline or crystal-like 1.mH₂O, which comprises analyzing 1.mH₂O by XRPD or single-crystal X-ray crystallography.

EXAMPLES

[0091] The following examples are intended to provide one of ordinary skill with a better understanding the disclosed embodiments.

Preparation of 1

[0092] Compound 1 may be prepared by stereoselective or non-stereoselective means. A stereoselective process is

described below, as well as U.S. Provisional Patent Application No. 61/319,548, which is incorporated by reference. A non-stereoselective process is also described below, as well as in U.S. patent application Ser. No. 12/645,765, the subject matter of which is incorporated by reference. Production of 1 via the non-stereoselective process, which produces a diastereomeric mixture containing 1 and its Rp-isomer, further includes, as detailed below, crystallization of the diastereomeric mixture to obtain 1 or chromatographic separation of the diastereomeric mixture by way of SMB chromatography.

Example 1

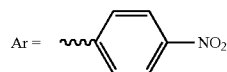
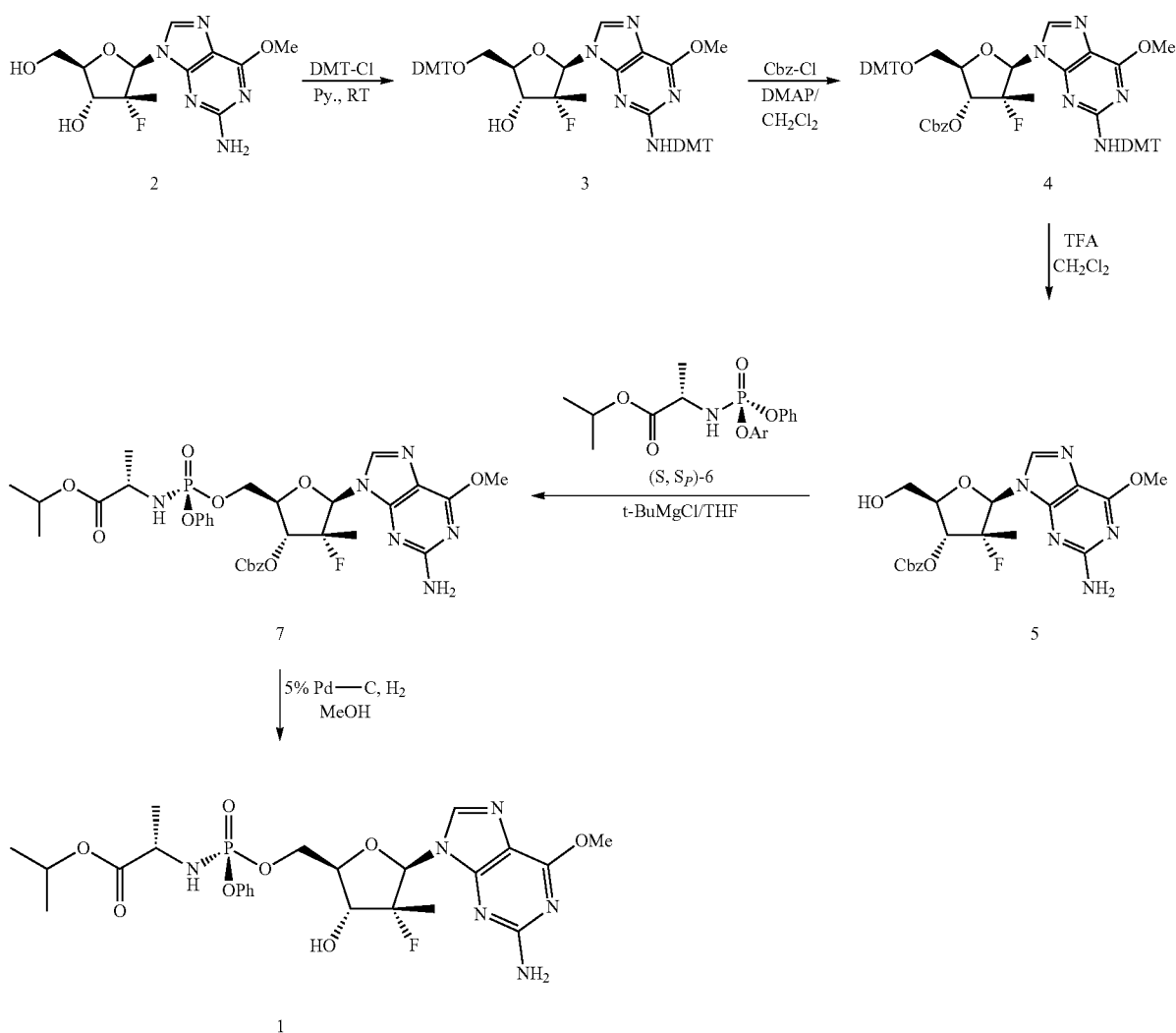
Stereoselective Preparation of 1

[0093]

Example 1-1

Synthesis of (2R,3R,4R,5R)-2-((bis(4-methoxyphenyl)(phenyl)methoxy)methyl)-5-(2-((bis(4-methoxyphenyl)(phenyl)methyl)amino)-6-methoxy-9H-purin-9-yl)-4-fluoro-4-methyltetrahydrofuran-3-ol (3):

[0094] To a solution of (2R,3R,4R,5R)-5-(2-amino-6-methoxy-9H-purin-9-yl)-4-fluoro-2-(hydroxymethyl)-4-methyltetrahydrofuran-3-ol (2, 4 g, 12.8 mmol) in anhydrous pyridine (100 mL) cooled at 0° C. was added DMT-Cl portion-wise under nitrogen. The brown solution was stirred at ambient temperature for 24 hours. The mixture was concentrated under reduced pressure to remove most of solvent and



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sat. NaHCO_3 (20 mL) was added. The mixture was diluted with water (150 mL) and EtOAc (120 mL). The organic layer was separated and washed with water (5 \times 120 mL), brine and dried over Na_2SO_4 . After removal of solvent, the residue was purified via column chromatography (20% EA in hexanes to 80% EA in hexanes) to afford 11.6 g of product, 3, as a white foam solid (quantitative yield). ^1H -NMR ($\text{DMSO}-d_6$): δ 7.94 (s, 1H), 7.39-7.37 (m, 3H), 7.26-7.14 (m, 17H), 6.84-6.80 (m, 8H), 5.58 (s, 1H), 4.04 (br, 1H), 3.71-3.70 (m, 14H), 3.68 (m, 1H), 3.48 (br, 2H), 3.20 (d, 1H), 0.88 (br, 3H).

Example 1-2

Synthesis of benzyl ((2R,3R,4R,5R)-2-((bis(4-methoxyphenyl)(phenyl)methoxy)methyl)-5-(2-((bis(4-methoxyphenyl)(phenyl)methyl)amino)-6-methoxy-9H-purin-9-yl)-4-fluoro-4-methyltetrahydrofuran-3-yl) carbonate (4)

[0095] To a solution of nucleoside 3 (2.52 g, 2.75 mmol) in anhydrous DCM (8 mL) was added DMAP (1.01 g, 8.2 mmol) and the solution was cooled at 0° C. in an ice-water bath. Cbz-Cl (0.77 g, 4.2 mmol) was added via a syringe to the mixture and resulted in a cloudy reaction mixture. The mixture was stirred at room temperature for 24 hours and sat. NaHCO_3 (10 mL) was added. The mixture was partitioned in DCM and water. The organic layer was dried over Na_2SO_4 and concentrated to a white foam solid. The residue was purified via column chromatography (10–60% EtOAc in hexanes) to afford 2.74 g product, 4, as a white foam solid (yield, 95%). ^1H -NMR (CDCl_3): δ 7.87 (s, 1H), 7.41-7.16 (m, 24H), 6.79-6.75 (m, 8H), 6.28 (s, 1H), 5.65 (br, 1H), 5.15 (s, 2H), 4.28 (d, 1H), 3.79-3.71 (m, 15H), 3.55-3.52 (m, 1H), 3.39-3.36 (m, 1H), 0.93 (br, 3H).

Example 1-3

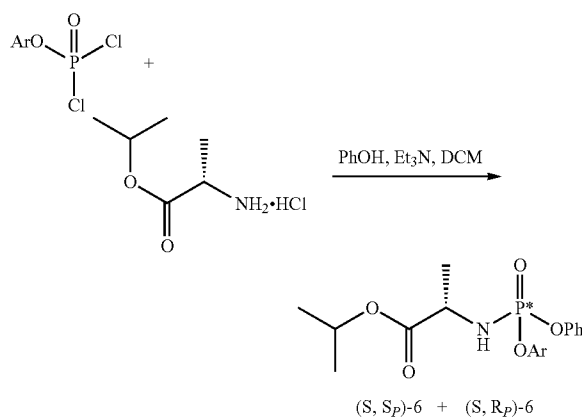
Synthesis of (2R,3R,4R,5R)-5-(2-amino-6-methoxy-9H-purin-9-yl)-4-fluoro-2-(hydroxymethyl)-4-methyltetrahydrofuran-3-yl benzyl carbonate (5)

[0096] A 1 vol % of TFA solution in DCM (50 mL) was added to a flask loaded with 4 (2.69 g, 2.56 mmol). The mixture was stirred at room temperature for 2 h and it was complete. Sat. NaHCO_3 (20 mL) was added and the mixture was partitioned in water and DCM. The organic layer was concentrated and solid residue was purified by column chromatography (silica gel, 0–5% 2-PrOH in DCM) to afford 1.01 g of product, 5, as a white foam solid (yield 88%). ^1H -NMR (CDCl_3): δ 7.82 (s, 1H), 7.39-7.33 (m, 5H), 6.02 (d, 1H, $J=19.2$ Hz), 5.77 (dd, 1H, $J=20.8, 8.8$ Hz), 5.32-5.30 (m, 1H), 5.20 (s, 2H), 5.04 (s, 2H), 4.34 (d, 1H, $J=8.8$ Hz), 4.15 (m, 1H), 4.04 (s, 3H), 3.85-3.79 (m, 1H), 1.21 (d, 3H, $J=22.8$ Hz).

Example 1-4

Preparation of (S)-2-[(4-nitro-phenoxy)-phenoxy-phosphorylamino]propionic acid isopropyl ester (Mixture of Diastereomers (S, S_P)-6 and (S, R_P)-6)

[0097]

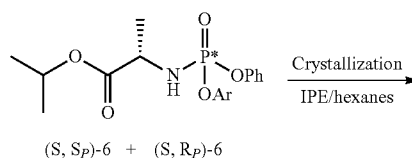


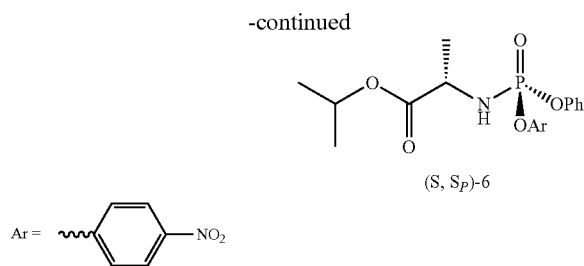
[0098] To a stirred solution of 4-nitrophenyl phosphorodichloridate 12.8 g, 50 mmol) in dichloromethane (100 mL) was added a solution of phenol and triethylamine (7.7 mL, 55 mmol) in dichloromethane (100 mL) at –78° C. over a period of 20 min. The mixture was stirred at this temperature for 30 min and then transferred to another round bottom flask containing L-alanine isopropyl ester hydrochloride (8.38 g, 50 mmol) in dichloromethane (100 mL) at 0° C. To the mixture was added second lot of triethylamine (14.6 mL, 105 mmol) over a period of 15 min. The mixture was stirred at 0° C. for 1 h and then the solvent was evaporated. The residue was triturated with ethyl acetate (150 mL) and the white solid was filtered off. The filtrate was concentrated under reduced pressure to give pale yellow oil. The crude compound was chromatographed using 0-20% ethyl acetate/hexanes gradient to give product (17 g, 83% yield) as a mixture of diastereomers in about 1:1 ratio. ^{31}P NMR (162 MHz, CDCl_3): δ –2.05, –2.10; ^1H NMR (400 MHz, CDCl_3): δ 8.22 (d, $J=9.2$ Hz, 2H), 7.41-7.33 (m, 4H), 7.26-7.18 (m, 3H), 5.05-4.96 (m, 1H), 4.14-4.05 (m, 1H), 3.93-3.88 (m, 1H), 1.38 (d, $J=6.8$ Hz, 3H), 1.22 (dd, $J=6.2$ & 3.0 Hz, 6H); MS (ESI) m/z 407 ($\text{M}-1$) $^+$.

Example 1-5

Crystallization of (S)-2-[(S)-(4-nitro-phenoxy)-phenoxy-phosphorylamino] propionic acid isopropyl ester ((S, S_P)-6

[0099]





[0100] (S)-2-[(4-Nitro-phenoxy)-phenoxy-phosphoryl-amino]-propionic acid isopropyl ester (3.4 g) was dissolved in IPE (6 mL). To the above solution were added hexanes (1 mL) while hand shaking until the solution was turbid. Few drops of IPE were then added to the mixture to get a clear solution. The mixture was gently stirred at room temperature for 20 h. A white and fine crystalline solid obtained was filtered, washed with 1:1 mixture of IPE/hexanes and dried to give white fluffy solid (820 mg, 24% yield) mp 52 (shrinks) 62-66 (melts). ³¹P NMR (162 MHz, CDCl₃): δ -2.05; ¹H NMR (400 MHz, CDCl₃): δ 8.22 (d, J=9.2 Hz, 2H), 7.41-7.33 (m, 4H), 7.26-7.18 (m, 3H), 5.05-4.96 (m, 1H), 4.14-4.05 (m, 1H), 3.93-3.88 (m, 1H), 1.38 (d, J=6.8 Hz, 3H), 1.22 (dd, J=6.2 & 3.0 Hz, 6H); MS (ESI) m/z 407 (M-1)⁺. The stereochemistry of (S, *S_p*)-6 as having the CIP configuration of S has been confirmed by single crystal X-ray crystallography, see U.S. 61/319,548, filed on Mar. 31, 2010.

Example 1-6

Synthesis of *S_p*-(2S)-isopropyl 2-((((2R,3R,4R,5R)-5-(2-amino-6-methoxy-9H-purin-9-yl)-3-(((benzyloxy)carbonyl)oxy)-4-fluoro-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphoryl)amino)propanoate (7)

[0101] To a solution of the nucleoside 5 (150 mg, 0.34 mmol) in 1.5 mL of anhydrous THF was added a solution of t-BuMgCl in THF (1.0 M, 0.41 mL) at 0° C. The cloudy mixture was stirred at ambient temperature for 1 h and then a solution of phosphoramidate reagent (ca 95% chiral purity) (S)-2-[(S)-(4-nitrophenoxy)phenoxyphosphorylamino]propionic acid isopropyl ester, (S, *S_p*)-6, (162 mg, 0.4 mmol) in 1.5 mL of THF was added to the mixture via a syringe dropwise. (Compound 6 is prepared according to the procedures outlined in U.S. patent application Ser. No. 12/645,765.) The mixture was stirred at ambient temperature for 20 h and ca 29% of starting material remained. The reaction was quenched by adding sat. NH₄Cl (4 mL) and 20 mL of EtOAc was added. After separation, organic layer was washed with water (3×25 mL), brine and dried over Na₂SO₄. After removal of solvent, the oil residue was checked by ¹H-NMR and ³¹P-NMR. The ratio of two isomers was ca. 12.5:1. The major isomer, ¹H-NMR (CDCl₃): δ 7.73 (s, 1H) (not completed); ³¹P-NMR (CDCl₃): δ 4.02.

Example 1-7

Synthesis of *S_p*-(2S)-isopropyl 2-((((2R,3R,4R,5R)-5-(2-amino-6-methoxy-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphoryl)amino)propanoate (1)

[0102] To a solution of crude phosphoramidate 7 in MeOH (2.5 mL) was added 5% Pd on charcoal (40 mg). The atmo-

sphere in the flask was exchanged with hydrogen twice. The mixture was stirred at ambient temperature under one atmosphere of hydrogen for 1 h. The mixture was filtered through a short pad of Celite and the filtrate was concentrated. The crude residue was checked by ¹H-NMR and ³¹P-NMR and ratio of two isomers was ca. 17:1 *S_p* isomer (1) and also matched the *S_p*-isomer by thin layer chromatography. ³¹P-NMR (DMSO-d₆): δ 4.91.

[0103] Alternatively, 1 can be prepared directly from 2, as illustrated below.

Example 2

Synthesis of *S_p*-(2S)-isopropyl 2-((((2R,3R,4R,5R)-5-(2-amino-6-methoxy-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphoryl)amino)propanoate, 1

[0104] To a 50 mL of dry flask were added the nucleoside (2, 100 mg, 0.32 mmol) and 1.5 mL of anhydrous THF. The suspension was cooled in an ice bath and a solution of t-BuMgCl in THF (1.7 M, 0.35 mL, 2 eq) was added via a syringe slowly. The resulting clear solution was stirred at room temperature for one hour. A solution of chiral enriched phosphoramidate reagent (98:2 (S, *S_p*)-6/(S, *R_p*)-6, 156 mg, 0.383 mmol) in 1.5 mL of anhydrous THF was added via a syringe at room temperature dropwise. After 48 h, TLC indicated approximately 35% of the starting nucleoside remained. The reaction was quenched by adding sat. NH₄Cl (6 mL). Ethyl acetate (20 mL) was added and organic layer was separated. Aqueous layer was extracted with ethyl acetate (10 mL). Combined organic layer was washed with water (2×20 mL), sat NaHCO₃ (15 mL), water (3×25 mL), brine, and dried over Na₂SO₄. After removal of solvent, the crude residue was checked by NMR. ³¹P NMR indicated a ratio of diastereomers was 75:1 (1/*R_p*-isomer of 1). The mixture was purified via column (silica gel, 0-8% MeOH in DCM) to afford product as a white foam solid (35.8 mg, 19%).

Non-Stereoselective Preparation of 1

Example 3

Synthesis of (2S)-isopropyl 2-((((2R,3R,4R,5R)-5-(2-amino-6-methoxy-9H-purin-9-yl)-4-fluoro-3-hydroxy-4-methyltetrahydrofuran-2-yl)methoxy)(phenoxy)phosphoryl)amino)propanoate (1)

[0105] To a 250 mL dry round-bottomed flask were loaded phenyl dichlorophosphate (2.66 g, 12.61 mmol) and anhydrous dichloromethane (40 mL). The amino ester salt (2.60 g, 15.53 mmol) was added to the solution and the mixture was cooled to -5° C. N-Methyl imidazole (7.7 mL, 97 mmol) was then added quickly via a dry syringe at -5° C. and the solution was stirred at -5° C. for 1 h. The nucleoside (1, 3.04 g, 9.7 mmol) was added from a vial in one portion at -5° C. and the solid was slowly dissolved in 20 minutes. The reaction temperature was allowed to rise to ambient temperature over 2 h. After 17 h, the reaction was not complete. More reagents were made (as described above from phosphate (2.66 g), aminoester (2.60 g), and NMI (3.8 mL, 48 mmol)) and added to the reaction mixture at -5° C. The reaction was stirred at room temperature for 2 more hours. The reaction was almost complete as shown by TLC result and diluted with 70 mL of dichloromethane. HCl solution (1 N, 70 mL) was added. The aqueous layer was separated and extracted with dichlo-

romethane. The organic layer was washed with saturated NaHCO_3 , water, brine and dried over MgSO_4 . After removal of the solvent under reduced pressure, the sticky residue was purified through automated column chromatography using a 240 g cartridge and a gradient of 0-8% 2-PrOH in dichloromethane to afford product as a foam solid (4.16 g, 7.14 mmol, 73% yield). HPLC purity 97.4%. NMR spectra of product showed it is a mixture of two diastereoisomers with a ratio of 1.2:1.

[0106] ^1H -NMR ($\text{DMSO}-d_6$): δ =7.98 (1H, s, 8-H of one isomer), 7.95 (1H, s, 8-H of another isomer), 7.37-7.32 (2H, m, arom-H), 7.22-7.15 (3H, m, arom-H), 6.6 (2H, s, NH_2), 6.11 (1H, d, C1'-H of one isomer), 6.09 (1H, d, C1'-H of another isomer), 6.09-5.98 (1H, m, amide NH), 5.88 (1H, d, 3'-OH of one isomer), 5.81 (1H, d, 3'-H of another isomer), 4.85-4.75 (1H, hepta, methine H of iso-propyl), 4.46-4.27 (2H, m, C4'-H, α -H of amino ester), 4.15-4.07 (1H, m, C3'-H), 3.96 (3H, s, OCH_3), 3.82-3.72 (2H, m, C5'-H $_{\alpha}$ and C5'-H $_{\beta}$), 1.23-1.06 (9H, m, CH_3 's of amino ester), 1.03 (3H, d, C2'- CH_3).

[0107] ^{31}P -NMR ($\text{DMSO}-d_6$): δ =4.91 (one isomer), 4.72 (another isomer).

[0108] An alternate purification method is to chemically alter the minor 3' phosphoramidate by-product in order to simplify the chromatographic separation. The crude phosphoramidate product is dissolved in anhydrous pyridine (5 mL/g), and is treated with 0.5 molar equivalents of t-butyltrimethylsilyl chloride at ambient temperature to react selectively with the free 5' primary hydroxyl of the 3' isomer impurity. Reaction progress can be monitored by LC/MS. Once the 3' isomer is converted to a 5'-tBDMS-3'-phosphoramidate derivative, the reaction is quenched with methanol (3 eq), concentrated under reduced pressure, partitioned between ethyl acetate and 5% citric acid and then the organic layer is concentrated. The residue is then subjected to chromatography which can now be done with a higher loading and a faster gradient and achieve a higher purity.

[0109] Production of 1 via a non-stereoselective process provides for a diastereomeric mixture containing 1 and the Rp-isomer, whereby 1 can be further purified by crystallization of the mixture or by chromatographic separation, as illustrated below.

Example 4

Separation of 1 from a Mixture Containing 1 and the Rp-Isomer

[0110] Separate a total of 11.9 Kg of a diastereomeric mixture containing 1 and the Rp-isomer to obtain an approximate 85% yield of both isomers (1 and Rp-isomer) under cGMP protocols. The diastereomeric purity requirement is >98% d.e. for both diastereomers.

Preparative Chromatographic Conditions

[0111] The chromatographic equipment and separation method are as follows:

[0112] A 5-cm SMB system, consisting of 8 columns, each holding 100 grams of CHIRALPAK®IATM, was used for processing of the diastereomeric mixture. The mobile phase was 100% ethyl acetate.

Separation and Isolation

[0113] The solubility of this material was 75 g/l in the mobile phase. The material was dissolved in the mobile phase at 40° C. with stirring.

[0114] The extract stream from the SMB was first concentrated using a 1/2 sq ft horizontal thin-film evaporator (Protherm) and then dried down in the rotovap at 40° C. The flow rate of the raffinate stream was too low to be handled by the Protherm. The raffinate was directly dried down using the rotovap. The products were then dried to constant weight in a vacuum oven at 40° C. The residual level of ethyl acetate however was found to exceed the desired level (<0.4 wt %) even after constant weight had been reached. After further drying under vacuum at 50° C., the level remained at approximately 1 wt %. The products were re-dissolved in acetone and then evaporated to dryness in the rotovap. This step effectively removed the residual ethyl acetate level to below 0.1 wt %. The products were then further dried in the vacuum oven to reduce the residual acetone to the desired level of <0.4 wt %.

Results

[0115] Extract—1. A quantity of 6,864 g of amorphous 1 was recovered with a diastereomeric purity of 99.2% d.e. Raffinate—The Rp-isomer. A quantity of 3,707 g of the Rp-isomer was recovered, of which 1,844 g had a diastereomeric purity of 98.1% d.e.

Example 5

Kiloscale Crystallization of 1.H₂O from Amorphous Solid 1 (Obtained from SMB Chromatography) with Anisole/Ethyl Acetate/Water

[0116] A 10 L rotary evaporator flask was equipped with a mechanical stirrer. 1000 g of amorphous solid (98.9% HPLC purity, 0.25 molar equiv of water present) was added followed by ethyl acetate (1.00 L) and anisole (99% grade, 4.00 L) at ambient temperature. The suspension was rapidly stirred until all the solid dissolved (15 min). Stirring was slowed to 88 rpm. Crystalline seeds (40 mg) were added, followed by water (31 mL, 1.0 eq). The solution became cloudy after about 30 minutes and showed heavy precipitate after 3-4 h with no visible water remaining. The suspension was stirred for a total of 20 h at ambient temperature. The crystalline solid was collected by vacuum filtration on a 25 cm Buchner funnel. The cake washed with a 50:50 mixture of heptanes and t-butyl methyl ether (3x1 L). The cake washed easily and did not require pressing. After air-drying for 15 min, the solid was transferred to a 8x14" drying pan and dried under vacuum (0.2 mm Hg, 50° C.) to a constant weight (4 h) and then held under vacuum at ambient temperature for 17 h to yield 840 g (ca 82% for the difference in hydration) of fine broken crystalline laths and needles less than 150 micrometers on a side. HPLC purity 99.7%. The apparent melting point showed shrinking starting at 88° C. and melting at 93-100° C. As a hydrate, this represents a combination of dehydration to an amorphous solid and then the phase transition temperature.

[0117] The crystallization was repeated on 990 g more in a proportional manner to yield another 840 g (ca 83%) in the same purity. The filtrates were combined and stripped under reduced pressure to a yellow oil, crude wt 450 g which still contained roughly 150 g of anisole. To this was added ethyl acetate (350 mL), more anisole (850 mL), water (9 g) and seeds (20 mg). The solution was stirred as before. It became cloudy after 4 h and stirred at ambient temperature for 24 h. The solid was collected by filtration and rinsed with 50:50 heptanes and t-butyl methyl ether while mechanically breaking up lumps and washing until a yellow tinge was gone

(4×450 mL). The solid was dried in a similar manner to give 200 g more product in 98.9% HPLC purity which is suitable for an additional recrystallization. The mother liquor was stripped to 175 g of light brown oil (85% HPLC purity) still containing some anisole.

Example 6

Crystallization of 1.H₂O from Amorphous 1 (Obtained from SMB Chromatography) with Heptanes/Ethyl Acetate

[0118] 1 g of the amorphous 1 (98.9% HPLC purity) was dissolved in mixture of ethyl acetate (4 mL) and heptanes (2 mL) by shaking on a Vortex mixer. To this solution was added a few seeds of crystalline product. The suspension was stirred at ambient temperature in an open vessel exposed to atmospheric moisture for 6 h. The solid was collected by filtration and washed with a mixture of ethyl ether and hexanes (1:1, 3×3 mL) and dried to 910 mg of crystalline solid. HPLC purity 99.5%.

Example 7

Crystallization of 1.H₂O from Amorphous 1 (Obtained from SMB Chromatography) with Xylenes/Ethyl Acetate/Water

[0119] 1 g of the amorphous 1 (98.9% HPLC purity) was dissolved ethyl acetate (1.5 mL) and then xylenes was added until cloudy (3.2 mL). The mixture was heated to 45° C. to give a clear solution and then cooled to ambient temperature. Water (50 µL) was added and the solution was stirred with a magnetic stirrer. No seeds were added. After 72 h, the resulting precipitate was collected by filtration, washed with 35% ethyl acetate in xylenes (2.5 mL) and dried (0.2 mm Hg, ambient temperature) to give 950 mg of product as fine white crushed crystals. HPLC purity 99.5%.

Discussion on Purification from Mixture.

[0120] A practical alternative to separating out 1 from a mixture containing 1 and its corresponding Rp-diastereomeric counterpart, by SMB chromatography and then crystallizing it is to crystallize 1 (or 1.H₂O) directly from the mixture. The overall recovery is nearly as high while avoiding the expense and time consumption of the SMB process. Direct crystallization of material produced with a chiral synthesis with a greatly enhanced S_p/R_p ratio is expected to be even more efficient.

Example 8

Crystallization of 1.H₂O from Purified Amorphous Solid of Mixture of Isomers (S_p/R_p (1.6:1))

[0121] A mixture containing 1 and its diastereomeric counterpart (the Rp isomer) (1.0 g) was dissolved in ethyl acetate (1 mL). Anisole (4 mL) was added slowly to form a clear solution with rapid stirring. Water (22 mg, 1.0 eq based on 1) was added followed by crystalline 1 seeds (~2 mg). The solid started to form in 10 min. Stirring was continued for 36 h. White solid was collected by filtration and washed with cold mixture of ethyl ether/hexanes (1:1, 3 mL×3). Yield=520 mg (96% 1 by P-NMR). 470 mg of this material was recrystallized by dissolving in warm ethyl acetate (0.7 mL, 45° C.) over 2 min, then with stirring, adding anisole (2.8 mL) with no additional water or seeds. The solution became cloudy after 5 min. It was stirred for 48 h at ambient temperature. The solid

was collected by filtration in a sintered glass funnel, and washed with a cold mixture of ethyl ether/hexanes (1:1, 2 mL×3) and dried under vacuum to a white solid, 451 mg (>99% pure by P-NMR). Recovery percentage based on the 615 mg of 1 theoretically present in the starting mixture was 73%.

Discussion on Alternative Solvent Mixtures to Crystallize the S_p-Isomer from the Mixture.

[0122] A mixture containing 1 and the Rp-isomer (1 g) dissolved in ethyl acetate (4 mL/g) with a molar equivalent of water followed by evaporation to ca 1 mL/g yielded 396 mg of 93% pure material. A 375 mg sample of this was recrystallized from ethyl acetate (2.5 mL) and an equivalent of water to yield 275 mg of 99.1% pure material. Another 1 g of the diastereomeric mixture was crystallized from ethyl acetate/heptanes 2:1 (6.5 mL/g) with one equivalent of water to give 433 mg of 95% purity.

[0123] It is contemplated that 1.H₂O can be obtained by crystallization of crude mixtures of 1 and the R_p-isomer.

[0124] Alternatively, the S_p-isomer can also be isolated without any chromatography through direct crystallization of the monohydrate from the crude product formed from the chlorophosphate reagent (see non-stereoselective process above). Anisole-water is a suitable solvent combination to initiate the crystallization followed by dilution with ethyl ether. Other solvent combinations such as used on the purified 1 could also be used.

Example 9

Isolation of 1 by Crystallization Directly from the Crude Product

[0125] To a 50 mL dry round-bottomed flask were loaded phenyl dichlorophosphate (2.68 g, 12.7 mmol) and anhydrous dichloromethane (40 mL). The amino ester salt (2.58 g, 15.4 mmol) was added to the solution and the mixture was cooled to -30° C. N-Methyl imidazole (6.3 g, 76 mmol) was then added quickly via a dry syringe at -30° C. and the solution was stirred at -5° C. for 20 min. The nucleoside (7, 2.00 g, 6.38 mmol) was added from a vial in one portion at -5° C. and the solid was slowly dissolved in 20 minutes. The reaction temperature was allowed to rise to ambient temperature and stirred for 1 h. TLC indicated approximately 95% completion. The reaction was diluted dichloromethane (50 mL) and washed with 1 N HCl (2×50 mL), 1:1 mixture of sat'd bicarbonate and brine, dried over sodium sulfate (5 g), filtered, concentrated under reduced pressure and then high vacuum to 4.5 g of crude product. P-NMR indicated a 1.5:1 mixture of the S_p/R_p isomers.

[0126] A small portion (100 mg) of the crude oil was dissolved in anisole (0.30 mL) and the solution was filtered through a syringe filter (22 micron). Water (8 mg) was added slowly to the stirred solution at ambient temperature followed by seed crystals (ca 1 mg). The solution became cloudy in 5 min. Ethyl ether (1 mL) was dropped in slowly and the suspension was stirred for 2 h and then the precipitate was collected by filtration, washed with a cold 1:1 mixture of ether ether-hexane (3×1 mL) and then dried under vacuum at ambient temperature to 17 mg of the Sp-isomer. NMR purity 98%. This represents a yield of 21% from the starting nucleoside and 35% of the theoretical yield of the amount of 1 formed in the crude mixture.

Discussion of Solid State Characterization.

[0127] Single crystal x-ray from methylene chloride/hexanes/atmospheric water indicated a single water molecule

incorporated into the crystal structure. Samples from three solvent systems (anisole/ethyl acetate, heptanes/ethyl acetate, xylenes/ethyl acetate) all showed the same crystalline form by XRPD. The anisole sample was used for additional studies. Karl Fisher analysis showed 3.1% water which corresponds to a monohydrate. TGA analysis indicated a loss of a half hydrate between 80-100° C. and the second half hydrate at 110 to 170° C. DSC under heating at 10° C./min in an open pan indicated an endotherm starting at 75° C. for the dehydration with an onset of 93.56° C. and peak at 100.24° C. Variable temperature XRPD showed a loss of crystallinity between 90 and 100° C. GVS analysis showed very low hygroscopicity of the monohydrate with a reversible 0.47% w/w gain at 90% RH and stability of the monohydrate at 0% RH.

Example 10

XRPD of 1.H₂O (Form 1)

Experiment

[0128] X-Ray Powder Diffraction patterns were collected on a Bruker D8 diffractometer using Cu K α radiation (40 kV, 40 mA), 0-20 goniometer, and divergence of V4 and receiving slits, a Ge monochromator and a Lynxeye detector. The instrument is performance checked using a certified Corundum standard (NIST 1976). The software used for data collection was Diffrac Plus XRD Commander v2.5.0 and the data were analysed and presented using Diffrac Plus EVA v 11.0.0.2 or v 13.0.0.2.

[0129] Samples were run under ambient conditions as flat plate specimens using powder as received. Approximately 20 mg of the sample was gently packed into a cavity cut into polished, zero-background (510) silicon wafer. The sample was rotated in its own plane during analysis. The details of the data collection are:

[0130] Angular range: 2 to 42 °2 θ

[0131] Step size: 0.05 °2 θ

[0132] Collection time: 0.5 s.step⁻¹

[0133] An XRPD pattern was obtained providing the following °2 θ /° intensity data:

TABLE 1

XRPD data of 1•H ₂ O.			
Angle 2-Theta °	Intensity %	Angle 2-Theta °	Intensity %
8.0	9.6	23.4	18.8
8.7	48.2	24.4	9.1
9.1	12.2	24.7	21.0
10.3	6.0	25.2	10.8
11.1	30.5	25.5	7.1
11.3	11.6	25.9	20.6
11.8	7.7	26.5	5.5
13.6	34.9	27.0	9.9
14.1	19.2	27.1	13.7
14.8	100.0	27.4	23.8
15.3	6.4	28.1	8.6
15.6	5.1	28.5	11.5
16.1	18.2	28.9	5.1
16.8	24.2	29.8	6.9
17.6	99.8	30.1	7.7
17.8	37.5	30.5	7.4
18.8	4.7	31.7	7.4
19.4	8.3	32.7	5.0
20.4	64.0	33.4	5.4
21.7	8.5	33.9	4.1

TABLE 1-continued

XRPD data of 1•H ₂ O.			
Angle 2-Theta °	Intensity %	Angle 2-Theta °	Intensity %
22.1	23.2	35.0	5.0
23.0	12.0	36.1	5.5

[0134] FIG. 1 contains an XRPD spectrum of 1.H₂O.

Example 11

X-ray Structure Determination of 1.H₂O

[0135] Compound 1.H₂O, C₂₄H₃₄N₆PO₉F, crystallizes in the orthorhombic space group P2₁2₁2₁ (systematic absences h00: h=odd, 0k0: k=odd, and 001: l=odd) with a=10.9918(8) Å, b=13.0925(9) Å, c=20.3570(13) Å, V=2929.6(3) Å³, Z=4, and d_{calc}=1.362 g/cm³. X-ray intensity data were collected on a Bruker APEXII CCD area detector employing graphite-monochromated Mo-K α radiation (k=0.71073 Å) at a temperature of 143(1)K. Preliminary indexing was performed from a series of thirty-six 0.5° rotation frames with exposures of 30 seconds. A total of 1790 frames were collected with a crystal to detector distance of 37.522 mm, rotation widths of 0.5° and exposures of 30 seconds:

scan type	2 θ	ω	ϕ	χ	frames
ϕ	-15.50	258.48	-351.72	19.46	739
ω	-5.50	2.88	-8.86	-31.86	116
ϕ	-10.50	300.13	18.75	39.97	196
ϕ	19.50	59.55	-11.29	-16.16	739

[0136] Rotation frames were integrated using SAINT (Bruker (2009) SAINT. Bruker AXS Inc., Madison, Wis., USA.), producing a listing of unaveraged F² and σ (F²) values which were then passed to the SHELXTL (Bruker (2009) SHELXTL. Bruker AXS Inc., Madison, Wis., USA.) program package for further processing and structure solution on a Dell Pentium 4 computer. A total of 47654 reflections were measured over the ranges 1.85° ≤ θ ≤ 25.06°, -13° ≤ h ≤ 13, -15° ≤ k ≤ 15, -24° ≤ l ≤ 22 yielding 5187 unique reflections (Rint=0.0261). The intensity data were corrected for Lorentz and polarization effects and for absorption using SADABS ((Sheldrick, G. M. (2007) SADABS. University of Gottingen, Germany.) (minimum and maximum transmission 0.6749, 0.7452).

[0137] The structure was solved by direct methods (SHELXS-97 (Sheldrick, G. M. (2008) Acta Cryst. A64, 112-122)). Refinement was by full-matrix least squares based on F² using SHELXL-97 (Sheldrick, G. M. (2008) Acta Cryst. A64, 112-122.) All reflections were used during refinement. The weighting scheme used was w=1/[(σ^2 (F_o²)+(0.0413P)²+0.4916P] where P=(F_o²+2F_c²)/3. Non-hydrogen atoms were refined anisotropically and hydrogen atoms were refined using a riding model. Refinement converged to R1=0.0244 and wR2=0.0657 for 5000 observed reflections for which F>4 σ (F) and R1=0.0259 and wR2=0.0669 and GOF=1.059 for all 5187 unique, non-zero reflections and 377 variables (R1=[$\sum ||F_o| - |F_c|| / \sum |F_o|$]; wR2=[$\sum w(F_o^2 - F_c^2)^2 / \sum w(F_o^2)^2$]^{1/2}; z; GOF=[$\sum w(F_o^2 - F_c^2)^2 / (n-p)$]^{1/2}; where n=the number of reflections and p=the number of parameters refined.) The maximum Δ/σ in the final cycle of least squares was 0.001 and the two most prominent peaks in the final difference Fourier were +0.170 and -0.248 e/Å³.

[0138] Table 2 lists cell information, data collection parameters, and refinement data. Final positional and parameters are given in Table 3. FIG. 2 is an ORTEP ("ORTEP-II: A Fortran Thermal Ellipsoid Plot Program for Crystal Structure

Illustrations.” C. K. Johnson (1976) ORNL-5138) representation of the 1.H₂O with 30% probability thermal ellipsoids displayed.

TABLE 2

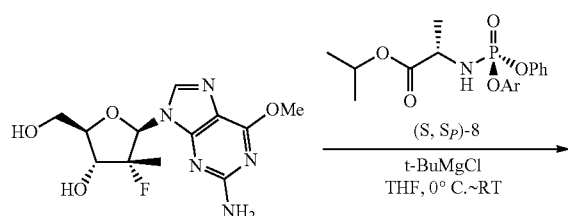
Summary of Structure Determination of 1•H ₂ O	
Empirical formula	C ₂₄ H ₃₄ N ₆ O ₉ F
Formula weight	600.54
Temperature	143(1) K
Wavelength	0.71073 Å
Crystal system	orthorhombic
Space group	P2 ₁ 2 ₁ 2 ₁
Cell constants:	
a	10.9918(8) Å
b	13.0925(9) Å
c	20.3570(13) Å
Volume	2929.6(3) Å ³
Z	4
Density (calculated)	1.362 Mg/m ³
Absorption coefficient	0.160 mm ⁻¹
F(000)	1264
Crystal size	0.22 × 0.18 × 0.10 mm ³
Theta range for data collection	1.85 to 25.06°
Index ranges	-13 ≤ h ≤ 13, -15 ≤ k ≤ 15, -24 ≤ l ≤ 22
Reflections collected	47654
Independent reflections	5187 [R(int) = 0.0261]
Completeness to theta = 25.06°	99.8 %
Absorption correction	Semi-empirical from equivalents
Max. and min. transmission	0.7452 and 0.6749
Refinement method	Full-matrix least-squares on F ²
Data/restraints/parameters	5187/0/377
Goodness-of-fit on F ²	1.059
Final R indices [I > 2sigma(I)]	R1 = 0.0244, wR2 = 0.0657
R indices (all data)	R1 = 0.0259, wR2 = 0.0669
Absolute structure parameter	0.03(6)
Largest diff. peak and hole	0.170 and -0.248 e.Å ⁻³

[0139] The following embodiments provide examples where various phosphoramidate reagents are used to prepare 1, its R_P-isomer, or a diastereomeric mixture of 1 and its R_P-isomer.

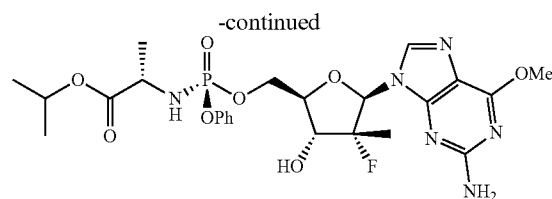
Example 12-1

Synthesis of (S)-2-[(S)-[(1R,4R,5R)-5-(2-Amino-6-methoxy-purin-9-yl)-4-(R)-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid isopropyl ester monohydrate (1) via (S)-isopropyl 2-(((S)-(perfluorophenoxy)(phenoxy)phosphoryl)amino)propanoate ((S, S_P)-8) and isolation by chromatography and crystallization

[0140]



2



Ar = —C₆F₅

a) Preparation of (S)-2-[(2,3,4,5,6-pentafluoro-phenoxy)-phenoxy-phosphorylamino] propionic acid isopropyl ester ((S, S_P)-8 and (S, R_P)-8) and isolation of (S)-2-[(S)-[(2,3,4,5,6-pentafluoro-phenoxy)-phenoxy-phosphorylamino] propionic acid isopropyl ester ((S, S_P)-8) via crystallization-induced dynamic resolution in a single crop

[0141] To a 1 L of dry three-necked flask fitted with a low-temperature thermometer and a mechanical stirrer was loaded phenyl phosphorodichloridate (25 g, 118.5 mmol). Anhydrous dichloromethane (125 mL) was added and the solution was cooled to 0° C. The alanine ester salt (oven dried) (19.86 g, 1 eq) was added quickly under N₂ while agitated. The solution was cooled to ca -50° C. (internal temperature (in an acetone/dry ice bath under N₂)). A solution of triethylamine (25.2 g, 2.1 eq) in DCM (125 mL) was added dropwise via an addition funnel over 0.5 h at -50° C. and the resulting white slurry was stirred at about -50° C. for 0.5 h. The mixture was allowed to warm up to 0° C. over 1.5 h and then a pre-mixed cooled solution of pentafluorophenol (21.82 g, 1 eq) and TEA (13.2 g, 1.1 eq) (caution: heat released while mixing pentafluorophenol and TEA) in 75 mL of DCM was added over 0.5 h at 0° C. via an addition funnel. The mixture was stirred at 0° C. for additional 4 h.

[0142] The mixture was filtered through a Buchner funnel and the collected solid triethylamine hydrochloride was rinsed with DCM (3×40 mL). The filtrate was checked by ³¹P-NMR (ratio ca 1.14:1 favored the S_P-diastereomer—downfield peak) and was divided into two parts of equal weight. One of them was concentrated under reduced pressure. The white solid residue (31 g) was triturated in a mixture of EtOAc and hexanes (150 mL, 20:80, v/v) at RT for 17 h allowing time for dynamic resolution of the less soluble S_P-isomer. The white slurry was filtered and solid was rinsed with 20% EtOAc in hexanes (2×25 mL). The solid (22.58 g) was checked by ¹H-NMR and ³¹P-NMR and it contained product as one isomer contaminated with triethylamine hydrochloride salt. The solid was dissolved and partitioned in 310 mL of EtOAc and 100 mL of water. After separation of the organic layer, the aqueous layer was back-extracted with EtOAc (50 mL). The combined organic layer was washed with water (3×80 mL), brine (50 mL) and dried over MgSO₄. The solution was concentrated under reduced pressure and then dried under high vacuum at RT to a constant weight to furnish 17.36 g of product as a white solid from the one half of the reaction. The yield is 64%. The mother liquor from above was concentrated to a gummy residue (7.89 g) that contained the reagents with a ratio of 1:1.2 ((S, S_P)-8/(S, R_P)-8) based on ³¹P-NMR.

b) Preparation of 1 from (S, S_P)-8 and 2

[0143] To a 250 mL of dry three-necked round flask was added 5.06 g (16.15 mmol) of the purine nucleoside (2). The

solid was suspended in 40 mL of anhydrous THF and cooled in an ice-water bath. The Grignard reagent (1 M solution in THF) was added dropwise via a syringe and a clear solution was formed. The mixture was stirred at 0° C. for 30 minutes and a solution of (S, S_p)-8 (8.32 g, 18.35 mmol) in 40 mL of THF was added via an addition funnel over 50 minutes. After finishing addition, the reaction mixture was stirred at room temperature for 3 hours. The reaction was quenched by adding 20 mL of sat NH₄Cl at 0° C. The mixture was diluted with 100 mL of ethyl acetate. Two layers were separated and aqueous layer was extracted with 50 mL of ethyl acetate. Organic layer was combined and washed with water (60 mL), sat sodium bicarbonate (2×60 mL), water (60 mL), brine (40 mL), and dried over sodium sulfate. Solvent was removed under reduced pressure to afford an amorphous solid residue.

[0144] To the crude residue 7 mL of ethyl acetate was added and followed by 26 mL of anisole. The mixture was stirred until a solution was formed. Water (320 mg) was added and 20 mg of crystal seeds of product (1) was added. The mixture was cooled at -5° C. overnight. White solid was formed and collected by filtration. Solid was rinsed with pre-cooled mixture of heptane and TBME (1:1, 3×2 mL) and weighed 3.3 g after drying. The mother liquor was concentrated under reduced pressure and the residue was purified via column chromatography (5~7% 2-propanol in DCM). Product was obtained as a white amorphous solid (4.5 g).

[0145] Solids from above were combined (7.8 g) and mixed with 7.7 mL of ethyl acetate. To the slurry, 31 mL of anisole was added and the mixture was stirred until a uniform solution was formed. To the solution 160 mg of water was added and followed by 20 mg of crystal seeds of product (1). The mixture was stirred slowly at room temperature and white solid precipitated. The mixture was kept at -5° C. for 2 hours and solid was collected via filtration. Solid was rinsed with pre-cooled mixture of heptane and TBME (1:1, 4×5 mL) and dried in vacuo. Product weighed 6.69 g (69% yield).

Example 12-2

Synthesis of (S)-2-[(S)-[(1R,4R,5R)-5-(2-Amino-6-methoxy-purin-9-yl)-4-(R)-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid isopropyl ester monohydrate (1) via (S)-isopropyl 2-(((S)-(perfluorophenoxy)(phenoxy)phosphoryl)amino)propanoate ((S, S_p)-8) and isolation by crystallization only

[0146] To a 250 mL of dry three-necked round flask were loaded 5 g (15.96 mmol) of the nucleoside and 40 mL of anhydrous THF. The suspension was cooled in an ice-water bath and 20 mL of the Grignard reagent (1 M solution in THF, 20 mmol) was added via a syringe over 10 minutes. The clear reaction mixture was stirred at 0° C. for half hour and then a solution of the phosphorus reagent ((S, S_p)-8) in 40 mL of THF was added via an addition funnel in 2 hours. The reaction was allowed to warm up to ambient temperature slowly and stirred for overnight. The mixture was cooled to 0° C. and 50 mL of 1 N diluted HCl was added. Most of THF was removed under reduced pressure and the mixture was diluted with 200 mL of ethyl acetate. The organic layer was separated and aqueous layer was extracted with 30 mL of ethyl acetate. The combined organic layer was washed with water (60 mL), sat'd. sodium bicarbonate (2×50 mL), 5% sodium carbonate (70 mL), water (50 mL), and brine (50 mL). Organic solution

was dried over magnesium sulfate and solvent was removed under reduced pressure to afford an amorphous solid residue.

[0147] The crude residue was dissolved in 41 mL of anisole at room temperature. To the solution, 24 mL of xylenes was added and followed by 410 mg of water. The mixture was stirred slowly at room temperature and crystal seeds of 1 (10 mg) were added. White solid precipitated and the mixture was kept at -5° C. for 2 hours. Solid was collected via filtration and rinsed with a pre-cooled mixture of heptane and TBME (1:1, 3×2 mL). Solid weighed 5.83 g after drying. The mother liquor was concentrated to dryness under reduced pressure. The residue was dissolved in 7.2 mL of anisole and 10.7 mL of xylenes was added. To the solution, 178 mg of water was added and 5 mg of crystal seeds of 1 were added. The mixture was slowly stirred at room temperature for overnight. White solid was formed and collected via filtration. Solid was rinsed with a pre-cooled mixture of heptane and TBME (1:1.3×1 mL) and weighed 1.17 g.

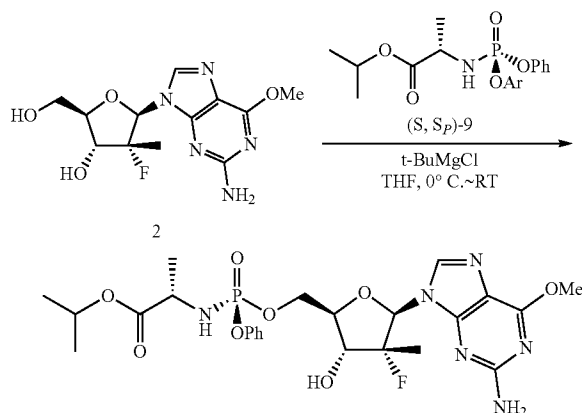
[0148] Solids obtained above were combined (7.0 g) and added 7 mL of ethyl acetate. After addition of 27 mL of anisole, a clear solution was formed. To the solution, 200 mg of water was added and then added 5 mg of crystal seeds of 1. The mixture was stirred at ambient temperature and white solid precipitated. The mixture was kept at -5° C. for overnight. Crystalline solid was collected by filtration and rinsed with a pre-cooled mixture of heptane and TBME (1:1, 3×5 mL). The resultant product (1) weighed 5.66 g with purity of 98.3% by HPLC.

[0149] The above solid was purified again via crystallization from a combination of 5.6 mL ethyl acetate and 22.6 mL of anisole. After filtration and drying, 4.48 g (47%) of product was obtained and purity was 99.18% by HPLC. Spectral (¹H- and ³¹P-NMR, MS) and physical properties (HPLC retention, melting point and appearance) matched an authentic sample.

Example 12-3

Synthesis of (S)-2-[(S)-[(1R,4R,5R)-5-(2-Amino-6-methoxy-purin-9-yl)-4-(R)-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid isopropyl ester (1) via (S)-isopropyl 2-(((S)-(2,4-dinitrophenoxy)(phenoxy)phosphoryl)amino)propanoate ((S, S_p)-9)

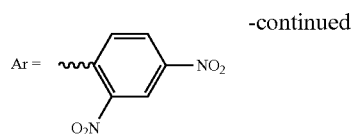
[0150]



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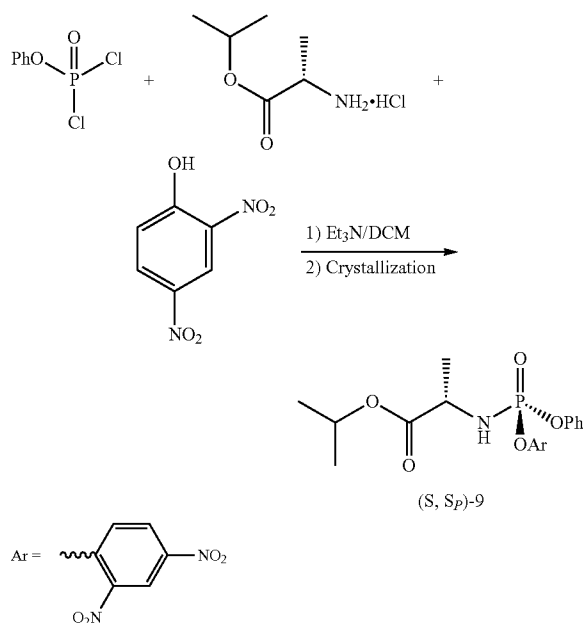
Oct. 20, 2011

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a) Preparation of (2*S*)-isopropyl 2-(((2,4-dinitrophenoxy)(phenoxy)phosphoryl)amino)propanoate diastereomeric mixture ((*S*, *S_P*)-9 and (*S*, *R_P*)-9) and isolation of the single isomer (2*S*)-isopropyl 2-(((*S*)-(2,4-dinitrophenoxy)(phenoxy)phosphoryl)amino)propanoate ((*S*, *S_P*)-9) by crystallization

[0151]



[0152] Phenyl phosphorodichloridate (10.0 g, 47.4 mmol) was dissolved in 60 mL of dry DCM and subsequently cooled to -78°C . A premixed solution of 2,4-dinitrophenol (8.72 g, 47.4 mmol) and triethylamine (7.27 mL, 52.1 mmol) in 20 mL of DCM was slowly added at -78°C over a period of 30 min. The reaction was brought to 0°C and stirred for 2.5 h at this temperature before (L)-alanine isopropyl ester (7.95 g, 47.4 mmol) was added as a solid in one batch. Stirring for 40 min at 0°C was then followed by addition of more triethylamine (13.9 mL, 99.54 mmol) and additional stirring for 3 h at 0°C or until judged complete by TLC (ethyl acetate/hexane=1/3). The reaction mixture was subsequently evaporated under reduced pressure, residue redissolved in MTBE (100 mL), solids filtered off and filtrate evaporated to dryness to give yellow syrup. NMR of the crude sample indicated mixture of 2 isomers ((*S*, *S_P*)-9 and (*S*, *R_P*)-9) in the ratio of 1:1. A mixture of EtOAc:Hexanes (1:1) (50 mL) was added and mixture allowed to stir for 15 h. The white solid thus formed was filtered off and rinsed with EtOAc:Hexanes (1:1) (20 mL) and dried under vacuum to give 6.0 g (28%) of (*S*, *S_P*)-9 a single isomer.

[0153] Data: ^1H NMR (CDCl_3 , 400 MHz) δ : 8.82-8.81 (m, 1H), 8.43-8.40 (m, 1H), 7.89-7.86 (m, 1H), 7.36-7.32 (m, 2H), 7.23-7.19 (m, 3H), 4.96 (hepta, 1H), 4.19-4.08 (m, 2H), 1.42 (d, 3H), 1.20 (d, 6H). ^{31}P NMR (CDCl_3 , 162 MHz) δ : -1.82.

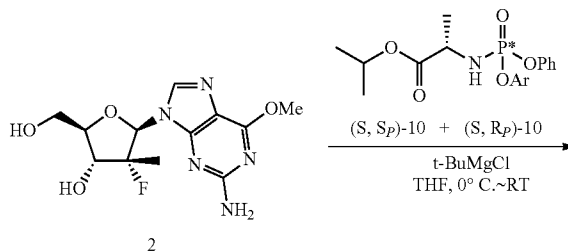
b) Preparation of 1 from (*S*, *S_P*)-9 and 2

[0154] To a 50 mL of dry round-bottomed flask were added 80 mg (0.255 mmol) of 2 and 1 mL of anhydrous THF. The suspension was cooled in an ice water bath and 0.33 mL of Grignard reagent was added via a syringe under nitrogen. A clear solution was formed and stirred at 0°C for half hour. A solution of (*S*, *S_P*)-9 (133 mg, 0.294 mmol) in 1.5 mL of THF was added via a syringe. The orange-colored, clear, reaction mixture was checked by TLC in 20 minutes at 0°C and the reaction was almost complete. Product was formed as well as the 3',5'-bisphosphoramidate by-product. The reaction was quenched by adding sat NH_4Cl after one and half hour. The mixture was diluted with 20 mL of ethyl acetate. Organic layer was separated and aqueous layer was extracted with ethyl acetate (20 mL). The combined organic layer was washed with water (50 mL), sat sodium bicarbonate (2x40 mL), sat sodium carbonate (40 mL), water (40 mL), and brine (30 mL). The light yellow color organic layer was dried over sodium sulfate. The solution was concentrated under reduced pressure and an amorphous solid residue resulted was purified via column chromatography. The bis-phosphoramidate by-product was eluted out at 1% methanol in DCM as a foam solid (32.4 mg) and 1 was eluted out at 3% methanol in DCM (74 mg, 0.127 mmol, 49.6%).

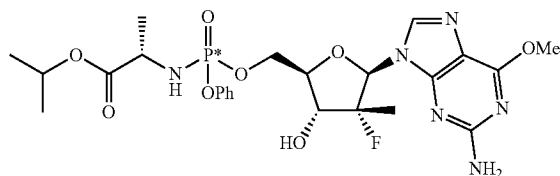
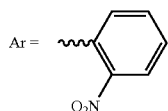
Example 12-4

Synthesis of (*S*)-2-[[[(1*R*,4*R*,5*R*)-5-(2-Amino-6-methoxy-purin-9-yl)-4-(*R*)-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid isopropyl ester diastereomeric mixture (1 and β isomer) via (2*S*)-isopropyl 2-(((2-nitrophenoxy)(phenoxy)phosphoryl)amino)propanoate ((*S*, *S_P*)-10 and (*S*, *R_P*)-10)

[0155]



-continued

1 and R_P -isomera) Preparation of ((S, S_P)-10 and (S, R_P)-10)

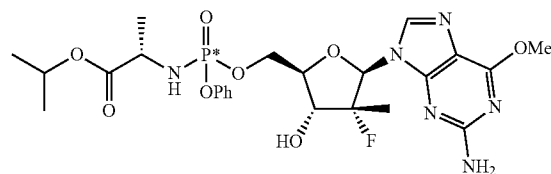
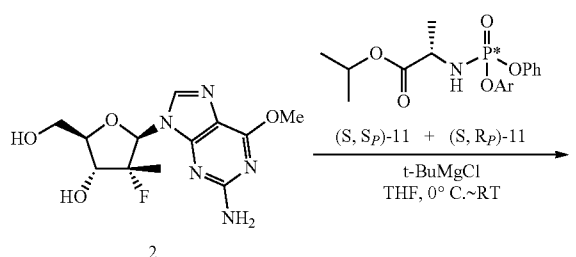
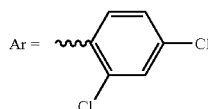
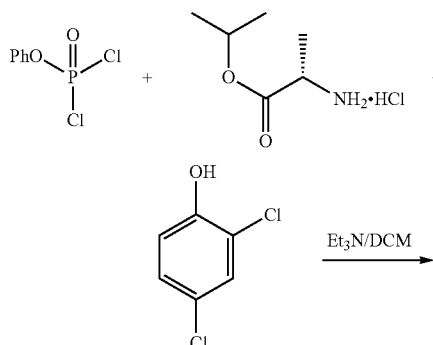
[0156] To a solution of phenyl phosphorodichloridate (30 g, 142.2 mmol) in dichloromethane (150 mL) at -70°C . under nitrogen atmosphere was added drop wise a pre-prepared mixture of o-Nitro phenol (19.76 g, 142.2 mmol) and triethylamine (19.8 mL, 142.2 mmol) in dichloromethane (150 mL) through addition funnel for 1 h at above temperature. Stirring was continued for additional 2 h and was slowly brought to 0°C . L-alanine isopropyl ester hydrochloride salt (26.2 g, 156.3 mmol) was added as solid and then followed by triethylamine (43.7 mL, 313.4 mmol) in dichloromethane (150 mL) drop wise at 0°C . for 20 min. and the reaction mass was continued stirring at the same temperature for additional one hour. The reaction mixture was filtered and concentrated and was finally purified by column chromatography (20% EtOAc/hexanes) on a silica gel to yield ((S, S_P)-10 and (S, R_P)-10) as diastereomeric mixture (14.4 g, 25%). ^1H NMR (CDCl_3 , 400 MHz) δ : 7.94-7.90 (m, 1H), 7.67-7.63 (m, 1H), 7.57-7.54 (m, 1H), 7.33-7.26 (m, 3H), 7.23-7.14 (m, 3H), 5.04-4.89 (m, 1H), 4.21-4.04 (m, 2H), 1.38 (d, 3H, isomer I), 1.33 (d, 3H, isomer II), 1.23-1.17 (m, 6H). ^{31}P NMR (CDCl_3 , 162 MHz) δ : -1.55 (isomer I), -1.76 (isomer II).

b) Preparation of Diastereomeric Mixture of 1 and its R_P -Isomer from ((S, S_P)-10 and (S, R_P)-10) and 2

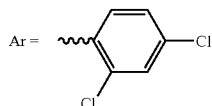
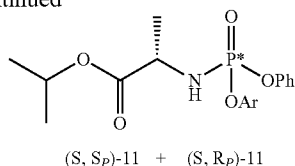
[0157] To a 50 mL of dry round-bottomed flask were added 80 mg (0.255 mmol) of 2 and 1 mL of anhydrous tetrahydrofuran. The suspension was cooled in an ice-water bath and a solution of Grignard reagent (1 M in THF, 0.32 mmol) was added via a syringe. The clear solution thus formed was stirred at 0°C . for half hour and then a solution of phosphorus reagent (120 mg, 0.296 mmol, mixture of isomers) in 1 mL of THF was added dropwise at 0°C . The mixture was stirred at room temperature for 44 hours and quenched by addition of 1 N diluted HCl. After aqueous work-up as usual, the crude residue was purified via column chromatography (silica gel, 3% methanol in DCM) to afford 33.9 mg (0.058 mmol, 22.8%) of 1 and its R_P -isomer as a 1:1 mixture of two isomers.

Example 12-5

Synthesis of (S)-2-[[[(1R,4R,5R)-5-(2-Amino-6-methoxy-purin-9-yl)-4-(R)-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid isopropyl ester diastereomeric mixture (1 and its R_P -isomer) via diastereomeric mixture of (2S)-isopropyl 2-(((2,4-dichlorophenoxy)(phenoxy)phosphoryl)amino)propanoate ((S, S_P)-11 and (S, R_P)-11)

[0158]1 and R_P -isomera) Preparation of (2S)-isopropyl 2-(((2,4-dichlorophenoxy)(phenoxy)phosphoryl)amino)propanoate diastereomeric mixture ((S, S_P)-11 and (S, R_P)-11)**[0159]**

-continued



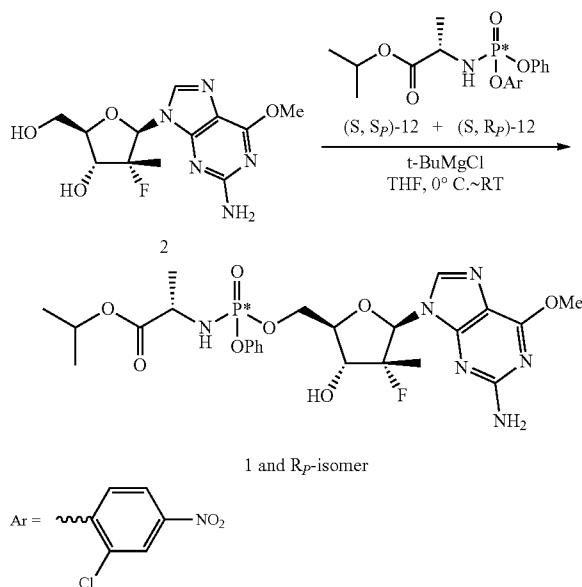
[0160] Phenyl phosphorodichloridate (10.0 g, 47.4 mmol) was dissolved in 60 mL of dry DCM and subsequently cooled to -78°C . Slow addition of a preformed mixture of 2,4-dichlorophenol (7.73 g, 47.4 mmol) and triethylamine (7.27 mL, 52.1 mmol) in 20 mL of DCM was followed by stirring at above temperature for 30 min. The reaction was brought to 0°C and stirred for 2.5 h at this temperature before (L)-alanine isopropyl ester (7.95 g, 47.4 mmol) was added as a solid in one batch. Stirring for 40 min at 0°C was then followed by addition of more triethylamine (13.9 mL, 99.54 mmol) and additional stirring for 3 h at 0°C or until judged complete by TLC (ethyl acetate/hexane=1/3). The reaction mixture was subsequently evaporated under reduced pressure and finally submitted to column chromatography (ethyl acetate in hexane) on silica gel to yield the product (mixture of two isomers) in 66% yield (13.6 g) as a viscous colorless oil. Data: ^1H NMR (CDCl_3 , 400 MHz) δ : 7.47-7.44 (m, 1H), 7.42-7.39 (m, 1H), 7.35-7.30 (m, 2H), 7.24-7.15 (m, 3H), 5.05-4.94 (m, 1H), 4.19-4.08 (m, 1H), 3.96-3.89 (m, 1H), 1.41-1.35 (m, 1H), 1.24-1.19 (m, 6H). ^{31}P NMR (CDCl_3 , 162 MHz) δ : -1.52 (one isomer), -1.54 (the other isomer).

b) Preparation of Diastereomeric Mixture of 1 and its *R_P*-Isomer from ((S, *S_P*)-11 and (S, *R_P*)-11) and 2

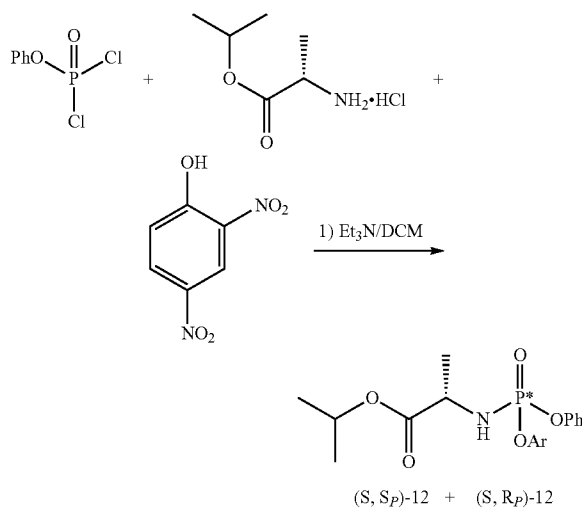
[0161] To a dry 50 mL of round-bottomed flask were added 181 mg (0.58 mmol) of 2 and 1.5 mL of anhydrous THF. The suspension was cooled in an ice-water bath. Grignard reagent (1 M solution in THF, 0.72 mmol) was added via a syringe dropwise over 5 minutes at 0°C . The clear solution was stirred at room temperature for half hour before a solution of ((S, *S_P*)-11 and (S, *R_P*)-11) (276 mg, 0.66 mmol) in 1.5 mL of THF was added over 10 minutes. The reaction was allowed to warm up to ambient temperature and stirred for 22 hours. Reaction was not complete and less than half of starting material was consumed. The reaction was quenched after additional three days by adding sat NH_4Cl (5 mL). The mixture was diluted with 20 mL of ethyl acetate. After work-up, the residue was purified via column chromatography (silica gel, 4% 2-propanol in DCM) to afford 63.1 mg (0.108 mmol, 19%) of 1 and its *R_P*-isomer as a mixture of two diastereomers. From column, 29.6 mg (0.094 mmol) of starting nucleoside was recovered.

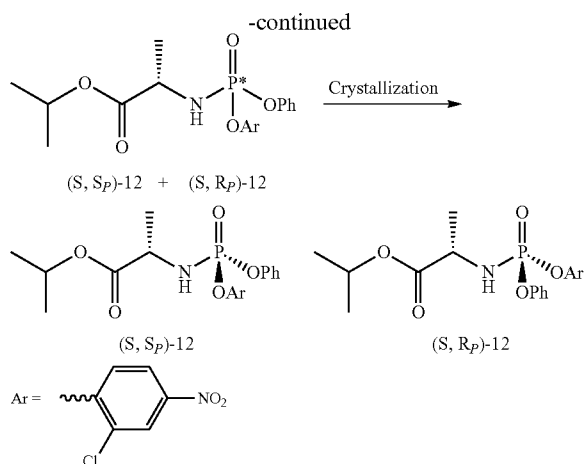
Example 12-6

Synthesis of (S)-2-[[[(1R,4R,5R)-5-(2-Amino-6-methoxy-purin-9-yl)-4-(R)-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid isopropyl ester diastereomeric mixture (1 and *R_P*-isomer) via (2S)-isopropyl 2-(((2-chloro-4-nitrophenoxy)(phenoxy)phosphoryl)amino)propanoate ((S, *S_P*)-12 and (S, *R_P*)-12)

[0162]

a) Preparation of ((S, *S_P*)-12 and (S, *R_P*)-12) and isolation of (S, *S_P*)-12 and (S, *R_P*)-12

[0163]



[0164] Phenyl phosphorodichloridate (10.0 g, 47.3 mmol) was dissolved in 50 mL of dry DCM and subsequently cooled to 0° C. After addition of solid (L)-alanine isopropyl ester HCl salt (7.94 g, 47.3 mmol), the reaction mixture was cooled to -70° C. and then treated with triethylamine (13.8 mL, 94.6 mmol) dissolved in 50 mL of dry DCM. The resulting mixture was stirred for 30 min at this temperature before being allowed to warm to 0° C. Subsequently, a preformed solution of 2-chloro-4-nitrophenol (8.2 g, 47.3 mmol) and triethylamine (6.6 mL, 47.3 mmol) dissolved in 20 mL of dry DCM was added over 5-10 min and was continued stirring for additional 2 h. The solution was filtered and the filtrate was concentrated under reduced pressure. The resulting residue was suspended in 50 mL of TBME and stirred for 10 min at room temperature. Subsequent filtration removed more triethylamine hydrochloride and yielded a filtrate that was again stripped of its solvent under reduce pressure. Column chromatography (dichloromethane) yielded the product (12.2 g, 27.6 mmol) as solid. The product was recrystallized using EtOAc/hexane (2:3) for two times to isolate (S, R_P)-12 (5.2 g, 25% yield) and upon cooling the mother liquor to -5° C. (S, S_P)-12 was obtained (1.5 g, 7% yield).

[0165] (S, S_P)-12 Data: ¹H NMR (CDCl₃, 400 MHz) δ: 8.33 (m, 1H), 8.13-8.10 (m, 1H), 7.73-7.71 (m, 1H), 7.36-7.33 (m, 2H), 7.25-7.18 (m, 3H), 5.00 (hepta, 1H), 4.19-4.10 (m, 1H), 4.02-3.97 (m, 1H), 1.43 (d, 3H), 1.23-1.21 (m, 6H).

[0166] ³¹P NMR (CDCl₃, 162 MHz) δ: -1.97.

[0167] (S, R_P)-12 Data: ¹H NMR (CDCl₃, 400 MHz) δ: 8.32-8.31 (m, 1H), 8.13-8.10 (m, 1H), 7.73-7.71 (m, 1H), 7.38-7.34 (m, 2H), 7.28-7.19 (m, 3H), 5.02 (hepta, 1H), 4.21-4.11 (m, 1H), 4.01-3.95 (m, 1H), 1.40 (d, 3H), 1.25-1.22 (m, 6H).

[0168] ³¹P NMR (CDCl₃, 162 MHz) δ: -2.02.

b) Preparation of Diastereomeric Mixture of 1 and its R_P-Isomer from ((S, S_P)-12 and (S, R_P)-12) and 2

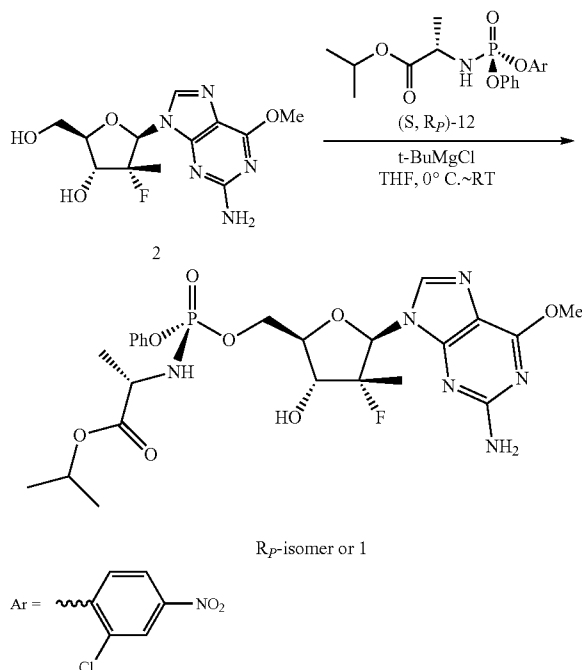
[0169] To a dry 50 mL of round-bottomed flask were added 181 mg (0.58 mmol) of 2 and 1.5 mL of anhydrous THF. The suspension was cooled in an ice-water bath under nitrogen. Grignard reagent (1 M solution in THF, 0.72 mmol) was added via a syringe and a clear solution was formed. The mixture was stirred at ambient temperature for half hour and then cooled to 0° C. again. A solution of ((S, S_P)-12 and (S, R_P)-12) (292 mg, 0.66 mmol) in 1.5 mL of THF was added via

a syringe over 10 minutes at 0° C. The resulting orange color reaction solution was stirred at room temperature for overnight (19 h) and reaction was almost complete as checked by TLC. The reaction was quenched by addition of sat NH₄Cl (5 mL) and diluted with 20 mL of ethyl acetate and 10 mL of water. Two layers were separated and aqueous layer was extracted with 20 mL of EtOAc. Organic layer was washed with water (20 mL), sat sodium bicarbonate (2×30 mL), 5% sodium carbonate (30 mL), water (20 mL), and brine (20 mL). Organic solution was dried over sodium sulfate and concentrated to a yellow color solid residue. The residue was purified via column chromatography (silica gel, 3% methanol in DCM) to afford 279 mg (0.48 mmol, 83%) of 1 and its R_P-isomer.

Example 12-7

Synthesis of (S)-2-[(R)-[(1R,4R,5R)-5-(2-Amino-6-methoxy-purin-9-yl)-4-(R)-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid isopropyl ester (R_P-isomer of 1) via (2S)-isopropyl 2-(((R)-2-chloro-4-nitrophenoxy)(phenoxy)phosphoryl)amino)propanoate ((S, R_P)-12)

[0170]



[0171] To a 50 mL of dry round-bottomed flask were charged 70 mg (0.223 mmol) of 2 and 1 mL of anhydrous THF. The flask was cooled in an ice-water bath and Grignard reagent (1 M solution in THF, 0.32 mL) was added dropwise at 0° C. After stirred at 0° C. for half hour, a solution of the (S, R_P)-12 (129 mg, 0.29 mmol) in 1 mL of THF was added via a syringe. A clear brown color solution was formed and gradually warmed up to ambient temperature. After overnight (19 h) at room temperature, the reaction was quenched by adding 1 N of diluted HCl at 0° C. The mixture was diluted with ethyl acetate (20 mL) and water (10 mL). After separa-

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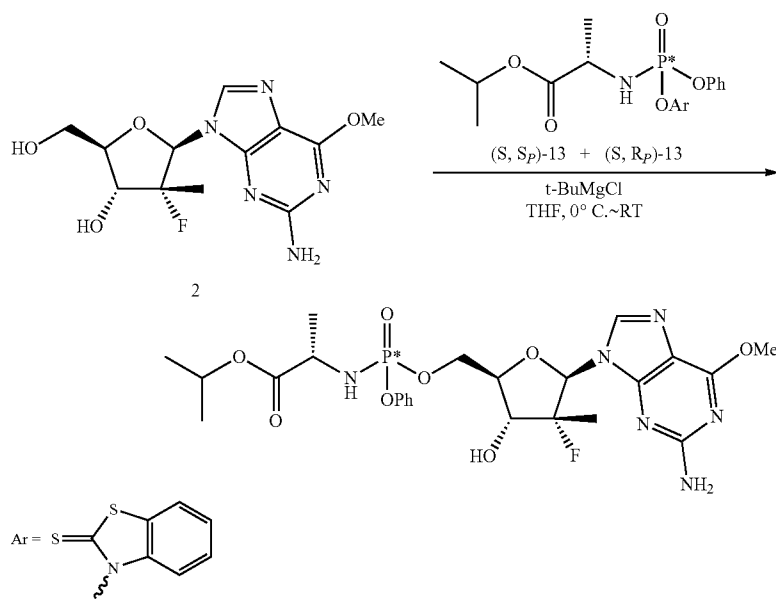
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tion of two layers, aqueous layer was extracted with EtOAc (10 mL). Organic layer was washed with water (10 mL), sat sodium bicarbonate (3×15 mL), water (10 mL), brine (10 mL), and dried over sodium sulfate. After concentration, the solid residue was purified via column chromatography (silica gel, 3% methanol in DCM) to afford 100 mg (0.17 mmol, 77%) of product as a white solid and single isomer.

Example 12-8

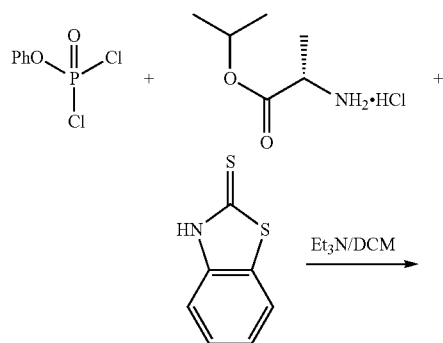
Synthesis of (S)-2-[[[(1R,4R,5R)-5-(2-Amino-6-methoxy-purin-9-yl)-4-(R)-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino]-propionic acid isopropyl ester diastereomeric mixture (1+R_P-isomer of 1) via diastereomeric mixture (2S)-isopropyl 2-((phenoxy(2-thioxobenzo[d]thiazol-3(2H)-yl)phosphoryl)amino)propanoate ((S, S_P)-13 and (S, R_P)-13)

[0172]

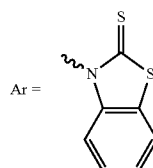
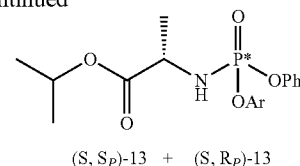


a) Preparation of ((S, S_P)-13 and (S, R_P)-13)

[0173]



-continued



[0174] Phenyl phosphorodichloridate (6.37 g, 30.19 mmol) was dissolved in 40 mL of dry DCM and subsequently cooled to 0° C. After addition of solid (L)-alanine isopropyl ester

(5.06 g, 30.19 mmol), the reaction mixture was cooled to -78° C. and then treated with triethylamine (8.84 mL, 63.3 mmol) dissolved in 20 mL of dry DCM. The resulting mixture was stirred for 30 min at this temperature before being allowed to warm to 0° C. Subsequently, a preformed solution of benzo[d]thiazole-2(3H)-thione (5.05 g, 30.19 mmol) and triethylamine (4.63 mL, 33.21 mmol) dissolved in 20 mL of dry DCM was added over 5-10 min whereupon the mixture was allowed to warm to RT over night. The cloudy mixture was then cooled back to 0° C. and filtered to remove all solids. The filtrate was stripped of all solvent under reduced pressure. The resulting residue was suspended in 50 mL of TBME and stirred for 1 h at RT. Subsequent filtration removed more triethylamine hydrochloride and yielded a filtrate that was again stripped of its solvent under reduce pressure. Column chromatography (DCM) yielded ((S, S_P)-13 and (S, R_P)-13) (3:1, isomer I/isomer II) in 15% (1.97 g) yield as viscous oil. [0175] Data: ¹H NMR (CDCl₃, 300 MHz) δ: 8.63-8.59 (m, 1H), 7.37-7.27 (m, 7H), 7.18-7.14 (m, 1H), 6.05-5.97 (m,

1H), 5.04 (hepta, 1H, isomer II), 4.91 (hepta, 1H, isomer I), 4.37-4.24 (m, 1H), 1.45-1.42 (d, 3H, isomer I), 1.41-1.39 (d, 3H, isomer II), 1.26-1.22 (m, 6H), 1.09-1.02 (m, 6H). ^{31}P NMR (CDCl_3 , 121 MHz) δ : -0.43 (isomer I), -1.29 (isomer II).

b) Preparation of Diastereomeric Mixture of 1 and its R_P -isomer from ((S, S_P)-13 and (S, R_P)-13) and 2

[0176] To a dry round-bottomed flask were added 120 mg (0.38 mmol) of 2 and 1.5 mL of anhydrous THF. The mixture was cooled to 0° C. and 0.5 mL of Grignard reagent (0.5 mmol) was added dropwise. The clear solution was stirred at 0° C. for half hour. A solution of ((S, S_P)-13 and (S, R_P)-13) (197 mg, 0.45 mmol) in 1.5 mL of THF was added via a syringe. The resulting mixture was allowed to warm up to room temperature and stirred for overnight (19 h). TLC showed reaction was not complete and product was found together with bis-phosphoramidate by-product. Reaction was quenched by addition of 1 N of diluted HCl at 0° C. and mixture was diluted with 20 mL of ethyl acetate. After work-up, as noted above, an oily residue was obtained and it was purified via column chromatography (silica gel, 3% methanol in DCM) to afford 78.6 mg (0.13 mmol, 35%) of 1 and its R_P -isomer as a mixture of two diastereomers. From column, 36.4 mg of bis-phosphoramidate by-product was isolated.

Example 13

Biological Data of Compound 1

[0177] HCV replicon assay. HCV replicon RNA-containing Huh7 cells (clone A cells; Apath, LLC, St. Louis, Mo.) were kept at exponential growth in Dulbecco's modified Eagle's medium (high glucose) containing 10% fetal bovine serum, 4 mM L-glutamine and 1 mM sodium pyruvate, 1× nonessential amino acids, and G418 (1,000 $\mu\text{g}/\text{mL}$). Antiviral assays were performed in the same medium without G418. Cells were seeded in a 96-well plate at 1,500 cells per well, and test compounds were added immediately after seeding. Incubation time 4 days. At the end of the incubation step, total cellular RNA was isolated (RNeasy 96 kit; Qiagen). Replicon RNA and an internal control (TaqMan rRNA control reagents; Applied Biosystems) were amplified in a single-step multiplex RT-PCR protocol as recommended by the manufacturer. The HCV primers and probe were designed with Primer Express software (Applied Biosystems) and covered highly conserved 5'-untranslated region (UTR) sequences (sense, 5'-AGCCATGGCGTTAGTA(T)GAGTGT-3', and antisense, 5'-TTCCGCAGACCAC-TATGG-3'; probe, 5'-FAM-CCTCCAGGAC-CCCCCTCCC-TAMRA-3').

[0178] To express the antiviral effectiveness of a compound, the threshold RT-PCR cycle of the test compound was subtracted from the average threshold RT-PCR cycle of the no-drug control ($\Delta\text{Ct}_{\text{HCV}}$). A ΔCt of 3.3 equals a 1-log 10 reduction (equal to the 90% effective concentration [EC_{90}]) in replicon RNA levels. The cytotoxicity of the test compound could also be expressed by calculating the $\Delta\text{Ct}_{\text{rRNA}}$ values. The $\Delta\Delta\text{Ct}$ specificity parameter could then be introduced ($\Delta\text{Ct}_{\text{HCV}} - \Delta\text{Ct}_{\text{rRNA}}$), in which the levels of HCV RNA are normalized for the rRNA levels and calibrated against the no-drug control. Compound 1 was tested for its biological properties based on the preceding assay. The results of these tests are disclosed below.

Compd. No.	CloneA EC_{90} (μM)
1	0.02

[0179] Replicon assay results show activity when compound 1 is assayed in combination with compound C (designated as compound 19 in US 2010/0081628, and the individual diastereomers (19a and 19b) disclosed in the same application); compound D (disclosed in US 2010/0016251); compound E (disclosed in U.S. Ser. No. 12/783,680 as Sp-4); ITMN-191 (disclosed in US 2009/0269305 at Example 62-1); and ANA-598 (disclosed in F. Ruebasam et al. Biorg. Med. Chem. Lett. (2008) 18: 3616-3621 as compound 31). Surprisingly, replicon assay results show synergism when compound 1 is assayed in combination with any one of compound C (19a or 19b); compound E (S_P -4), ITMN-191; and ANA-598.

[0180] This application claims priority to U.S. 61/319,513, filed on Mar. 31, 2010; U.S. 61/319,548, filed on Mar. 31, 2010; and U.S. 61/355,940, filed on Jun. 17, 2010, the subject matter of which is incorporated by reference in its entirety.

[0181] The contents of U.S. Provisional Patent Application No. 61/319,548 and U.S. patent application Ser. Nos. 12/645,765, filed on Dec. 23, 2009; 12/053,015, filed on Mar. 21, 2008; 12/783,680, filed on May 20, 2010; and 13/076,552, filed on Mar. 31, 2011 are hereby incorporated by reference in their entirety. Moreover, the patent and non-patent references disclosed herein are incorporated by reference. In the event that the incorporated subject matter contains a term that conflicts with a term disclosed in the present application text, the meaning of the term contained in the present application controls provided that the overall meaning of the incorporated subject matter is not lost.

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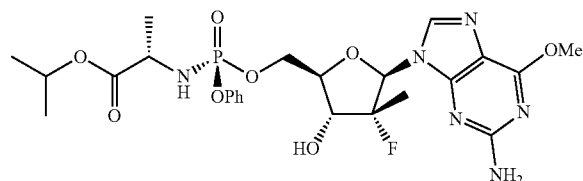
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20

1. A compound represented by formula 1 its hydrate or solvate thereof in crystalline or crystal-like form.



2. The compound hydrate of claim 1, represented by 1.mH₂O, wherein m varies in an integer or non-integer amount from about 0 to about 5.

3. The compound of claim 2, wherein m is about 1.

4. The compound of claim 2, wherein m is about 1/2.

5. The compound of claim 2, wherein m is about 1, that further comprises an amount of adsorbed water.

6. The compound of claim 5, wherein the amount of adsorbed water ranges from about 0 wt. % to about 10 wt. % based on the weight of the 1.H₂O.

7. Crystalline 1.mH₂O.

8. Crystalline 1.mH₂O of claim 7 having an XRPD 2θ-reflection (°) at about: 14.8.

9. Crystalline 1.mH₂O of claim 7 having XRPD 2θ-reflections (°) at about: 14.8 and 17.6.

10. Crystalline 1.mH₂O of claim 7 having XRPD 2θ-reflections (°) at about: 14.8, 17.6, and 20.4.

11. Crystalline 1.mH₂O of claim 7 having XRPD 2θ-reflections (°) at about: 8.7, 14.8, 17.6, and 20.4.

12. Crystalline 1.mH₂O of claim 7 having XRPD 2θ-reflections (°) at about: 8.7, 13.6, 14.8, 17.6, and 20.4.

13. Crystalline 1.mH₂O of claim 7 having XRPD 2θ-reflections (°) at about: 8.7, 11.1, 13.6, 14.8, 17.6, and 20.4.

14. Crystalline 1.mH₂O of claim 7 having an XRPD diffraction pattern substantially as that shown in FIG. 1.

15. Orthorhombic crystalline 1.mH₂O.

16. Orthorhombic crystalline 1.mH₂O of claim 15, having the following unit cell parameters a~10.99 Å, b~13.09 Å, and c~20.36 Å.

17. A composition comprising the compound of claim 1.

18. A method for treating HCV in a patient in need thereof, which comprises administering to the patient a therapeutically effective amount of the compound of claim 1.

19. A method for treating HCV in a patient in need thereof, which comprises administering to the patient a therapeutically effective amount of the compound of claim 1.

20. A method for treating HCV in a patient in need thereof, which comprises administering to the patient a therapeutically effective amount of the compound of claim 1 in combination or in alternation with a therapeutically effective amount of another antiviral.

21. The method of claim 20, wherein the other antiviral is selected from among compound A; compound B; compound C; compound D; compound E; telaprevir; boceprevir; BMS-790052; ITMN-191; ANA-598; TMC435; and combinations thereof.

22. A co-therapy method for treating HCV in a patient in need thereof, which comprises administering to the patient a therapeutically effective amount of the compound of claim 1 in combination or in alternation with a therapeutically effective amount of another antiviral.

23. The co-therapy method of claim 22, wherein the other antiviral is selected from among compound A; compound B; compound C; compound D; compound E; telaprevir; boceprevir; BMS-790052; ITMN-191; ANA-598; TMC435; and combinations thereof.

24. A process for preparing the compound of claim 1, which comprises crystallizing 1.

25. The process of claim **24**, which further comprises dissolving or suspending **1** in a solvent or solvent mixture.

26. The process of claim **25**, wherein the solvent or solvent mixture is selected from among anisole, ethyl acetate; xylenes; toluene; isopropanol; acetone; dichloromethane; diethyl ether; isopropyl acetate; t-butyl methyl ether; and combinations thereof.

27. The process of claim **25**, wherein the solvent mixture is selected from among anisole/ethyl acetate; heptanes/ethyl acetate; xylenes/ethyl acetate/water; anisole/water; ethyl

acetate/xylenes; isopropanol/xylenes; acetone/xylenes; dichloromethane/xylenes; dichloromethane/hexanes; ethyl acetate/toluene; diethyl ether/xylenes; isopropyl acetate/xylenes; isopropyl acetate/heptanes; ethyl acetate/water; t-butyl methyl ether/water; t-butyl methyl ether/ethyl ether; and t-butyl methyl ether.

28. A method for determining the crystallinity of the compound of claim **1**, which comprises analyzing the compound by XRPD or single-crystal X-ray crystallography.

* * * * *



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The Use of Bioisosterism in Drug Design and Molecular Modification

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ABSTRACT

Bioisosteres are atoms or group of molecules that fit the broadest definition for isosteres. They have chemical and physical similarities thus producing broadly similar biological properties. Many heterocycles, when appropriately substituted exhibits bioisosterism. Bioisosterism represents an approach used by the medicinal chemist for the rational modification of lead compounds into safer and more clinically effective agents. It has significant value in drug design and lead optimization process as it may enhance the desired biological or physical properties of a compound, reduce toxicity and also alter the metabolism of the lead. Bioisosteric replacement is not simple replacement with another isostere but they are firstly analyzed by structural, solubility and electronic parameters to obtain molecules having similar biological activity. Few of the popular examples of the successful use of bioisosteres have been included. The objective of this review is to provide an overview of bioisosteric replacements which can be used for advance drug development.

Keywords: Bioisostere, Isostere, Drug design, Replacement, Pseudoatoms

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INTRODUCTION

Bioisosteres are substituents or groups with similar physical or chemical properties which produce broadly similar biological properties to a chemical compound. In a biologically active molecule the replacement of an atom or a group of atoms by another one presenting the same physicochemical properties is based on the concept of isosterism. The notion of isosterism was introduced in 1919 by Langmuir. The extensive application of isosterism to modify a part of a biologically active molecule to get another one of similar activity, has given rise to the term of bio-isosterism.

In drug design, the purpose of exchanging one bioisostere for another is :

1. To enhance the desired biological or physical properties of a compound without making significant changes in chemical structure.
2. To attenuate toxicity.
3. To modify the activity of the lead compound.
4. To alter the metabolism of the lead.

Depending on the molecule used in the substitution, little change in activity (i.e. either increase or decrease in affinity or efficacy) can occur as it is dependent on factors such as electronegativity, size, pka, solubility which are important for target binding such as electronegativity, size, pka, solubility etc. ¹.

1.1. History: Development of the isosterism concept

1. a. Molecular number ²

Allen, in 1918 defined the molecular number of a compound in a similar way of the atomic number:

$$N = aN_1 + bN_2 + cN_3 + \dots + zN_i$$

Where N = Molecular number

$N_1, N_2, N_3 \dots N_i$ = Respective atomic numbers of each element of the molecule.

$a, b, \dots z$ = Number of atoms of each element present in the molecule.

Example: Comparison of the ammonium and the sodium cations. The atomic number of nitrogen is 7 and that of hydrogen is 1. Thus the molecular number of the ammonium cation can be calculated and compared to that of the sodium ion (**Table 1**).

Possessing the same molecular number, the ammonium cation should resemble the sodium cation. Two compounds with identical molecular numbers present some similar physical properties (e.g. specific heat).

Table 1: Molecular number of ammonium and sodium cations

	Atomic number	Molecular number
NH_4^+	$7 + (4 \times 1)$	11
Na^+	11	11

1.2. Isosterism concept³

Langmuir in 1919 defined the concept of isosterism as follows:

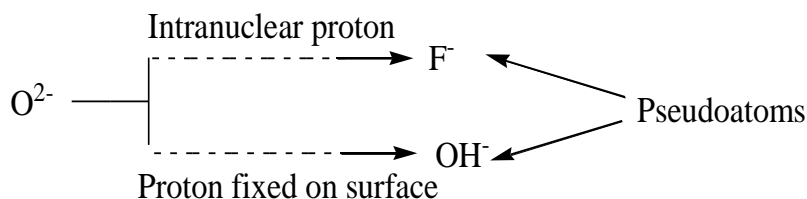
Comolecules are isosteric if they contain the same number and arrangement of electrons. The comolecules of isosteres must, therefore contain the same number of atoms. The essential differences between isosteres are confined to the charges on the nuclei of the constituent atoms (Table 2).

Table 2: Groups of isosteres⁴

Groups	Isosteres
1	H, He, Li^+
2	O^{2-} , F^- , Ne, Na^+ , Mg^{2+} , Al^{3+}
3	S^{2-} , Cl^- , Ar, K^+ , Ca^{2+}
4	Cu^{2-} , Zn^{2+}
...	...
...	...
8	N_2 , CO, CN^-
9	CH_4 , NH_4^+
10	CO_2 , N_2O , N_3 , CNO^-
...	...
...	...
20	MnO_4^- , CrO_4^{2-}
21	SeO_4^{2-} , AsO_4^{3-}

1.3. Grimm's hydride displacement law⁵

Later on, in 1925, Grimm formulated the "Hydride displacement law" according to which the addition of hydrogen to an atom confers on an aggregate the properties of the atom of next highest atomic number. An isoelectronic relationship exists among such aggregates which were named pseudoatoms. Example, when a proton is "added" to the O^{2-} ion in the nuclear sense, an isotope of fluorine is obtained.

**Figure 1: Pseudoatoms.**

Here the hydrogen ion has been penetrated into the electronic shell of the oxygen, which is assumed to be masked by the greater atom and exerts only negligible effect toward the outside.

The fluoride anion F^- and the hydroxyl anion OH^- show therefore some analogies. The generalization of the pseudoatom concept represents the “Hydride displacement law” (**Table 3**).

Table 3: Hydride Displacement law ⁶

Number of electrons					
6	7	8	9	10	11
$\begin{array}{c} \\ -C- \\ \end{array}$	$\begin{array}{c} \\ -N- \\ \end{array}$	-O-	-F	Ne	Na^+
	$\begin{array}{c} \\ -CH \\ \end{array}$	-NH-	-OH	FH	
		-CH ₂ -	-NH ₂	OH ₂	
			-CH ₃	NH ₃	OH^{3+}
				CH ₄	NH^{4+}

1.4. Erlenmeyer's expansion of the isosterism concept

⁷

Erlenmeyer proposed his own definition of isosteres as elements, molecules or ions, in which the peripheral layers of electrons may be considered identical.

Erlenmeyer also proposed three expansions of the isosterism concept:

1. To the whole group of elements present in a given column of the periodic table. Thus silicon becomes isosteric to carbon, sulfur to oxygen, etc.
2. To the pseudoatoms, with the aim of including groups which at a first glance seem totally different, but which in practice, possess rather similar properties. This is the case for the pseudohalogens (e.g. Cl, CN, SCN, etc.)
3. To the ring equivalents: the equivalence between $-CH=CH-$ and $-S-$ explains the well known analogy between benzene and thiophene.

$$M_{(-CH=CH-)} = 26$$



$$M_{(S)} = 32$$



Figure 2: Analogy between benzene and thiophene.

1.5. Friedman's and Thornber's definitions

Friedman proposed to call bioisosteres compounds: “which fit the broadest definition of isosteres and have the same type of biological activity” ⁸. Friedman considered that, isosteres that exhibit opposite properties (antagonists) have also to be considered as bioisosteres, as they interact with the same recognition site. eg. Para-amino benzoic acid and Para-amino benzene-sulfonamide ^{9, 10}. Thornber proposed a loose and flexible definition of the term bioisostere as: “Bioisosteres are groups or molecules which have chemical and physical similarities producing broadly similar biological effects” ¹¹.

2. CLASSIFICATION OF BIOISOSTERISM^{4,12}

In 1970, Burger classified and subdivided bioisosteres into two broad categories according to the degree of electronic and steric factors.

2.1. Classic isosteres:

2.1. A. Monovalent atoms or groups

2.1. B. Divalent atoms or groups

2.1. C. Trivalent atoms or groups

2.1. D. Tetravalent atoms

2.1. E. Ring equivalents

2.2. Non-classical isosteres:

2.2. A. Cyclic vs. Non cyclic

2.2. B. Non classic bioisosterism of functional groups

2.2. B.1. Carboxylic group bioisosteres

2.2. B.2 Hydroxyl group bioisosteres

2.2. B.3. Amide group bioisosteres

2.2. B.4. Halogen bioisosteres

2.1. Classic isosteres

They obey steric and electronic definition (Table 4).

Table 4: Classic bioisostere atoms and groups

Monovalent	Divalent	Trivalent	Tetravalent
-OH, -NH ₂ , -CH ₃ , -OR	-CH ₂ -	=CH-	=C=
-F, -Cl, -Br, -I, -SH, -PH ₂	-O-	=N-	=Si=
-Si ₃ , -SR	-S-	=P-	=N ⁺ =
	-Se-	=As-	=P ⁺ =
	-Te-	=Sb-	=As=
			=Sb ⁺ =

2.1.A. Monovalent atoms or groups

❖ Hydrogen vs. Fluorine Replacement¹³

Steric parameters for hydrogen and fluorine are similar, their Vander Waal's radii being 1.2 and 1.35 Å respectively. Thus, the difference in the electronic effects (fluorine being the most electronegative element in the periodic table) is often the basis for the major differences in the pharmacological properties of agents. Due to its electronegativity, fluorine exerts strong field and inductive effects on the adjacent carbon atom. However, fluorine can donate a lone pair of electrons by resonance. This is commonly referred to as its mesomeric effect. The opposing resonance and field effects can nearly cancel. The pharmacological differences can be attributed

to the influence of the electron withdrawing effect that the fluorine substitution causes on interaction with a biological receptor or enzyme, as well as its effect on the metabolic fate of the drug.

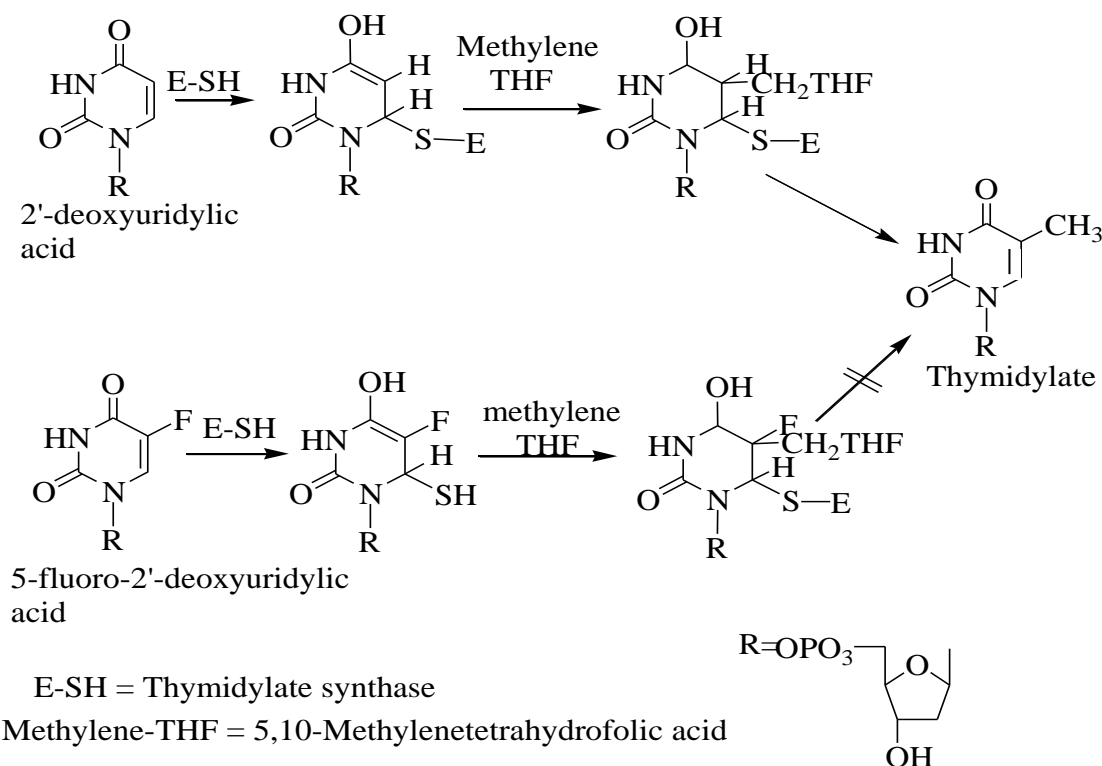


Figure 3: Hydrogen to fluorine replacement.

eg. 1 The biochemically altered form of 5-FU ie. 5-Fluoro-2-deoxyuridylic acid is ultimately responsible for the inhibition of thymidylate synthase, an enzyme involved in the conversion of uridylic acid to thymidylic acid and critical for DNA synthesis. The increased reactivity of 5-Fluoro-2-deoxyuridylic acid relative to 2'-deoxyuridylic acid is due to the inductive effect of fluorine which results in its covalent binding to thymidylate synthase.

❖ Interchange of Hydroxyl and Amino Groups

The best known example of classical isosteric substitution of an amino group for a hydroxyl group is illustrated by aminopterin (b) wherein the hydroxyl substituent of folic acid (a) has been substituted by an amino group. This represents a monovalent bioisosteric substitution at a carbon atom adjacent to a heterocyclic nitrogen atom. This bioisosteric replacement has the capability of mimicking even the tautomeric forms of folic acid.

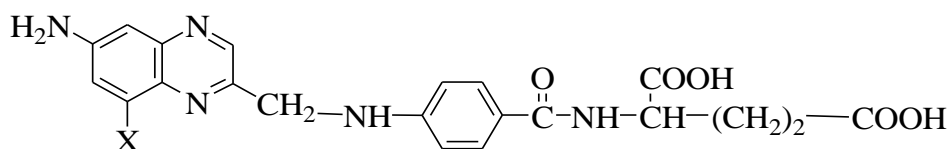


Figure 4: (a) X= OH folic acid, (b) X=NH₂ Aminopterin.

In the presence of electron donating atoms such as nitrogen in heterocyclic systems, it is known that there will be tautomerization where a neighboring C-OH will tautomerize to C=O¹⁴. In the case of a neighboring carbon containing C-NH₂ the preferred tautomer is the C-NH form^{14, 15}.

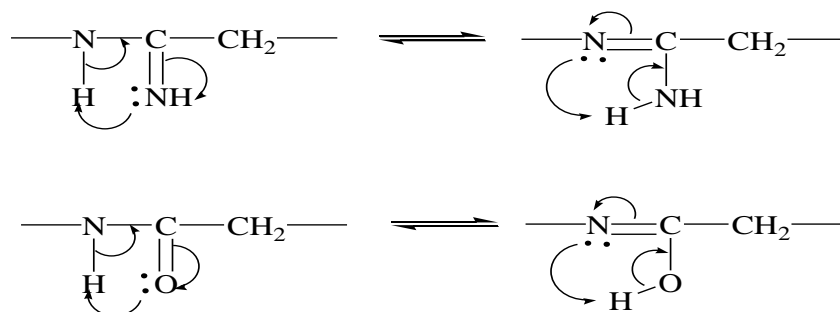


Figure 5: Tautomerization of cyclic nitrogen.

The similarities as well as the capability of the amino group to hydrogen bond and to the enzyme are two important factors that facilitate the binding of aminopterin to the enzyme dihydrofolate reductase¹⁶.

❖ Interchange of Hydroxyl and Thiol Groups

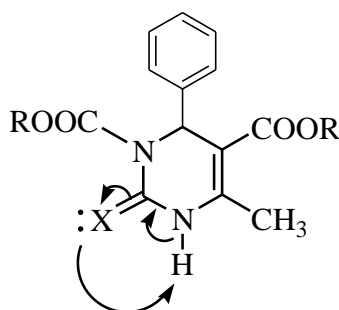


Figure 6: 1, 4-dihydropyrimidine.

In order to enhance the calcium channel blocking capacity of certain dihydropyrimidine agents, a number of isosteric analogues with the general structure were synthesized¹⁷. Substitution of the hydroxyl with an amino resulted in analogues with similar potency. However, substitution with the thiol resulted in enhanced potency. This is due to the size of the substituent, described here as the Vander Waal's radii and the hydrogen bonding ability. Therefore, replacement with the amino group, which has a similar size, resulted in similar potency and replacement with the sterically optimal thiol resulted in an analogue which was more potent (Table 5)¹⁸.

Table 5: Calcium channel blocking activity of 1, 4-Dihydropyrimidines

Compound	X	Vander Waal's radius (Å°)	IC ₅₀ (nM)
15a	=O	1.40	140
15b	=NH	1.50	160
15c	=S	1.85	17

❖ Replacement of chlorine with methyl¹⁹

The chlorine atom is often viewed to be isosteric and isolipophilic with the methyl group it is very often selected as a bioisosteric replacement because of its ability to alter the metabolism. Replacement of a chloro atom with a methyl substituent can facilitate metabolism of a xenobiotic. Lipid-soluble chemicals tend to be distributed into adipose tissue where, unless they are metabolized, they tend to accumulate for long periods of time, e.g. DDT. The replacement of the trichloromethyl moiety with a tert-butyl group results in diminished persistence of this pesticide. The methyl substituent provide a site which is susceptible to metabolic degradation.

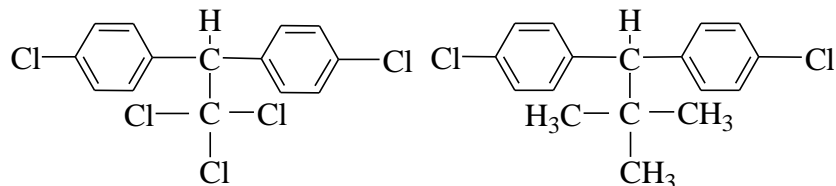


Figure 7: Replacement of chlorine with methyl in DDT.

2.3. B. Divalent atoms and groups

➤ Divalent replacements involving double bonds²⁰

This subclass includes replacements of groups such as C=S, C=O, C=NH and C=C.

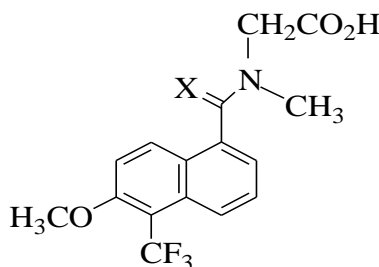


Figure 8: (a) X= S Tolrestat, (b) X= O Oxotolrestat.

The replacement of C=S with C=O in Tolrestat (a) an aldose reductase inhibitor, currently under study in human subjects for the treatment of diabetic neuropathy, resulted in oxo-Tolrestat (b) which retained activity both *in vitro* and *in vivo* (Table 6).

Table 6: Aldose Reductase inhibitory activity of Tolrestat and Oxo-Tolrestat

Compound	X	Aldose reductase inhibition	
		<i>In vitro</i>	<i>In vivo</i>
A	S	94	53
B	O	86	56

➤ Divalent Replacements Involving Two Single Bonds²¹

The second major class of divalent bioisosteres represents those atoms or groups which are attached to different substituent. The bond angle or the conformation associated with the use of

these divalent bioisosteres may be an important factor associated with retention of biological activity (Table 7).

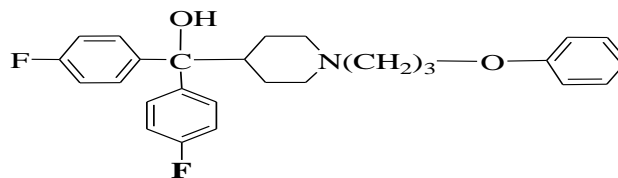


Figure 9: Divalent replacement involving two single bonds.

Table 7: Oral antiallegry activity in the passive foot anaphylaxis assay of analogues containing varied heteroatoms

Compound	X	Electronegativity	Bond angle(deg)	Passive foot anaphylaxis assay (10mg/kg)
a	-O-	3.51	108.0	+++
b	-S-	2.32	112.0	+
c	-CH ₂ -	2.27	111.5	+
d	-NH-	2.61	111.0	+

2.1. C. Trivalent atoms and groups

A classical trivalent bioisosteric replacement is $-\text{CH}=\text{}$ with $-\text{N}=\text{}$

- I. This replacement when applied to cholesterol resulted in 20, 25-diazacholesterol which is a potent inhibitor of cholesterol biosynthesis. The greater electronegativity of the nitrogen atom could be responsible for the biological activity of this bioisostere²².

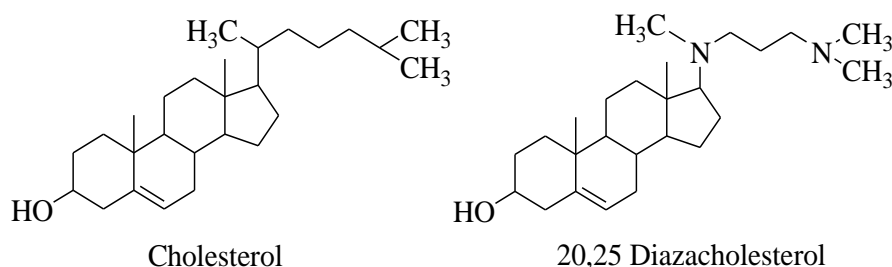


Figure 10: Trivalent bioisosteric replacement.

- II. The 4-dimethylamino-antipyrene and its carba-isostere are about equally active as antipyretics²³.

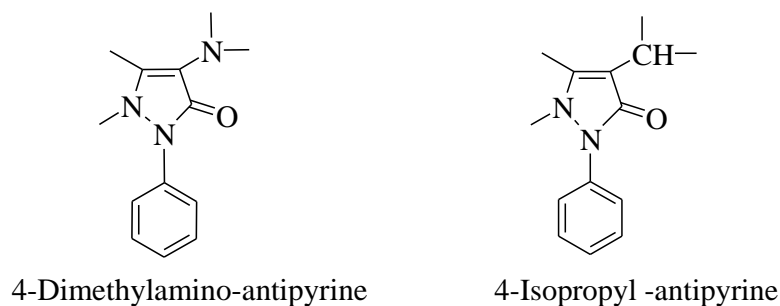


Figure 11: Antipyretics.

2.1. D. Tetra substituted Atoms²⁴

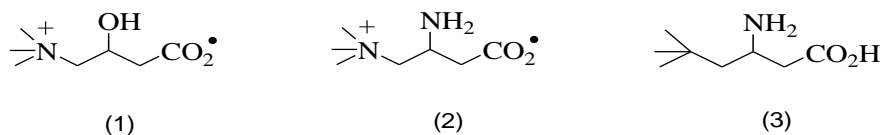


Figure 12: Replacement of tetravalent trimethyl ammonium group with tert-butyl group.

Certain simple acyl carnitine analogues are potent carnitine acyltransferase (CAT) inhibitors (Table 8). Structure-activity studies in this series have included the bioisosteric replacement of the hydroxyl group of carnitine **Figure 12** (1) with an amino **Figure 12** (2) and replacement of the tetravalent trimethylammonium group with a tertiary butyl group **Figure 12** (3).

Table 8: Rate constants for carnitine and synthetic analogues with Pigeon breast carnitine acyltransferase

X	Ki (mM)
1	4.0
2	2.6

2.1. E. Ring equivalents

1. In sulphonamide antibacterials phenyl group may be replaced by heterocyclic group to give active compound eg. sulfadiazine and sulfamethoxazole etc. In this case no essential activity difference is found between the original drug and its isostere²⁵.



Figure 13: Sulfonamide antibacterial.

2. In class of arylthiazine-1, 1-dioxides the newest member was found where the benzothiazinic nucleus was replaced by the thienothiazinic moiety. This example represents the bioisosteric relationship existing between aromatic heterocyclic rings and the phenyl group. The profile of pharmacotherapeutic activity proved to be comparable because of its long plasmatic half-life, a desirable quality for cases of arthritis as well as osteoarthritis. Both derivatives act by the same mechanism of action, at the same receptor level, i.e. cyclooxygenase, an enzyme involved in arachidonic acid metabolism²⁶.



Figure 14: Ring equivalents.

(i) Bioisosteres of pyridine ²⁷⁻²⁹

The pyridine ring of nicotine can be replaced by different other rings like methyl-isoxazole or methylisothiazole. The bioisosteric replacement of the isoxazole ring in the (3-methyl-5-isoxazolyl) methylene-azacyclic compound with pyridine, pyrazine, oxadiazole or an acyl group resulted in ligands with moderate to high affinity for the central nicotinic cholinergic receptors.

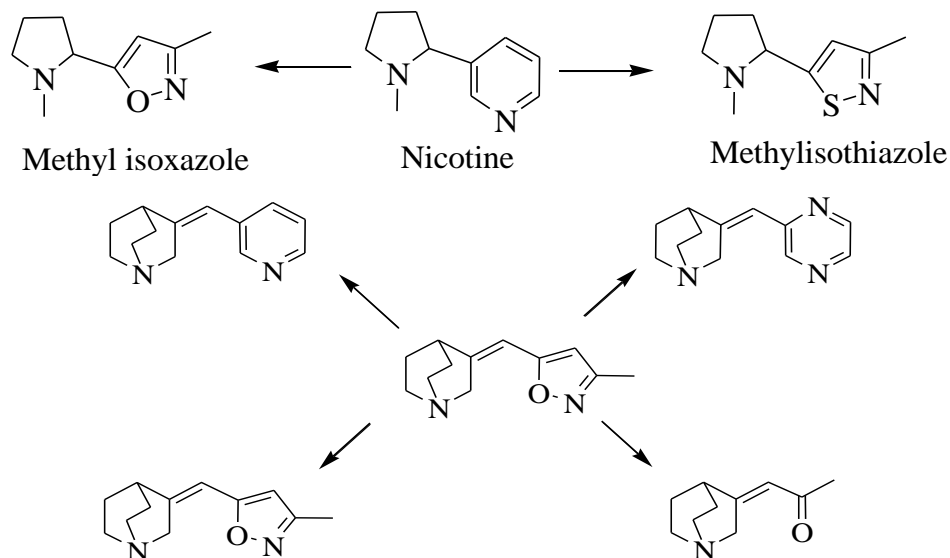


Figure 15: Non classical bioisosteres of pyridine ring.

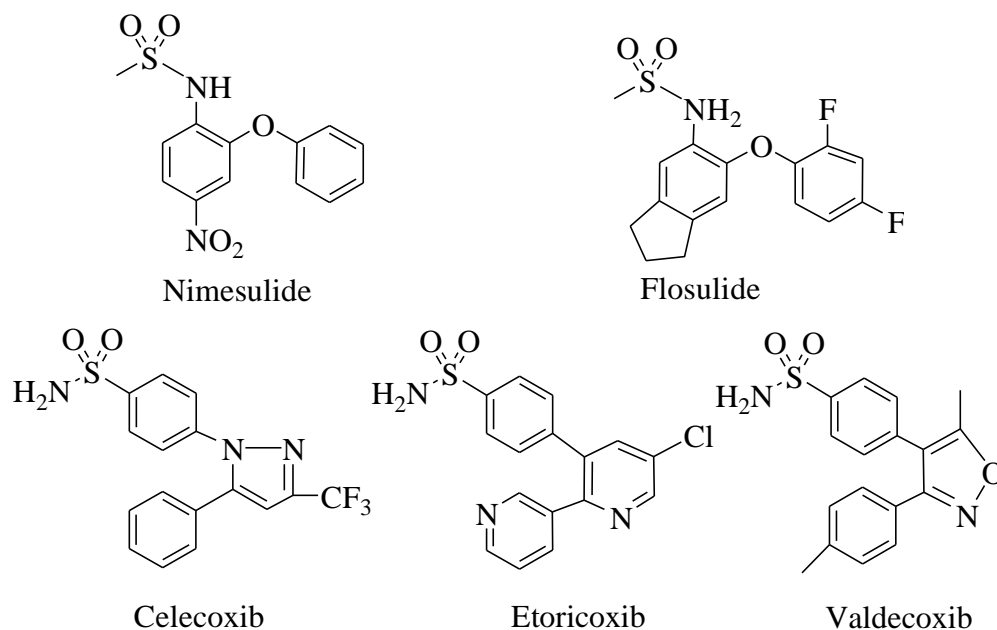















(ii) Bioisosteres of other heterocycles ²⁶

Figure 16: COX-2 inhibitors.

Selective cyclooxygenase-2 inhibitors (COX-2 inhibitors) is a nice example of bioisosters of heterocycles. The comparison of the most potent selective COX-2 inhibitors suggests that

isoxazoles, pyridines and pyrazoles are good bioisosteres of each other as well as nitrophenol and indanones (**Table 9**).

Table 9: Ring replacements

	 ,  ,  , 
	 ,  , 
	 , 
	 , 

2.2. Non classic bioisosteres ⁴

- They do not obey the steric and electronic definition of classical isosteres.
- They do not have the same number of atoms as the substituent or moiety for which they are used as a replacement (**Table 10**).

Table 10: Non-Classic bioisosteres

-CO-	-COOH	-SO ₂ NH ₂	-H	-	-COOR	-CONH ₂
-CO ₂ -	-SO ₃ H	-	-F	CONH-	-ROCO-	-CSNH ₂
-SO ₂ -	-tetrazole	PO(OH)NH ₂		-		
-SO ₂ NR-	-SO ₂ NHR		-OH	NHCO-	-catechol	
			-CH ₂ OH			
-CON-	-SO ₂ NH ₂				-benzimidazole	
-CH(CN)-	-3-hydroxy isoxazole		-NHCONH ₂			C ₄ H ₄ S
R-S-R	-2-hydroxy chromones		-NH-CS-NH ₂			-C ₅ H ₄ N
(R-O-R')						-C ₆ H ₅
R-N(CN)-	=N-		-NH-C(=CHNO ₂)-			-
			NH ₂ -NH-			C ₄ H ₄ NH
			C(=CHCN)-NH ₂			
-halide	C(CN)=R'					
	-CF ₃					
	-CN					
	-N(CN) ₂					
	-C(CN) ₃					

2.4.A. Cyclic vs. Non cyclic³⁰

The molecular design of Hexestrol was carried out from the opening of rings B and C of the steroidal skeleton of estradiol. However, in analogy to what was observed for estradiol, the activity of Hexestrol is dependent on the configurational aspects, such that, the diastereoisomer *E* presents an estrogen profile significantly superior to the diastereoisomer *Z*, with reduced estrogen activity also being observed for the dihydrogenated compound that is compound Diethylstilbestrol.

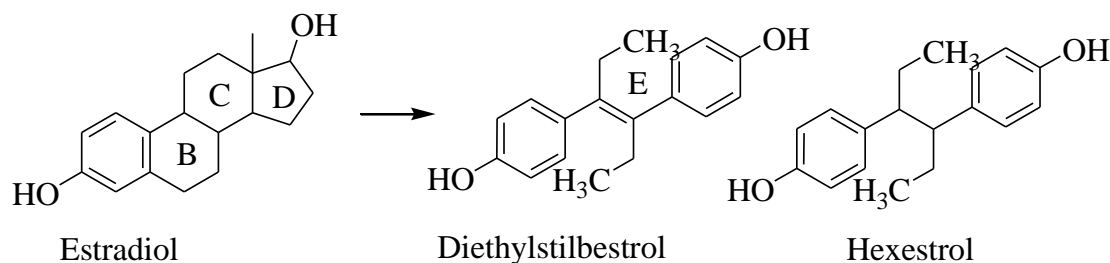


Figure 17: Ring opening bioisosterism.

2.2. B. Non classic bioisosterism of functional groups

2.2. B.1. Carboxylic group bioisosteres³¹

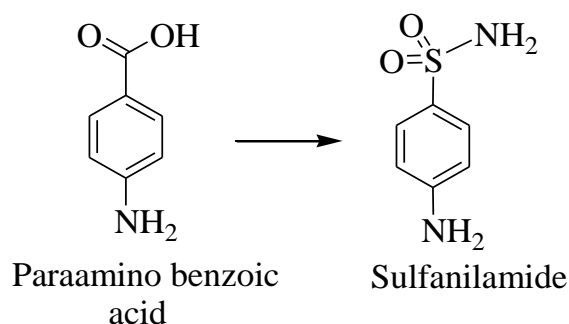


Figure 18: Carboxylic group bioisosteres.

Table 11: Carboxylic group bioisosteres

-COOH	-SO₂NH₂
	-CONHOH
	-SO₂OH
	-CONHCN

Evidence of the similarity between sulfanilamide and paraminobenzoic acid was found during elucidation of its mechanism of molecular action. This similarity was based on electronic and conformational aspects as well as the physicochemical properties such as pKa and log P. This denotes an authentic bioisosteric relationship between the sulfonamide (SO₂NH₂) and carboxylic acid functionalities (CO₂H) (**Table 11**).

2.2. B.2. Hydroxyl group bioisosteres

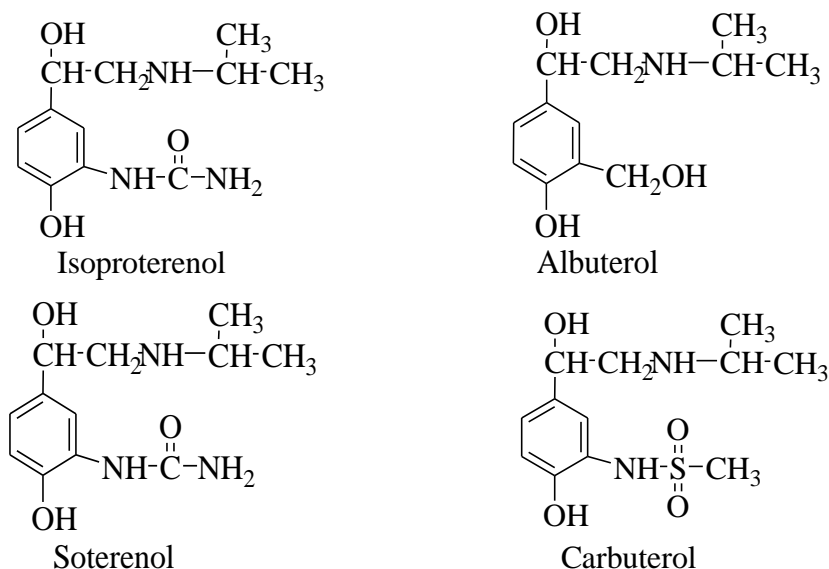


Figure 19: β -adrenoceptor agonists.

Notable example of β -adrenoceptor agonists that is isoproterenol, is widely used clinically as a bronchodilator; in which a 3-hydroxyl group has been replaced with bioisosteric groups which include albuterol 3-CH₂OH, soterenol 3-NHSO₂CH₃³² and carbuterol 3-NHCONH₂³³. This results in agents with potent and selective activities (**Table 12**).

Table 12: Hydroxyl group bioisosteres

-OH	-CH ₂ OH
	-NHCONH ₂
	-NHCOCH ₃
	-NHSO ₂ CH ₃
	-NHCN

2.2. B.3. Amide group bioisosteres³⁴

Bioisosteric replacements for the amide are done because of its implications in peptide chemistry and the development of peptide mimetic. Peptide bonds and peptide fragments have been replaced with a wide variety of structural moieties in attempts to convert peptides into chemically stable and orally available molecules.

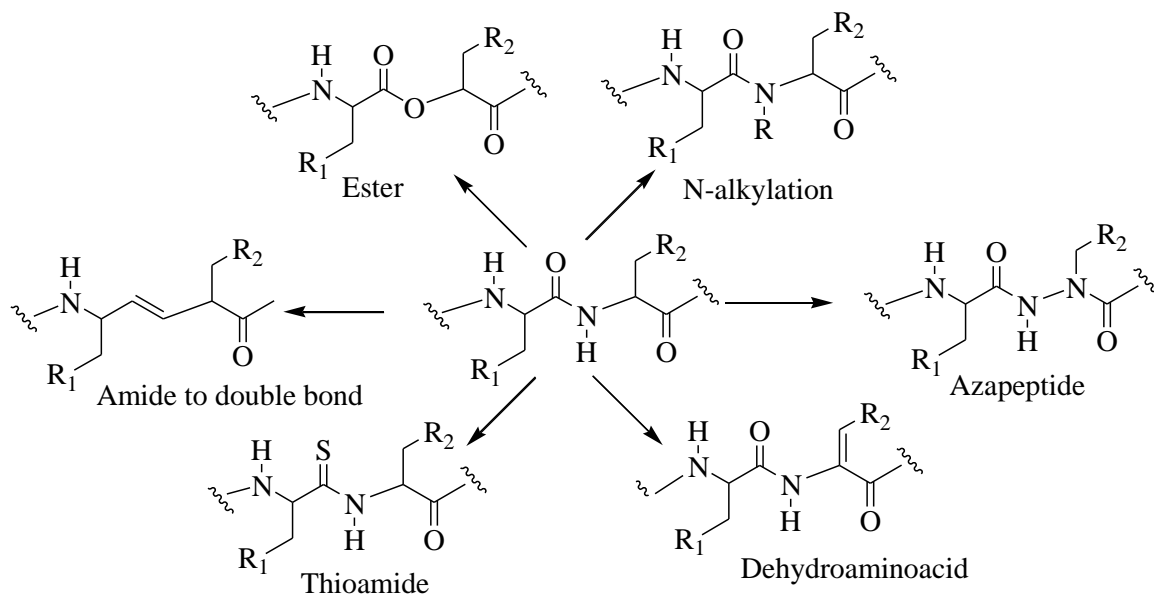


Figure 20: Amide group bioisosteres.

Heterocyclic bioisosteres of the amide bond are as follows (**Table 13**).

Table 13: Bioisosteres of the amide bond

<p>Amide</p>	<p>2-isoxazoline</p>
	<p>Imidazoline</p>
	<p>1,2,4-oxadiazoles</p>
	<p>1,3,4- oxadiazoles</p>
	<p>1,2,4-triazoles</p>

2.2. B.4. Halogen Bioisosteres³²

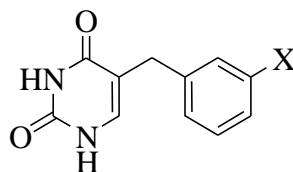


Figure 21: Halogen Bioisosteres.

Replacements of this type were observed in a series of 1-[(2- hydroxyethoxy) methyl]-5-benzyluracils that were tested for inhibition of liver uridine phosphorylase (UrdPase)³⁵. Uridine phosphorylase is an enzyme that catalyzes the reversible phosphorolysis of pyrimidine

nucleosides. Uridine phosphorylase is responsible for the degradation of chemotherapeutic agents such as 5-fluoro-2-deoxyuridylic acid ³⁶. Within the series of 5-benzyluracils, it was suggested that electron-withdrawing groups at the 3-position decreased potency. This hypothesis was supported by the observation that replacement of the chloro atom with stronger electron-withdrawing groups such as the cyano or the trifluoromethyl resulted in less potent analogues (Table 14).

Table 14: Uridine phosphorylase inhibition of 5-Benzyluracil

Compound	X	IC ₅₀
1	Cl	2.5
2	CN	13.2
3	CF ₃	21.4

3. ANALYSIS OF THE MODIFICATIONS RESULTING FROM ISOSTERISM

In general the isosteric replacement, even though it represents a subtle structural change, results in a modified profile: some properties of the parent molecule will remain unaltered, others will be changed. Isosteric modification can be governed by following parameters:-

- structural parameters
- electronic parameters
- solubility parameters

A. Structural parameters

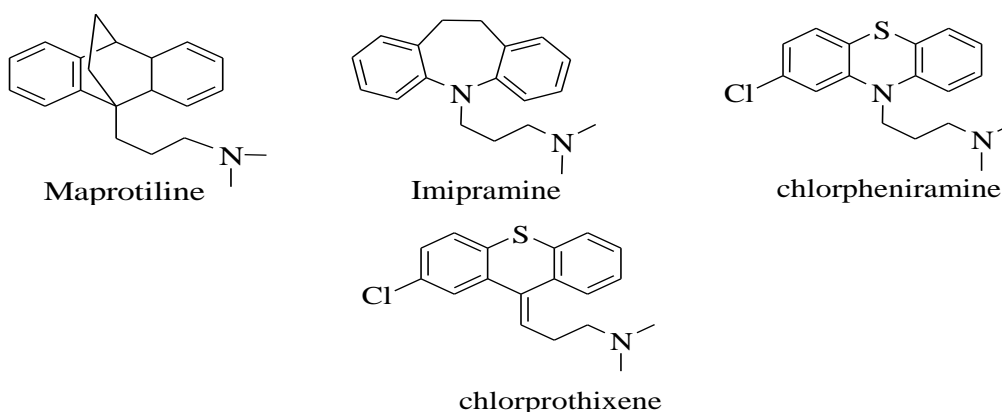


Figure 22: Dihedral angle formed by the two benzo rings dibenzazepine & dibenzocycloheptadiene.

These will be important when the portion of the molecule involved in the isosteric change serves to maintain other functions in a particular geometry. That is the case for tricyclic psychotropic drugs. In the two antidepressants (imipramine and maprotiline) the bioisosterism is geometrical that is the dihedral angle α formed by the two benzo rings is comparable: $\alpha = 65^\circ$ for the dibenzazepine and $\alpha = 55^\circ$ for the dibenzocycloheptadiene ³⁷. This same angle is only 25° for the

neuroleptic phenothiazines and the thioxanthenes. In these examples the part of the molecule modified by isosterism is not involved in the interaction with the receptor. It serves only to position correctly the other elements of the molecule³⁸.

B. Electronic parameters³⁹

It governs the nature and the quality of ligand–receptor or ligand enzyme interactions. The relevant parameters will be inductive or mesomeric effects, polarizability, pKa, capacity to form hydrogen bonds etc. Despite their very different substituents in the meta-position, the two epinephrine analogs exert comparable biological effects: they are both β -adrenergic agonists. In fact the key parameter resides in the very close pKa.

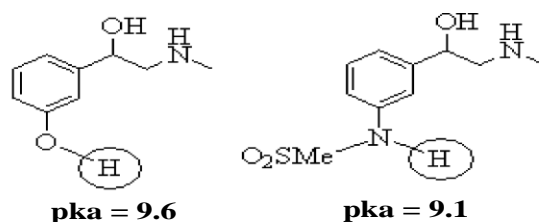


Figure 23: Non-classical isosterism, methyl sulfonamide substituent has comparable acidity to the phenolic hydroxyl group.

C. Solubility parameters⁴⁰

When the functional group involved in the isosteric change plays a role in the absorption, distribution or excretion of the active molecule, the hydrophilic–lipophilic parameters become important.

For example in an active molecule the replacement of $-\text{CF}_3$ ($\pi = +0.88$) by $-\text{CN}$ ($\pi = -0.57$), the electron-attracting effect of the two groups will be comparable, but the molecule with the cyano function will be clearly more hydrophilic. This loss in lipophilicity can then be corrected by attaching elsewhere on the molecule a propyl, isopropyl, or cyclopropyl group.

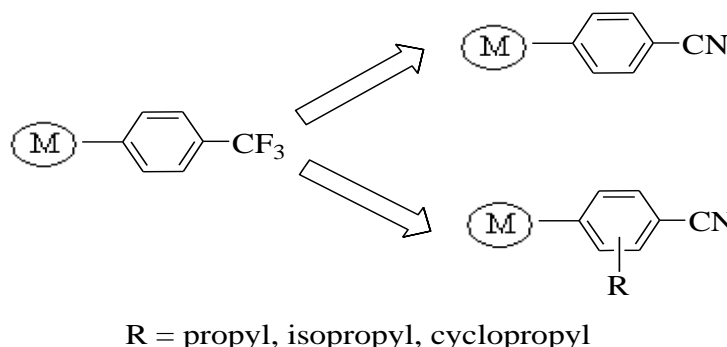


Figure 24: Replacement of CF_3 with CN .

4. MINOR METALLOIDS-TOXIC ISOSTERS

A. Bioisosteres involving selenium

Selenium can be considered the best isoster of sulfur as it is just below it in the periodic table. These two atoms have very similar physical properties: the radius of selenium is only 12.5% bigger than that of sulfur and their electro negativity is rather similar. Selenium and its derivatives are highly toxic, with the exception of ^{75}Se derivatives which serve diagnostic purposes (e.g. ^{75}Se -selenomethionine is used as a radioactive imaging agent in pancreatic scanning). Selenium bioisosteres of sulfur compounds are mainly used as research tools (e.g. bis [2-chloroethyl] selenide as selenium bioisostere of the classical sulfur mustards). Selenocysteine is present in the catalytic site of mammalian glutathione-peroxidase and this explains the importance of selenium as an essential trace. The only selenium-containing drug candidate is *ebselen* which owes its antioxidant and anti-inflammatory properties to its interference with the selenoenzyme glutathione-peroxidase ⁴¹. Because of its strongly bound selenium moiety only metabolites of low toxicity are formed ⁴².

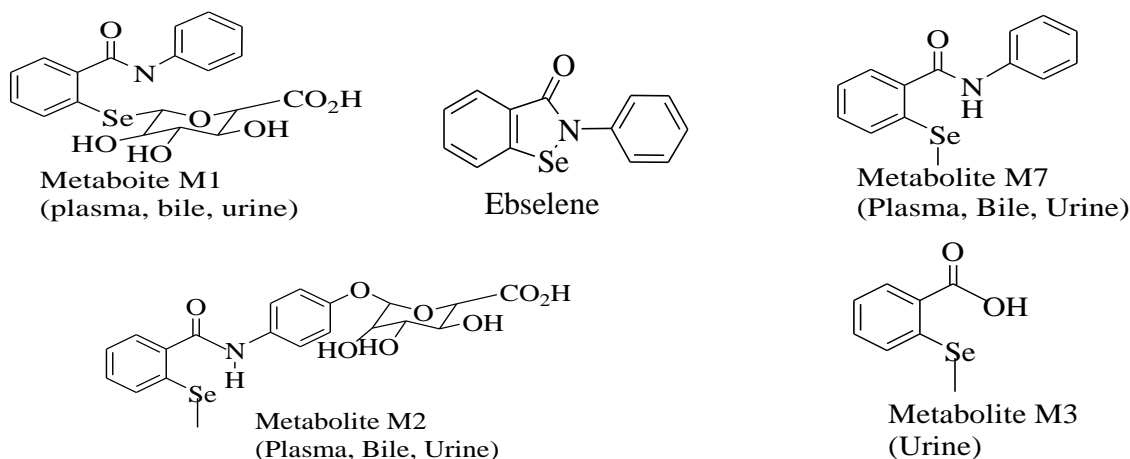


Figure 25: Ebselen and its main metabolites.

B. Carbon–boron isosterism

Compounds-containing carboxy boranes have shown anticancer, hypolipidemic and antifungal activity. Diazaborines are active against malaria and oxazaborolidines possess antibacterial activity ⁴³. Boronic chalcones are reported to be antitumor agents ⁴⁴. Organoboron derivatives, even more than organosilicon compounds, are sensitive to hydrolytic degradation that always leads to the final formation of boric acid. But boric acid has teratogenic properties in chickens. It produces the same malformations as those produced by a riboflavine (vitamin B₂) deficiency and the administration of riboflavine prevents these toxic effects. In man the chronic utilization of boron derivatives results in cases of borism (dry skin, cutaneous eruptions, and gastric troubles) tumors by Boron Neutron ⁴⁵.

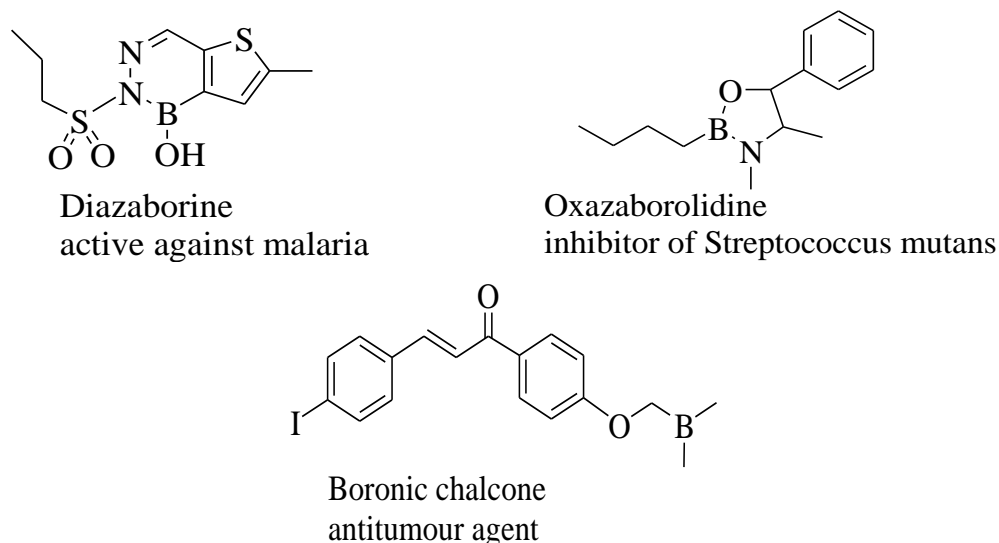


Figure 26: Boron-containing molecules with a biological activity.

A classical illustration of tetra substituted isosteres involves replacement of the quaternary ammonium group in case of cholinergic agonists with the phosphonium and arsonium analogues. In this study, it was observed that such replacements resulted in less potent analogues with greater toxicity. Activity was found to decrease as size of the onium ion increased. The decreased potency and greater toxicity of these higher elements has diminished interest in replacements of this type for the development of direct-acting cholinergic agonists⁴⁶.

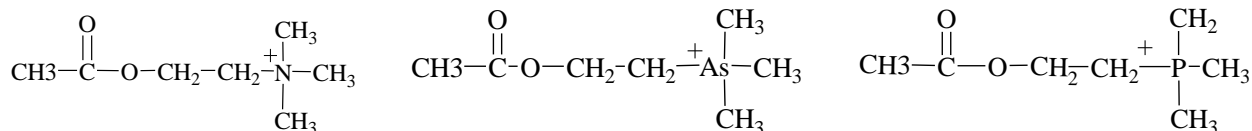


Figure 27: Cholinergic agonists.

CONCLUSION

- The bioisosteric replacements have significant value in lead optimization process. Examples of classical and non classical bioisosteric replacements in the hit to lead optimization process provide building blocks for the synthesis of frequently used bioisosteres.
- In drug design, the purpose of exchanging one bioisostere for another is to enhance the desired biological or physical properties of a compound without making significant changes in chemical structure.
- Bioisosteric replacement is not simple replacement with another isostere but we must analyse them by structural, solubility and electronic parameters to obtain molecules having similar biological activity.

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Activity and the metabolic activation pathway of the potent and selective hepatitis C virus pronucleotide inhibitor PSI-353661

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2'-Deoxy-2'-fluoro-2'-C-methylguanosine-5'-monophosphate

ABSTRACT

PSI-353661, a phosphoramidate prodrug of 2'-deoxy-2'-fluoro-2'-C-methylguanosine-5'-monophosphate, is a highly active inhibitor of genotype 1a, 1b, and 2a HCV RNA replication in the replicon assay and of genotype 1a and 2a infectious virus replication. PSI-353661 is active against replicons harboring the NS5B S282T or S96T/N142T amino acid alterations that confer decreased susceptibility to nucleoside/tide analogs as well as mutations that confer resistance to non-nucleoside inhibitors of NS5B. Replicon clearance studies show that PSI-353661 was able to clear cells of HCV replicon RNA and prevent a rebound in replicon RNA. PSI-353661 showed no toxicity toward bone marrow stem cells or mitochondrial toxicity. The metabolism to the active 5'-triphosphate involves hydrolysis of the carboxyl ester by cathepsin A (Cat A) and carboxylesterase 1 (CES1) followed by a putative nucleophilic attack on the phosphorus by the carboxyl group resulting in the elimination of phenol and the alaninyl phosphate metabolite, PSI-353131. Histidine triad nucleotide-binding protein 1 (Hint 1) then removes the amino acid moiety, which is followed by hydrolysis of the methoxyl group at the O⁶-position of the guanine base by adenosine deaminase-like protein 1 (ADAL1) to give 2'-deoxy-2'-fluoro-2'-C-methylguanosine-5'-monophosphate. The monophosphate is phosphorylated to the diphosphate by guanylate kinase. Nucleoside diphosphate kinase is the primary enzyme involved in phosphorylation of the diphosphate to the active triphosphate, PSI-352666. PSI-352666 is equally active against wild-type NS5B and NS5B containing the S282T amino acid alteration.

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1. Introduction

Selective inhibitors of HCV replication that target the NS3 protease and the NS5B RNA-dependent RNA polymerase (RdRp) in particular have been pursued as potential new therapies for chronic HCV infection. Nucleoside and nucleotide inhibitors of

Abbreviations: HCV, hepatitis C virus; HPLC, high pressure liquid chromatography; PCR, polymerase chain reaction; ddC, dideoxycytidine; GMP, guanosine-5'-monophosphate; GDP, guanosine-5'-diphosphate; RdRp, HCV RNA-dependent RNA polymerase; NS5B, HCV non-structural protein 5B; Cat A, cathepsin A; CES1, carboxylesterase 1; Hint 1, histidine triad nucleotide-binding protein 1; ADAL1, adenosine deaminase-like protein 1; hGUK1, human guanylate kinase; NDPK, human nucleoside diphosphate kinase; PK, pyruvate kinase; 3-PGK, 3-phosphoglycerate kinase; K_m , Michaelis constant; k_{cat} , turnover number; k_{cat}/K_m , catalytic efficiency.

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HCV are a desirable approach to treating HCV infection because of their high barrier to resistance and pan-genotype activity (Herlihy et al., 2008; McCown et al., 2008). The active triphosphates of nucleoside analog inhibitors of HCV replication act as non-obligate chain terminators of the polymerization process. Several nucleoside polymerase inhibitors have been in development. 2'- α -OH-2'- β -C-methylcytidine, the active component of valopicitabine, was the first polymerase inhibitor to enter clinical trials, but further development of this compound was terminated due to limited efficacy and side effects (Afdahl et al., 2004). R1626, the prodrug of 4'-azidocytidine (R1479), and RG7128, the diisobutyrate prodrug of 2'-deoxy-2'- α -fluoro-2'- β -C-methylcytidine (PSI-6130), were shown to have efficacy against HCV in clinical studies. However, the development of R1626 was stopped because of haematological side effects (Nelson et al., 2008). RG7128 is the most advanced anti-HCV nucleoside and is currently in Phase IIb clinical studies. We recently reported *in vitro* results for PSI-7851, a phosphorami-

date prodrug of 2'-deoxy-2'- α -fluoro-2'- β -C-methyluridine 5'-monophosphate (Lam et al., 2010). PSI-7851 was synthesised as a way of circumventing the lack of activity associated with 2'-deoxy-2'- α -fluoro-2'- β -C-methyluridine resulting from the inability of the compound to be phosphorylated to the corresponding 5'-monophosphate (Murakami et al., 2008). PSI-7851 consists of a mixture of two diastereoisomers, PSI-7976 and PSI-7977. Both PSI-7851 and the purified isomers demonstrated potent, specific and broad genotypic anti-HCV activity in replicon assays and infectious virus assays with PSI-7977 being the more active of the two isomers (Lam et al., 2010; Sofia et al., 2010). PSI-7977 is currently being evaluated in Phase II clinical trials.

A number of 2'-deoxy-2'- α -fluoro-2'- β -C-methylpurine analogs, including 2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine (PSI-6567), have weak activity in the replicon assay. This lack of activity was most likely due to the failure of cellular enzymes to efficiently metabolize the compound to the corresponding 5'-triphosphate, a result of the inefficient phosphorylation of the nucleoside to the 5'-monophosphate. Therefore, phosphoramidate prodrug methodology was employed as an approach to bypass the non-productive first phosphorylation step and to intracellularly deliver 2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine 5'-monophosphate. Our subsequent work led to the identification of the phosphoramidate prodrug PSI-352879, (S)-2-(((2R,3R,4R,5R)-5-(2-amino-6-methoxy-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy)-phenoxy-phosphorylamino)-propionic acid isopropyl ester), which has potent activity in the HCV replicon assay (Chang et al., 2010).

PSI-352879 is a mixture of two diastereoisomers, of which PSI-353661 ((S)-2-(((S)-[(1R,4R,5R)-5-(2-amino-6-methoxy-purin-9-yl)-4(R)-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino)-propionic acid isopropyl ester)) is the more active isomer in the replicon assay (Chang et al., 2010). Herein we describe the *in vitro* activity and metabolic pathway of the single isomer PSI-353661, a novel phosphoramidate prodrug of β -D-2'-deoxy-2'- α -fluoro-2'- β -C-methyl-6-methoxyguanosine-5'-monophosphate that has broad genotypic activity. Furthermore PSI-353661 remains active against HCV replicons containing the S282T mutation, a mutation that confers resistance to 2'- β -C-methyl-nucleoside/nucleotide analogs including PSI-6130, PSI-7851 and PSI-7977 (Lam et al., 2010; Sofia et al., 2010; Stuyver et al., 2006).

2. Materials and methods

2.1. Compounds

The following compounds were synthesised at Pharmasset and their purity determined by proton NMR, MS and HPLC analysis: PSI-353661 ((S)-2-(((S)-[(1R,4R,5R)-5-(2-amino-6-methoxy-purin-9-yl)-4(R)-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy]-phenoxy-phosphorylamino)-propionic acid isopropyl ester) (98.9% pure), PSI-6567 (2'-F-2'-C-methylguanosine) (>99% pure), PSI-353131 ((S)-2-(((2R,3R,4R,5R)-5-(2-amino-6-methoxy-purin-9-yl)-4-fluoro-3-hydroxy-4-methyl-tetrahydro-furan-2-ylmethoxy)-hydroxy-phosphorylamino)-propionic acid) (96.3% pure), PSI-353224 (2-amino-6-methoxypurine 2'-deoxy-2'- α -fluoro-2'- β -C-methylribose-5'-monophosphate) (99.8% pure), PSI-353222 (2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine-5'-monophosphate) (99.3% pure), PSI-353579 (2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine-5'-diphosphate) (99.7% pure), PSI-352878 ((2R,3R,4R,5R)-5-(2-amino-6-methoxy-purin-9-yl)-4-fluoro-2-hydroxymethyl-4-methyl-tetrahydro-furan-3-ol) (>97% pure), R1479 (4'-azidocytidine) (>95% pure), INX-08189 (a phosphoramidate prodrug of 2'- α -OH-2'- β -C-methylguanosine) (>99% pure), and the non-nucle-

oside NS5B inhibitors that include HCV-796 (5-cyclopropyl-2-(4-fluoro-phenyl)-6-[(2-hydroxy-ethyl)-methanesulfonyl-amino]-benzofuran-3-carboxylic acid methylamide) (>96% pure), a thiophene compound (3-((1*r*,4*r*)-*N*-isopropyl-4-methylcyclohexanecarboxamido)-5-phenylthiophene-2-carboxylic acid) (>99% pure), and a benzothiadiazine compound (*N*-{3-[4-hydroxy-1-(3-methyl-butyl)-2-oxo-1,2-dihydro-pyrrolo[1,2-*b*]pyridazin-3-yl]-1,1-dioxo-1,4-dihydro-1*λ*⁶-benzo[1,2,4]thiadiazin-7-yl]-*N*-methyl-methanesulfonamide) (>99% pure). A 2-phenol indole compound, 5a-amino-12-cyclohexyl-*N*-(*N,N*-dimethylsulfamoyl)-3-methoxy-4*b*,5*a*,6-tetrahydrobenzo [3,4] cyclopropano [5,6] azepino [1,2-*a*] indole-9-carboxamide, was synthesised by WuXi Apptec (Shanghai, China). PSI-352666 (2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine-5'-triphosphate) was synthesised by NuBlocks (Vista, CA). [³H]-labeled PSI-353661 was synthesised by Moravsek (Brea, CA) and [α -³²P]-UTP was purchased from Perkin Elmer (Waltham, MA). Gemcitabine (2'-deoxy-2'-difluorocytidine) was purchased from Hetero Drugs Ltd (Hyderabad, India). Zalcitabine (2',3'-dideoxycytidine, ddC) was purchased from RI Chemicals (Orange, CA). 3'-dCTP was purchased from Trilink Biotechnology (San Diego, CA). Telaprevir was synthesised by ACME Biosciences (Palo Alto, CA) and bis(4-nitrophenyl)phosphate (BNPP) was obtained from Sigma-Aldrich Corp. (St. Louis, MO).

2.2. Cells and viruses

The Clone A HCV genotype 1b (Con1 strain: GenBank Accession No. AJ238799.1) replicon cells (Apath LLC, Brooklyn, NY) were maintained in culture medium containing Dulbecco's Modified Eagle's medium (DMEM) with 10% fetal bovine serum (FBS), 1 mM sodium pyruvate, 4 mM L-glutamine, 0.1 mM non-essential amino acids (NEAA), 100 units/mL penicillin and 100 μ g/mL streptomycin and 1 mg/mL G418 (Invitrogen, Carlsbad, CA). The genotype 1b derived ET replicon (Con1 strain) with adaptative mutations E1202G, T1280I and K1846T (Lohmann et al., 2003) and the Lunet cell line (Koutsoudakis et al., 2007) were kindly provided by Dr. Bartenschlager (University of Heidelberg, Heidelberg, Germany). Plasmid DNA containing the genotype 1a replicon (H77 strain: NCBI reference NC_004102.1) with adaptive mutations P1496L and S2204I (Blight et al., 2003), and the genotype 2a J6/JFH-1 replicon were licensed from Apath. The J6/JFH-1 replicon encodes a partial core (first 19 amino acids) and 3'-non-translated region (NTR) from the J6 strain (GenBank Accession No. AF177036) and 5'-NTR and NS3 to NS5B region from the JFH-1 strain (GenBank Accession No. AB047639). Plasmids containing ET, H77 and J6/JFH-1 replicon were linearized with *Sca*I, *Hpa*I or *Xba*I, respectively. *In vitro* transcription and electroporation of the replicon RNA into Lunet cells was performed as described previously (Lam et al., 2010). Cells electroporated with the H77 replicon were selected in medium containing 0.75 mg/ml G418. Cells electroporated with the ET or J6/JFH-1 replicon were selected with medium containing 0.25 mg/ml G418. Huh7 and HepG2 cells (ATCC, Manassas, VA) were maintained in DMEM supplemented with 10% FBS, 4 mM L-glutamine, 0.1 mM NEAA, 100 units/mL penicillin and 100 μ g/mL streptomycin. CEM and BxPC3 cells (ATCC) were maintained in RPMI-1640 (Invitrogen) supplemented with 10% FBS, 100 units/mL penicillin and 100 μ g/mL streptomycin.

2.3. HCV RNA replication and viral inhibition assays

HCV replicon assays using Clone A or ET-Lunet cells were performed as described previously (Stuyver et al., 2006). H77-Lunet and J6/JFH-1-Lunet cells were tested in a similar manner. Briefly, replicon-containing cells were seeded at a density of 1500 or 3000 cells per well in a 96-well plate and were incubated with

serially diluted test compounds prepared in culture medium without G418 so that the final DMSO concentration was 0.5%. Plates were incubated at 37 °C in a 5% CO₂ atmosphere for 4 days. Inhibition of HCV RNA replication was determined by real time PCR (RT-PCR) (Stuyver et al., 2003) or by measuring the levels of luminescence expressed via the *firefly* luciferase (ET replicon) encoded within the replicon using the Bright-Glo luciferase reagent (Promega, Madison, WI). For the RT-PCR assay, total RNA was extracted using the RNeasy-96 kit as recommended by the Manufacturer (Qiagen, Valencia, CA), reverse transcribed into cDNA, and amplified using a primer and probe mix for HCV 5'-NTR RNA and human ribosomal RNA (rRNA) in a multiplex RT-PCR reaction as described previously (Stuyver et al., 2003). Primers and probes were designed for the HCV IRES region: sense primer 5'-AGC CAT GGC GTT AGT ATG AGT GT, antisense primer 5'-TTC CGC AGA CCA CTA TGG, and probes 5'-FAM-CCT CCA GGA CCC CCC CTC CC-TAM (GT 1b), 5'-FAM-CCT CCA GGC CCC CCC CTC CC-TAM (GT 2a).

Anti-HCV assays against the H77sV2 and JFH-1 infectious viruses were performed as previously described (Yi et al., 2006). Briefly, increasing concentrations of HCV inhibitors were added to En5-3 cells infected with the H77sV2 or JFH-1 virus. Fresh compound-containing medium was replaced every day for three days, after which cells were fixed and incubated with a primary HCV-core mouse monoclonal IgG1 antigen (Affinity BioReagents, Golden, CO), which was reacted with a FITC-labeled secondary antibody to mouse IgG (Kirkegaard & Perry Laboratories, Gaithersburg, MD). Clusters of infected cells were considered to constitute a single infectious focus. Foci counts were analyzed with a Zeiss LSM 510 laser scanning confocal microscope and EC₅₀ and EC₉₀ values were calculated using Graphpad Prism software (San Diego, CA).

2.4. Cytotoxicity assays

Cytotoxicity and mitochondrial toxicity assays with PSI-353661 were performed as described previously (Stuyver et al., 2002). For the cytotoxicity assay, Huh7 (2×10^3 cells/well), HepG2 (2×10^3 cells/well), BxPC3 (2×10^3 cells/well), or CEM (5×10^3 cells/well) cells were incubated with either PSI-353661, INX-08189 or Gemcitabine, for 8 days at 37 °C. At the end of the growth period, MTS dye from the CellTiter 96 Aqueous One Solution Cell Proliferation assay kit (Promega) was added to each well and the absorbance at 490 nm measured using a Victor3 plate reader (Perkin-Elmer, Boston, MA).

To assess the effect of PSI-353661 and INX-08189 on mitochondrial DNA synthesis, both compounds and ddC were serially diluted from 100 μM in assay medium containing DMSO and added to HepG2 or CEM cells seeded at 1×10^4 cells/well in a 24-well plate. Cells were incubated at 37 °C in a humidified 5% CO₂ atmosphere for 14 days, after which cells were harvested and total cellular DNA was extracted to perform a multiplex quantitative RT-PCR assay measuring the levels of the mitochondrial cytochrome C oxidase subunit II (COXII) gene and ribosomal DNA. The Δ Ct of mitochondrial COXII DNA (mtDNA) and Δ Ct of ribosomal DNA (rDNA) for each sample were determined as described previously (Stuyver et al., 2002). The fold difference in mitochondrial DNA normalized for ribosomal DNA relative to the no-drug control was calculated.

Lactic acid quantification was performed using the Enzy-Chrom™ L-Lactate Assay Kit (Bioassay System, Hayward, CA). Following a seven day incubation at 37 °C in the presence of various concentrations of PSI-353661, INX-08189, or ddC (0, 0.1, 1, 10 and 50 μM), the level of lactic acid in each sample of cell culture medium was determined as described in the Manufacturer's instructions. The total amount of lactic acid produced and the level of rDNA for each sample was determined as a percent of the

untreated control. The fold change in lactic acid production was calculated by dividing the percent of lactic acid by the percent of rDNA.

The human bone marrow stem cell toxicity assay was performed by Reachbio (Seattle, WA). Clonogenic progenitor cells of the erythroid (CFU-E and BFU-E) and myeloid (CFU-GM) lineages were set up in a methylcellulose-based medium. PSI-353661 was added to give final concentrations ranging from 0.5 to 50 μM. 5-Fluorouracil was used as a positive control at concentrations of 1, 0.1, and 0.01 μg/mL. The cultures were set up in triplicate at 2×10^4 cells per culture. Following 14 days of incubation the number of colonies was assessed.

2.5. Cross resistance studies

HCV Clone A replicon cells containing the S282T amino acid alteration in NS5B were established previously by selecting the cells in the presence of increasing concentrations of 2'-C-methyladenosine (Stuyver et al., 2006). The ET-S96T/N142T replicon was generated by site directed mutagenesis as previously described (Lam et al., 2010). Site directed mutagenesis was also used to generate the ET-C316Y, ET-M414T, ET-M423T, and ET-P495L mutations using the following primers and their complements: C316Y: 5'-CTG CAC GAT GCT CGT ATA CGG AGA CGA CCT TGT CG; M414T: 5'-CTA GGC AAC ATC ATC ACG TAT GCG CCC ACC, M423T: 5'-CCT TGT GGG CAA GGA CGA TCC TGA TGA CTC ATT TC, P495L: 5'-GGA AAC TTG GGG TAC TGC CCT TGC GAG TC (Integrated DNA Technologies, Coralville, IA). Cell lines containing each of these replicon mutants were generated by electroporation of the replicon RNA into Lunet cells and selecting for stable cells using 0.25 mg/ml G418. The anti-HCV activity of PSI-353661 against the Clone A wild type replicon and the replicon harboring the S282T amino acid alteration was evaluated using RT-PCR. The activity against replicons containing the S96T/N142T, C316Y, M414T, M423T, or P495L amino acid alteration was evaluated using genotype 1b ET-replicon cells and a *firefly* luciferase-based assay.

2.6. HCV replicon clearance and rebound studies

ET-Lunet replicon cells were seeded at 1×10^5 cells per well in a 6-well plate in culture medium without G418. PSI-353661 was added to cells at 0, 2, 5, 10 and 20-fold over its EC₅₀ value (4 nM) so that the final DMSO concentration was 0.5% in each well. When the cells reached approximately 90% confluency, they were passaged at ratios of 1:4 or 1:5 every 3–4 days for 2 weeks, and replenished with fresh medium containing the appropriate amount of PSI-353661. At each passage an aliquot of cells were harvested for RNA analysis. After 2 weeks, cells were passaged into culture medium containing 0.25 mg/ml G418 without inhibitor and cultured for an additional 2 weeks without passaging. At the end of the experiment, total RNA was extracted from all cell samples using the RNeasy-96 kit (Qiagen). Levels of HCV RNA and ribosomal RNA (rRNA) were determined using RT-PCR as described above. To determine relative log HCV RNA reduction, HCV RNA was normalized to rRNA by calculating Δ C_t (HCV C_t – rRNA C_t). $\Delta\Delta$ C_t was calculated by subtracting the average DMSO controls from each Δ C_t value. The level of HCV RNA was calculated as $\log 2^{-\Delta\Delta C_t}$. Results were expressed as log HCV RNA change in the compound-treated cells compared to the HCV RNA level in DMSO treated control cells. The effect of compound treatment was also examined by colony formation. Cells were washed with PBS, fixed using 7% formaldehyde, and stained with 1.25% crystal violet at the end of the four weeks experiment.

2.7. PSI-353661 metabolism studies in primary human hepatocytes

Primary human hepatocytes (CellDirect, Inc, Durham, NC) were seeded in cell plating medium (CellDirect, Inc) into 6-well plates at about 1×10^6 cells/well. After overnight incubation to allow the cells to attach, cells were incubated with [3 H]-PSI-353661 in cell maintenance medium (CellDirect, Inc) for up to 24 h at 37 °C in a 5% CO₂ atmosphere. At selected times, the medium was removed and the cell layer was washed with cold PBS. After trypsinization, cells were counted and centrifuged at 1200 rpm for 5 min. The cell pellets were suspended in 1 mL of cold 60% methanol and incubated overnight at –20 °C. The samples were centrifuged at 14,000 rpm for 5 min and the supernatants were collected and dried using a SpeedVac Concentrator (Thermo Electron Corporation), and stored at –20 °C until they were analyzed by high pressure liquid chromatography (HPLC). Residues were suspended in 100 μ L of water, and 50 μ L aliquots were injected onto the HPLC column. PSI-353661 and metabolites were separated by ion exchange HPLC with a Whatman 10 μ m SAX column (Whatman, Maidstone, England) using a Series 200 HPLC system (Perkin–Elmer, Waltham, MA). The mobile phase consisted of buffer A (0.02 M KH₂PO₄, pH 3.5) and buffer B (1 M KH₂PO₄, pH 3.5). Elution was performed using a linear gradient of buffer B from 0% to 100% for 100 min. Radioactivity was analyzed using a 610TR Radiometric Flow Scintillation Analyzer (Perkin–Elmer). PSI-353661 and its respective metabolites were identified based on the retention time of synthesised standards (Fig. 1). The intracellular concentration (pmol/10⁶ cells) of the metabolites was converted to μ M based on a 3 μ L cell volume per 10⁶ cells for normal human liver parenchymal cells (Duarte et al., 1989).

2.8. Hydrolysis of the carboxyl ester of PSI-353661

Hydrolysis of the carboxyl ester of PSI-353661 was evaluated using two enzymes, human cathepsin A (Cat A) (R&D Systems, Minneapolis, MN) and human carboxylesterase 1 (CES-1) (R&D Systems). For the CatA assay the enzyme was activated following the Manufacturer's instructions. Briefly, CatA (10 μ g/mL) was first activated by incubating with 1 μ g/mL cathepsin L (R&D Systems)

in 25 mM MES pH 6.0, 5 mM DTT for 30 min at 37 °C. Cat L was then inactivated by adding 10 μ M of the protease inhibitor E64. The hydrolysis of PSI-353661 by activated CatA was performed in reactions (100 μ L) containing 100 μ M PSI-353661 and 0.1 μ g of CatA in 25 mM MES pH 6.0, 0.1 M NaCl, 1 mM DTT, and 0.1% NP40. Hydrolysis of PSI-353661 by CES-1 was performed in reactions (100 μ L) containing 100 μ M PSI-353661 and 0.5 μ g of CES-1 in 50 mM Tris/HCl pH 7.5. After incubation at 37 °C for 30, 60, 90, 120 and 150 min, the reaction mixture was applied to an Ultra-cell 10 filter (Millipore, Billerica, MA) to remove the protein. The flow-through from the filter was collected and analyzed by reverse phase HPLC using a Gemini 5 μ m C18 column (Phenomenex, Torrance, CA). The mobile phase consisted of buffer A (water + 0.1% formic acid) and buffer B (acetonitrile + 0.1% formic acid). Elution was performed using a linear gradient of buffer B from 0% to 50% for 40 min. Amount of product formed was calculated based on the ratio of the peak areas of substrate and product, PSI-353131.

2.9. Hint1 studies

Hint1 was cloned, expressed and purified as described previously (Murakami et al., 2010). Hint1 reactions were performed at 37 °C in a reaction volume of 50 μ L containing 50 mM HEPES pH 7.2, 1 mM MgCl₂, 0.5 μ M Hint1, and varied concentrations of PSI-353131 (0.2–8.0 mM). After incubating the Hint1 reaction for different times (1, 2, 3, 4, and 5 h), the L-alanine dehydrogenase (Ala-DH) reaction was initiated by mixing with 150 μ L of 67 mM sodium carbonate buffer pH 10.0 containing 1.33 mM β -NAD and 0.067 U Ala-DH. After mixing, the pH of the solution was 9.5–10. It was confirmed that at this pH Hint1 activity was significantly reduced (<10% remaining). The initial rate of β -NADH product formation was measured. Amount of L-alanine in the mixture was calculated based on the rate of the reaction and the steady-state kinetic parameters for L-alanine (K_m and k_{cat}). Hint1 reaction rates were determined using varied concentrations of PSI-353131 based on the plots of the time dependent L-alanine formation. Steady-state kinetic parameters for PSI-353131 with Hint1 were determined by fitting the data to the Michaelis–Menten equation.

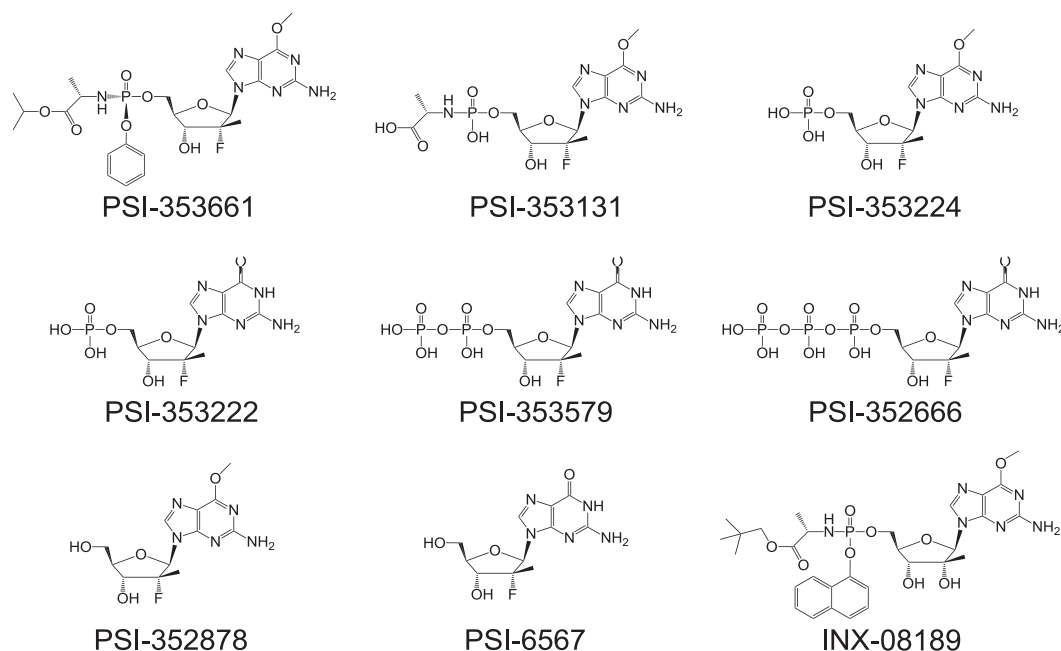


Fig. 1. Chemical structures of PSI-353661 and its metabolites

2.10. Human adenosine deaminase like protein1 studies

The cDNA fragment encoding ADAL1 was amplified from Huh7 cells using primers described by Schinkmanova et al. (2008): 5'-ATG ATA GAG GCA GAA GAG CAA CAG CCT TGC-3' and 5'-TTA AAT ATG TAA CAC TCT GGG CTT CAG GTG-3'. The amplified PCR product was confirmed by sequencing, cloned into the pET28a+ vector (Novagen, La Jolla, CA) and transformed into BL21-gold (DE3) cells (Stratagene, La Jolla, CA) for expression. ADAL1 was purified using a 1 mL Ni-affinity column (GE Healthcare, Piscataway, NJ) and eluted using a linear gradient (20–400 mM) of imidazole in buffer A, followed by a MonoQ 5/50 GL column (1 mL) (GE Healthcare) using a linear gradient of 0–1 M NaCl in 20 mM Tris/HCl pH 8.0, 10% glycerol, and 1 mM DTT. The fractions containing ADAL1 were identified by activity assay and SDS-PAGE analysis, which showed >95% purity. Protein concentration was determined based on the extinction coefficient ($30,160 \text{ M}^{-1} \text{ cm}^{-1}$) and the molecular weight of 40,263 Da using a Nanodrop spectrophotometer (Thermo Scientific, Wilmington, DE).

The ADAL1 assay was conducted in a 1 mL reaction mixture containing 50 mM potassium phosphate buffer pH 6.7, 2 mM DTT, 100 $\mu\text{g}/\text{mL}$ BSA, and varying concentrations of the test compound. The reactions were started by addition of the appropriate amount of ADAL1 and incubated at 37 °C in a Lambda 35 UV-Vis spectrophotometer (Perkin-Elmer). The reaction was followed by measuring the UV absorbance change at 258 nm for PSI-353224 ($\Delta\epsilon = 6,110 \text{ M}^{-1} \text{ cm}^{-1}$) and at 262 nm for *O*⁶-methyl-GMP ($\Delta\epsilon = 4,438 \text{ M}^{-1} \text{ cm}^{-1}$) and *O*⁶-methyl-dGMP ($\Delta\epsilon = 4,791 \text{ M}^{-1} \text{ cm}^{-1}$).

2.11. Phosphorylation of 2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine 5'-monophosphate (PSI-353222) by human guanylate kinase

Human guanylate kinase (hGUK1) was cloned from hGUK1 full-length cDNA obtained from Open Biosystems (Huntsville, AL) using a forward primer with a Nde I restriction site: 5'-CAG CCA TAT GTC GGG CCC CAG GCC TGT GGT GC-3', and a reverse primer with a Hind III site: 5'-CCG CAA GCT TCA GGC GCC GGT CCT TTG AGC-3'. The PCR product was digested and cloned into the pET28a+ vector (Novagen), expressed and purified by affinity chromatography as described above for ADAL1. The purified protein showed >95% purity based on SDS-PAGE analysis. hGUK1 concentration was measured using a spectrophotometer and calculated using an extinction coefficient of $7,450 \text{ M}^{-1} \text{ cm}^{-1}$ and the molecular weight (21.73 kDa). The purified protein was dialyzed against Buffer B (20 mM Tris/HCl pH 8.0, 150 mM NaCl, 10% glycerol, and 3 mM DTT) using a D-Tube Dialyzer Midi, MWCO 6–8 kDa (Novagen) overnight at 4 °C.

Phosphorylation of PSI-353222 by hGUK1 was studied spectrophotometrically as described previously (Murakami et al., 2007). The reaction was coupled with pyruvate kinase (PK) (Sigma-Aldrich) and lactate dehydrogenase (LDH) (Sigma-Aldrich). Oxidation of NADH (Sigma-Aldrich) was followed at 340 nm using a Lambda 35 UV/VIS Spectrometer (Perkin-Elmer). All assays were performed at 37 °C in a 1 mL reaction containing 64 mM Tris-HCl pH 7.5, 3.8 mM EDTA, 180 mM KCl, 12.8 mM MgCl_2 , 24 mM $(\text{NH}_4)_2\text{SO}_4$, 1 mM ATP, 0.5 mM phosphoenolpyruvate, 0.1 mM NADH, 5 IU/ml PK, 13.8 IU/ml LDH, nucleotide substrate, and purified hGUK1. The final enzyme concentration was 610 nM in reactions containing PSI-353222 and PSI-353224 as substrate and was 6.1 nM for GMP reactions. The reaction rate at different concentrations of the nucleotide substrate was determined and steady-state parameters were using GraphFit program version 5 (Erithacus Software, Horley, Surrey, UK).

2.12. Phosphorylation of 2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine-5'-diphosphate (PSI-353579)

Human nucleoside diphosphate kinase (NDPK) was cloned, expressed, and purified as described previously (Murakami et al., 2007). The phosphorylation of GDP and PSI-353579 by NDPK was studied spectrophotometrically using a coupled reaction as described above for hGUK1. The reaction was performed at 25 °C and the final concentration of NDPK in the reaction was 34.6 nM.

The phosphorylation of PSI-353579 by pyruvate kinase (PK) was studied spectrophotometrically. The reaction was followed at 340 nm using a Lambda 35 UV/VIS Spectrometer (Perkin-Elmer). All assays were performed at 25 °C in a 1 mL reaction mixture containing 50 mM imidazole, pH 7.5, 100 mM KCl, 60 mM MgSO_4 , 0.2 mM NADH, 1.5 mM phosphoenol pyruvate (PEP) and 5 U/ml of LDH, PSI-353579 and rabbit muscle PK. The final PK concentration in the reaction was 21 nM.

3-phosphoglycerate kinase (3-PGK) from bakers yeast was purchased from Sigma-Aldrich. The 3-PGK activity was measured using coupled reactions with a buffer containing 50 mM Tris-acetate (pH 7.5), 5 mM MgCl_2 , 1 mM NaF, 1 mM DTT, 10 mM sodium phosphate, 4 mM β -NAD, and 4 mM D,L-glyceraldehyde-3-phosphate as described previously (Krishnan et al., 2002). Prior to starting the reaction, phosphate donor (1.3-biphosphoglycerate) for the reaction was generated by preincubation with 5 units/ml of GAPDH (Sigma-Aldrich) for 20 min at 25 °C. The 3-PGK reaction was initiated by adding PSI-353579 (50, 200, and 500 μM) and 0.5 units/ml of 3-PGK and was incubated at 37 °C. After 0, 2, 5, 10, 20, and 40 min, a 100 μL aliquot was taken and the reaction was stopped on dry ice. The samples were thawed and filtered through 3K Amicon Ultra-0.5 mL centrifugal filters (Millipore) at 4 °C. Phosphorylation of PSI-353579 was analyzed by HPLC (Perkin-Elmer).

The reaction rate at different concentrations of the nucleotide substrate was determined and the steady-state parameters were determined with GraphFit. Since the NDPK reaction was coupled with the PK reaction, in order to determine NDPK-mediated phosphorylation rates, the rate for the PK reaction was determined at each concentration of substrate and subtracted from the rate of the NDPK-coupled reaction.

2.13. NS5B polymerase assay

IC_{50} values were determined for PSI-352666 or for 2'-C-methylguanosine 5'-triphosphate using recombinant HCV NS5B from wild-type or S282T genotype 1b NS5B (Con1 strain). Reactions were performed in a 20 μL mixture containing varying concentrations of the test compound, 5 μM of the four natural ribonucleotides, [α -³²P]UTP, 20 ng/ μL of genotype 1b (–) IRES RNA template, 1 unit/ μL of SUPERase in (Ambion, Austin, TX), 40 ng/ μL of NS5B, 1 mM MgCl_2 , 0.75 mM MnCl_2 , and 2 mM DTT in 50 mM HEPES buffer (pH 7.5). The reaction was quenched by adding 80 μL of stop solution (12.5 mM EDTA, 2.25 M NaCl, and 225 mM sodium citrate) after incubating at 27 °C for 30 min. The radioactive RNA products were separated from unreacted substrates by passing the quenched reaction mixture through a Hybond N+ membrane (GE Healthcare) using a dot-blot apparatus. The RNA products were retained on the membrane and the free nucleotides were removed by washing the membrane four times with a solution containing 0.6 M NaCl and 60 mM sodium citrate. After rinsing the membrane with water followed by ethanol, the membrane was exposed to a phosphorscreen and the products were visualized and quantified using a phosphorimager. The IC_{50} values were calculated using GraphFit. All the reactions were performed in triplicate and the results were reported as $\text{IC}_{50} \pm$ standard deviation (SD). PSI-352666 was also tested for activity

against recombinant NS5B from genotypes 2a, 3a, and 4a. The genotype 1b (Con1 strain) enzyme and 3a and 4a enzymes (cloned from genotype 3a or 4a HCV human serum samples obtained from SeraCare Life Sciences, Milford, MA) were prepared internally at Pharmasset as previously described (Lam et al., 2010). Genotype 2a enzyme (JFH-1 strain) was kindly provided by Dr. David Frick (University of Wisconsin, Milwaukee, WI) and Dr. Julie Heck (New York Medical College, Valhalla, NY). The experiments were essentially the same as described above except that the reactions contained 1 μ M of the four natural ribonucleotides and the reaction times were 20 min for genotype 1b, 2 h for genotype 2a, and 3 h for genotypes 3a and 4a NS5B.

3. Results

3.1. Inhibition of HCV replicon RNA and infectious viral replication

PSI-6567 (2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine) (Fig. 1) was shown previously to be a poor inhibitor of HCV replicon RNA replication (Clark et al., 2006). The reduced activity of PSI-6567 was subsequently demonstrated to be the result of the inability of cellular kinases to efficiently phosphorylate the compound to the 5'-monophosphate (Murakami, unpublished data). Therefore the phosphoramidate prodrug, PSI-353661 (Fig. 1), of the 2'- α -fluoro-2'- β -C-methylguanosine-5'-monophosphate was synthesised as an approach to bypassing this non-productive phosphorylation step. We compared the anti-HCV activity of PSI-353661 to that of PSI-6567 and of the recently described pro-drug of 2'- α -OH-2'- β -C-methylguanosine 5'-monophosphate (INX-08189, Fig. 1) (McGuigan et al., 2010) (Table 1). When incubated for 4 days (96 h) with genotype 1b replicon cells, the EC_{50} and EC_{90} values for PSI-353661 were 0.0030 ± 0.0014 μ M and 0.0085 ± 0.0007 μ M, respectively, whereas the EC_{50} and EC_{90} values for PSI-6567, the parent nucleoside, were 22.0 ± 7.7 μ M and 69.2 ± 19.3 μ M, respectively. The EC_{50} and EC_{90} values for INX-08189 were similar to those of PSI-353661. PSI-353661 also demonstrated potent activity against a genotype 1a H77 subgenomic replicon and a genotype 2a J6/JFH-1 subgenomic replicon (Table 1). In addition, the compound showed potent anti-HCV activity in the HCV 1a H77Sv2 and HCV 2a JFH-1 infectious virus assays (Table 2).

3.2. In vitro safety profile of PSI-353661

PSI-353661 and INX-08189 were evaluated for cytotoxicity in an 8-day assay using four different human derived cell lines: Huh7 (human hepatoma), HepG2 (human hepatoma), CEM (human T lymphocyte), and BxPC3 (human pancreatic) cells. Gemcitabine was included as a positive control for cytotoxicity (Table 3). The results showed no measurable cytotoxicity for PSI-353661 toward HepG2, BxPC-3, or CEM cells at 100 μ M, the highest concentration tested ($CC_{50} > 100$ μ M). The CC_{50} value determined for PSI-353661 with Huh 7 cells was 80.0 ± 6.0 μ M. In contrast, INX-08189 was found to be significantly more cytotoxic than PSI-353661 with

Table 2

Activity of PSI-353661 against genotype 1a and 2a infectious virus.

Infectious virus	EC_{50} (nM)	EC_{90} (nM)
GT1a_H77	3.5	8.6
GT2a_JFH-1	1.6	11.2

Values are reported as the average of duplicate experiments. Infectious foci were determined using a primary HCV-core mouse monoclonal antibody reacted with a FITC-labeled secondary antibody.

Table 3

Cytotoxicity of PSI-353661 and INX-08189.

Compounds	CC_{50} (μ M)			
	Huh7	HepG2	BxPC3	CEM
PSI-353661	80.0 ± 6.0	>100	>100	>100
INX-08189	0.32 ± 0.03	2.23 ± 0.1	2.81 ± 1.52	15.0 ± 2.0
Gemcitabine	<1	<1	<1	<1

Cells were incubated with PSI-353661 or INX-08189 (both up to 100 μ M) for eight days prior to determining the effect of these compounds in cell viability using a MTS assay. Gemcitabine (1 μ M) was included as positive control. Values are reported as the mean \pm SD from at least three independent experiments performed in triplicate.

CC_{50} values of 0.32 ± 0.03 , 2.23 ± 0.10 , 2.81 ± 1.52 , and 15.0 ± 2.0 μ M in Huh7, HepG2, BxPC3, and CEM cells, respectively.

Mitochondrial toxicity has been associated with long-term use of some nucleoside analogs (Fleischer and Lok, 2009; Lewis et al., 2003; Moyle, 2000; Tanji et al., 2001). As a measure of mitochondrial toxicity we assessed the effect of PSI-353661 and INX-08189 on mitochondrial DNA content and lactic acid production (Table 4). Exposing CEM and HepG2 cells to PSI-353661 at concentrations up to 100 μ M, the highest concentration tested, for 14 days did not affect mitochondrial DNA content. Because of the cytotoxicity associated with INX-08189 (as measured by reduction in rDNA), it was difficult to determine if the compound affected mitochondrial DNA synthesis. Dideoxycytidine (ddC), the positive control for mitochondrial toxicity, reduced mitochondrial DNA levels in CEM and HepG2 cells by more than one \log_{10} at the lowest concentration tested (0.5 μ M).

To test the effect of PSI-353661 on mitochondrial function, the level of lactic acid in the cell culture medium was quantified. When CEM and HepG2 cells were incubated with PSI-353661 for seven days no increase in lactic acid was seen at any of the concentrations tested (Table 4). A 3.1 ± 0.61 fold increase in lactic acid was seen when HepG2 cells were incubated with 1.0 μ M INX-08189 (Table 4). No increase in lactic acid was observed when CEM cells were incubated with up to 10 μ M INX-08189. An increase in lactic acid was seen when CEM and HepG2 cells were incubated with ddC at 1 and 10 μ M (CEM cells) and 10 μ M (HepG2 cells) (Table 4). Because of the cytotoxicity associated with INX-08189 and ddC (as measured by reduction in rDNA), it was not possible to accurately quantify the level of lactic acid at the higher concentrations.

The effect of PSI-353661 on the proliferation of human erythroid and myeloid progenitor cells was evaluated using a 14-day colony formation assay. An IC_{50} of >50 μ M was calculated for

Table 1

Activity of PSI-353661, PSI-6567 and INX-08189 against genotype 1a, 1b, and 2a replicons.

Replicon	PSI-353661		PSI-6567		INX-08189	
	EC_{50} , nM	EC_{90} , nM	EC_{50} , μ M	EC_{90} , μ M	EC_{50} , nM	EC_{90} , nM
GT1a_H77	4.5 ± 0.7	10.5 ± 0.7	19.0 ± 8.6	53.4 ± 12.8	1.4 ± 0.6	4.0 ± 0.7
GT1b_Con1	3.0 ± 1.4	8.5 ± 0.7	22.0 ± 7.7	69.2 ± 19.3	2.6 ± 0.5	4.6 ± 3.2
GT2a_JFH-1	6.5 ± 2.1	14.0 ± 1.4	38.3 ± 18.6	60.0 ± 14.8	7.3 ± 2.9	12.8 ± 3.5

Cells were treated with PSI-353661, PSI-6567, or INX-08189 for 4 days prior to determining HCV inhibition. Quantitative real time PCR or a luciferase-based reporter assay was used to quantify levels of HCV inhibition. Values are reported as the mean \pm SD from at least three independent experiments performed in duplicate.

Table 4
Mitochondrial toxicity of PSI-353661 and INX-08189 in CEM and HepG2 cells.

	CC ₉₀ (μM) ^a		Lactic acid ^b (Fold change from untreated cells)			
	mtDNA	rDNA	0.1 μM	1.0 μM	10 μM	50 μM
<i>HepG2 cells</i>						
PSI-353661	>100	>100	1.2 ± 0.35	1.2 ± 0.29	1.1 ± 0.06	1.3 ± 0.31
INX-08189	0.97 ± 0.62	0.75 ± 0.27	1.1 ± 0.21	3.1 ± 0.61	Toxic	Toxic.
ddC	<0.5	12.9 ± 9.1	1.6 ± 0.68	1.4 ± 0.12	3.9 ± 0.93	Toxic.
<i>CEM cells</i>						
PSI-353661	>100	>100	1.4 ± 0.36	1.4 ± 0.10	1.4 ± 0.31	1.3 ± 0.40
INX-08189	16.9 ± 1.7	16.3 ± 4.1	1.4 ± 0.35	1.4 ± 0.69	1.5 ± 0.50	Toxic.
ddC	<0.5	22.9 ± 11.9	0.9 ± 0.3	2.2 ± 0.81	4.2 ± 1.0	Toxic

^a Cells were incubated with PSI-353661 (up to 100 μM) or INX-08189 (up to 50 μM) for 14 days prior to determination of mitochondria COXII DNA (mtDNA) and ribosomal DNA (rDNA) using real time PCR ddC (up to 100 μM) was included as positive control.

^b Lactic acid levels were measure after 7-days using a colorimetric assay. The effect on lactic acid values are expressed as the ratio of percent change in lactic acid divided by the percent change in rDNA in order to account for the toxicity of the compounds. The fold change is expressed as the mean ± SD of the ratio of treated to untreated from at least three independent experiments performed in triplicate.

the erythroid progenitor cells (CFU-E and BFU-E) and for myeloid progenitor cells (CFU-GM) the IC₅₀ was calculated to be 45 ± 2 μM, suggesting that PSI-353661 was not significantly toxic toward bone marrow progenitor cells. At 1 μg/mL (4 μM) the positive control, 5-fluorouracil, inhibited CFU-E progenitor cell colony formation by >80% and completely inhibited BFU-E and CFU-GM colony formation.

3.3. Cross resistance studies

PSI-353661 was assessed for activity against replicons harboring the NS5B S282T or S96T/N142T amino acid alterations that confer decreased susceptibility to nucleoside/tide analogs. The S96T/N142T amino acid alterations confer resistance to 4'-azido-cytidine (R1479) (Le Pogam et al., 2006a), while the S282T amino acid alteration confers resistance to various 2'-β-C-methyl-modified nucleoside analogs as well as the pyrimidine analogs PSI-6130, PSI-7851, and PSI-7977, which have a 2'-α-fluoro-2'-β-C-methyl substitution on the ribose (Lam et al., 2010; Sofia et al., 2010; Stuyver et al., 2006). As shown in Table 5, neither the S96T/N142T amino acid alterations nor the S282T amino acid

alteration conferred resistance to PSI-353661. The EC₉₀ value for the wild-type replicon and the S96T/N142T replicon were 0.012 ± 0.004 and 0.009 ± 0.004 μM, respectively. As expected, the S96T/N142T replicon cells were less susceptible to 4'-azido-cytidine compared to cells containing the wild-type replicon. The S282T replicon also remained as sensitive to inhibition by PSI-353661 (EC₉₀ = 0.014 ± 0.003 μM) as wild-type (EC₉₀ = 0.009 ± 0.004 μM). The S282T replicon was resistant to PSI-7851 and INX-08189.

We also tested PSI-353661 against replicons containing amino acid alterations that confer resistance to non-nucleoside inhibitors of NS5B: these included C316Y, which confers resistance to HCV-796; M414T, which confers resistance to benzothiadiazine compounds; M423T, which confers resistance to thiophene compounds; and P495L, which confers resistance to 2-phenol indol compounds (Howe et al., 2008; Le Pogam et al., 2006a; Le Pogam et al., 2006b; Nguyen et al., 2003; Tomei et al., 2003). Results showed that PSI-353661 remained fully active against HCV replicons harboring any one of these resistant mutations, which conferred various levels of resistance towards each of the corresponding reference compounds (Table 5).

Table 5
Activity of PSI-353661 and other nucleoside or non-nucleoside analogs against replicons containing mutations that confer resistance to NS5B inhibitors. Values are reported as the mean ± SD from at least three independent experiments performed in duplicate.

NS5B resistant mutations	HCV inhibitors	EC ₉₀ (μM)		EC ₉₀ fold change ^a
		WT replicon	Mutant replicon	
S96T/N142T ^b	R1479	15.76 ± 3.67	74.12 ± 12.12	4.7
	PSI-7851	0.43 ± 0.23	0.39 ± 0.19	0.9
	PSI-353661	0.012 ± 0.0040	0.0087 ± 0.0037	0.7
	INX-08189	0.0054 ± 0.0036	0.0059 ± 0.0027	1.1
S282T ^c	R1479	22.35 ± 4.58	12.53 ± 3.78	0.6
	PSI-7851	0.44 ± 0.21	7.36 ± 1.02	16.7
	PSI-353661	0.0093 ± 0.0041	0.014 ± 0.0026	1.5
	INX-08189	0.018 ± 0.001	0.11 ± 0.004	6.1
C316Y ^b	HCV-796	0.026 ± 0.017	1.92 ± 0.77	73.9
	PSI-353661	0.012 ± 0.0040	0.0098 ± 0.0035	0.8
M414T ^b	Benzothiadiazine	1.23 ± 0.55	28.39 ± 13.70	23.1
	PSI-353661	0.012 ± 0.0040	0.012 ± 0.0022	1.0
M423T ^b	Thiophene	3.65 ± 0.21	69.52 ± 9.24	19.1
	PSI-353661	0.012 ± 0.0040	0.0097 ± 0.0019	0.8
P495L ^b	2-phenol indole	1.17 ± 0.46	60.75 ± 19.90	51.9
	PSI-353661	0.012 ± 0.0040	0.012 ± 0.00034	1.0

^a The fold change in EC₉₀, the concentration at which 90% inhibition occurred, was calculated by normalizing the EC₉₀ value in the mutant replicon with that of the wild-type.

^b Activity was determined by luciferase-based assay.

^c Activity was determined by RT-PCR assay.

3.4. PSI-353661 clears HCV replicon RNA and prevents viral rebound

Using the replicon system, PSI-353661 was evaluated for its ability to clear HCV replicon RNA from ET-Lunet cells. Replicon cells were incubated with PSI-353661, at approximately 2, 5, 10, and 20 times its EC_{50} value (4 nM), for 14-days in the absence of G418 and then for another 14-days in the presence of G418 and the absence of compound. Control cells incubated in the absence of PSI-353661 maintained a stable level of replicon RNA throughout the course of the experiment (Fig. 2A). By the end of the first 14-days, a 1-log reduction of HCV replicon RNA was observed at 8 nM PSI-353661 (2X EC_{50}) and a >4-log decrease in HCV replicon RNA at the 20 nM dose. By day 10 HCV RNA was undetectable at the 40 and 80 nM doses (Fig. 2A).

To determine whether PSI-353661 had cleared the HCV replicon after 14-days of treatment, the compound was withdrawn and the cells were incubated in the presence of G418 for an additional 14-days. Cells that contained replicon RNA grew in the presence of G418 whereas cells that were cleared of the replicon did not survive G418 treatment. As can be seen in Fig. 2B, partial clearance of replicon RNA was achieved with 20 nM PSI-353661 whereas 40 and 80 nM PSI-353661 completely eliminated the replicon, which resulted in the clearance of cells from those cultures and prevented rebound in HCV replicon RNA.

3.5. Metabolic profile of PSI-353661 in primary human hepatocytes

Cellular extracts from primary human hepatocytes incubated with 5 μ M [3 H]-PSI-353661 were analyzed by ion exchange HPLC and the metabolites identified by comparing the retention time of the radiolabeled metabolites to the retention time of known, chemically synthesised standards (Fig. 1). As shown in Fig. 3A, incubating primary human hepatocytes for 30 min with 5 μ M [3 H]-PSI-353661 led to the formation of the following intracellular metabolites: PSI-6567, PSI-352878, PSI-353131, PSI-353224, PSI-353222, PSI-353579 and PSI-352666. After 4 hours incubation, the major metabolites were PSI-6567, PSI-353222, PSI-353579 and PSI-352666 with PSI-352666 being the predominant metabolite (Fig. 3B).

A time course experiment was performed to assess the effect of time on the metabolism of PSI-353661 during continuous exposure of primary human hepatocytes to the compound (Fig. 4A and B). Throughout the course of the experiment, PSI-353661 was undetectable. The predominant metabolite formed when incubating PSI-353661 with primary human hepatocytes was the 5'-triphosphate, PSI-352666. PSI-352666 reached its maximum concentration at about 4 h and then declined. The monophosphate, PSI-353222, and the diphosphate, PSI-353579, appeared to have reached their maximum concentration after about 1–2 h of

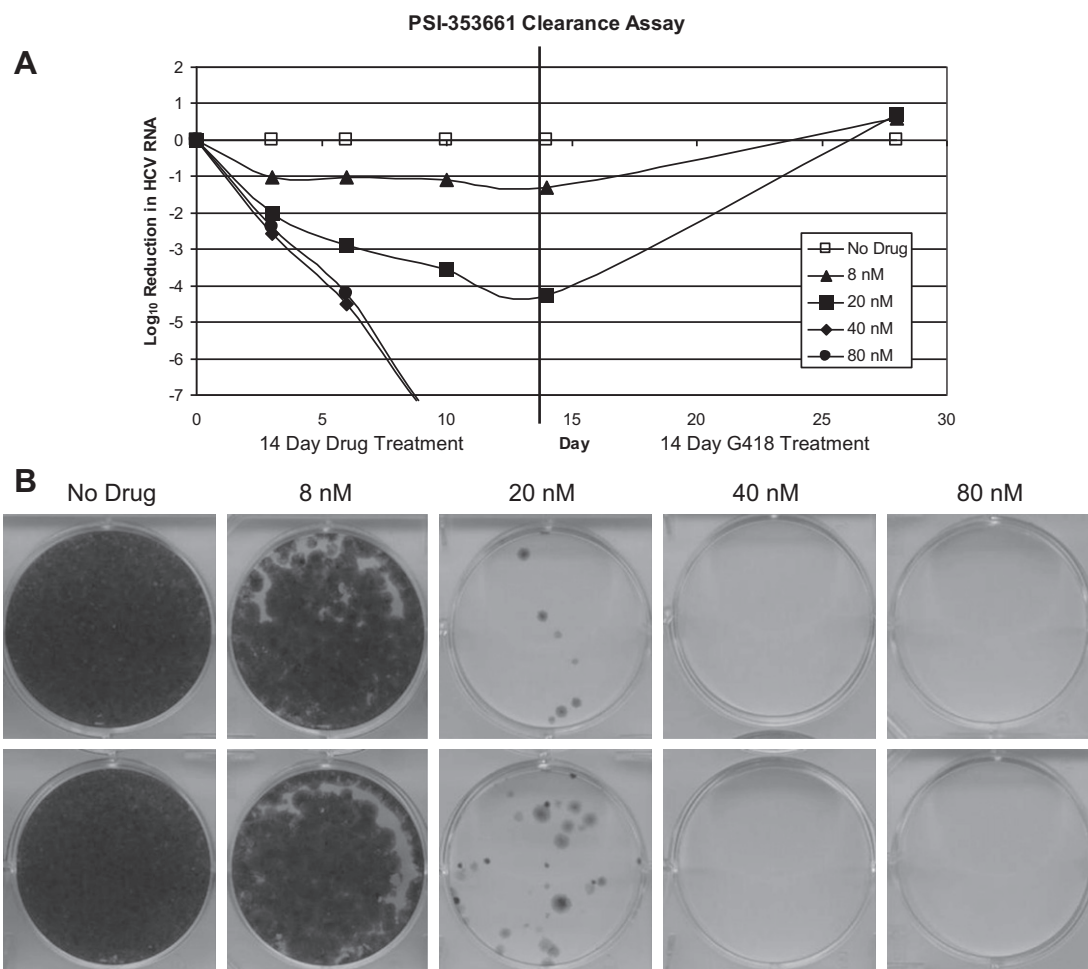


Fig. 2. Rebound and clearance assay. Replicon cells were treated with PSI-353661 for 2 weeks in the absence of G418. Fresh compound was added at every passage. After 2 weeks, cells were passaged into culture medium containing 0.25 mg/mL G418 without inhibitor and cultured for an additional 2 weeks without passaging. An aliquot of cells was harvested at each passage for replicon RNA analysis. Remaining cells were stained with crystal violet at the end of the experiment. (A) Effect of PSI-353661 on the level of HCV replicon RNA in the presence of 0 (□), 8 nM (▲), 20 nM (■), 40 nM (◆) and 80 nM (●) PSI-353661. Values for the level of HCV RNA are the average of two separate assays. (B) Colony formation in the presence of PSI-353661.

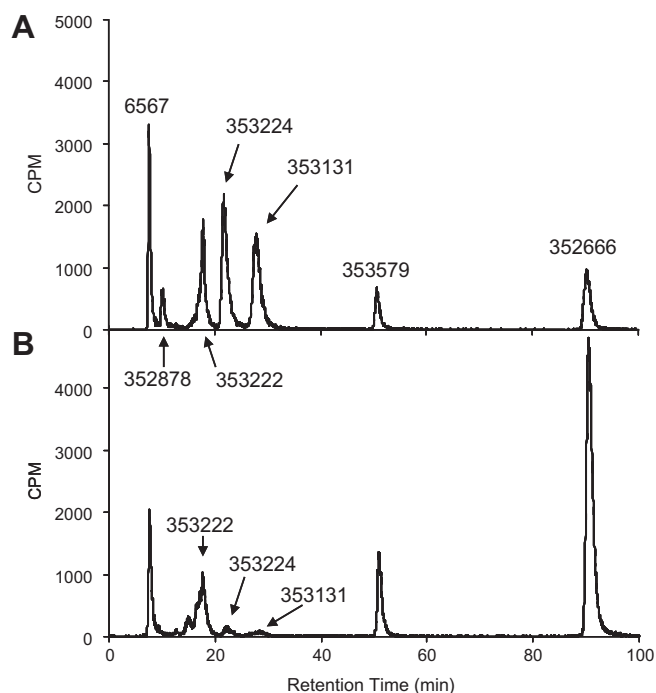


Fig. 3. Identification of metabolites. Primary human hepatocytes were incubated with 5 μM [^3H]-labeled PSI-353661 for 30 min (A) or 4 h (B) and formation of the intracellular metabolites were monitored by an anion exchange HPLC chromatography.

exposure. PSI-353131 reached its maximum concentration after 0.25 hours and declined rapidly thereafter. The concentration of the 6-methoxypurine monophosphate metabolite, PSI-353224,

declined rapidly after reaching its maximum concentration after 0.5–1 h of exposure. Both nucleoside analogs, PSI-352878 and PSI-6567, reached peak concentration after about 0.5–1 h of exposure.

In order to determine whether increasing the exposure of hepatocytes to higher concentrations of PSI-353661 would lead to increased formation of PSI-352666, primary human hepatocytes were incubated with PSI-353661 at different concentrations up to a maximal concentration of 100 μM for 4 h. PSI-353222, PSI-353579, and PSI-352666 increased with increasing concentrations of exogenous PSI-353661, but the rate of increase declined when the extracellular concentration of PSI-353661 was higher than 10 μM (Fig. 4C). However, the concentration of PSI-6567, PSI-353131 and PSI-353224 appeared to continue to increase as the extracellular concentration of PSI-353661 increased.

3.6. Identification of enzymes involved in metabolism of PSI-353661 to 2'-deoxy-2'-fluoro-2'-C-methylguanosine-5'-triphosphate

For PSI-353661 to be metabolized to PSI-352666 (2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine-5'-triphosphate), it must first be converted to 2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine-5'-monophosphate (PSI-353222), which involves a 4-step process. The first step is hydrolysis of the carboxylester by cellular enzymes. The second step is facilitated by the free carboxyl group performing a putative nucleophilic attack on the phosphorous releasing the phenol to produce the intermediate metabolite PSI-353131. The third step is removal of the amino acid moiety to form PSI-353224. The last step requires demethylation at the O^6 position of PSI-353224 to give PSI-353222.

We have previously demonstrated that the hydrolysis of the carboxylester linkage between the carboxylic acid and the alaninyl moiety isopropyl group of the phosphoramidate prodrug PSI-7977 is hydrolyzed by carboxylesterase 1 (CES1) and cathepsin A (Cat A)

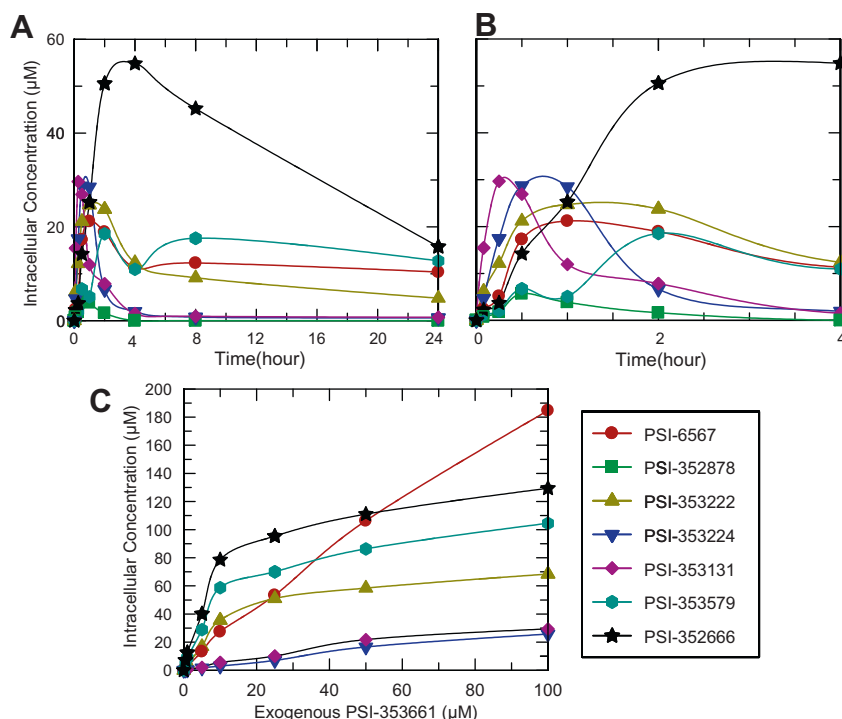


Fig. 4. Metabolic profile of PSI-353661. Time course of formation of PSI-353661 metabolites was monitored in primary human hepatocytes treated with 5 μM [^3H]-labeled PSI-353661 up to 24 h (A). The earlier time points were shown by plotting the same graph up to 4 h (B). Formation of the metabolites was followed in primary hepatocytes treated with varying concentrations of exogenous PSI-353661 for 4 h and the amount on intracellular metabolites were plotted against concentration of exogenously added PSI-353661 (C).

(Murakami et al., 2010). Here we demonstrate that PSI-353661 was hydrolyzed to PSI-353131 by CatA in a time-dependent manner with a specific activity of $0.80 \mu\text{mole min}^{-1} \text{mg}^{-1}$ (Fig. 5A). CES1 also hydrolyzed PSI-353661 but at a lower rate (estimated to be $0.034 \mu\text{mole min}^{-1} \text{mg}^{-1}$) and the reaction did not progress in a time-dependent manner (Fig. 5A).

To further substantiate that CatA and CES-1 were involved in the metabolism of PSI-353661, we studied the effect of telaprevir, an inhibitor of CatA and BNPP, a known inhibitor of CES-1 (Murakami et al., 2010), on the metabolism of PSI-353661 in primary human hepatocytes by following formation of the triphosphate, PSI-352666, in the presence of these inhibitors (Fig. 5B and C). BNPP inhibited the formation of PSI-352666 in hepatocytes from three different donors in a dose-dependent manner. However, as was observed with the phosphoramidate prodrug PSI-7977, the metabolism of PSI-353661 was not inhibited by telaprevir in hepatocytes from two of the three donors (Murakami et al., 2010).

The second enzymatic step in the activation of PSI-353661 can be catalyzed by histidine triad nucleotide-binding protein 1 (Hint1). The ability of Hint1 to catalyze the hydrolytic removal of L-alanine from PSI-353131 was tested using a two-step reaction. First, steady-state parameters for L-alanine with Ala-DH were determined as shown in Fig. 6A. The Michaelis constant (K_m) for alanine was $1227 \pm 65 \mu\text{M}$ and the V_{max} was $0.0206 \pm 0.0005 \Delta A_{340}/\text{min}$. This Ala-DH rate versus concentration curve was used to determine the concentration of alanine generated in the Hint1 reactions. After Hint1 reactions were performed, the concentration of the L-Ala product formed was quantified by performing the Ala-DH reaction and measuring the initial rate. The amount of L-Ala product was quantified at different incubation times to determine the rate of PSI-353224 formation in the Hint1 reaction (Fig. 6B inset). The steady-state kinetic parameters for PSI-353131 were determined by plotting the reaction rates against PSI-353131 concentration (Fig. 6B). The K_m value for PSI-353131 was $445 \pm 182 \mu\text{M}$ and k_{cat} was 3.44 min^{-1} .

It has been reported that an adenosine deaminase-like enzyme (ADAL1, N^6 -methyl-AMP aminohydrolase, abacavir monophosphate deaminase) is capable of catalyzing the hydrolytic deamination of natural substrates N^6 -methyl AMP, N^6,N^6 -dimethyl-AMP and N^6 -methyl dAMP and N^6 substituted purine-5'-monophosphate analogs (Schinkmanova et al., 2006; Schinkmanova et al., 2008). Because removal of the methyl group at the O^6 position of the guanine base is required during activation of PSI-353661 we examined the ability of ADAL1 to catalyze this reaction. PSI-353661, PSI-353131, and PSI-353224, along with N^6 -methyl AMP and O^6 -methyl GMP, were tested for their ability to serve as substrates for purified recombinant human ADAL1. Of the five compounds tested, only N^6 -methyl AMP, O^6 -methyl GMP and PSI-353224 were substrates for ADAL1 (Table 6). The K_m and k_{cat} values for PSI-353224 were $9.0 \pm 2.1 \mu\text{M}$ and $5.84 \pm 0.40 \text{ s}^{-1}$,

respectively, and a k_{cat}/K_m of $0.65 \mu\text{M}^{-1} \text{s}^{-1}$. K_m and k_{cat} values for O^6 -methyl GMP were $5.2 \pm 0.9 \mu\text{M}$ and $2.33 \pm 0.07 \text{ s}^{-1}$, respectively, and a k_{cat}/K_m value similar to that obtained for PSI-353224. N^6 -methyl AMP appeared to be a slightly less efficient substrate for ADAL1 with a k_{cat}/K_m of $0.13 \mu\text{M}^{-1} \text{sec}^{-1}$. Recombinant human adenylyl deaminase (AMPD) was also evaluated for its ability to use PSI-353224 as a substrate. PSI-353224 ($100 \mu\text{M}$) was incubated with AMPD for up to 72 h and the reaction mixture analyzed by HPLC. AMPD failed to convert PSI-353224 to PSI-353222, indicating that PSI-353224 was not a substrate for AMPD.

PSI-353222 was effectively phosphorylated to the corresponding 5'-diphosphate analog, PSI-353579, by recombinant hGUK1. Phosphorylation of PSI-353222 by hGUK1 was measured using a coupled spectrophotometric reaction. PSI-353222 was a substrate for purified hGUK1 with K_m and k_{cat} values of $64.9 \pm 1.6 \mu\text{M}$ and $0.31 \pm 0.03 \text{ s}^{-1}$, respectively. The natural substrate, guanosine-5'-monophosphate (GMP) was phosphorylated efficiently by hGUK1 with a k_{cat}/K_m value of $3.9 \mu\text{M}^{-1} \text{s}^{-1}$ (Table 7). However, compared with GMP, PSI-353222 was significantly less efficient as a substrate. The K_m for PSI-353222 indicated that PSI-353222 did not bind as tightly to the enzyme compared to GMP (3.4-fold weaker) and the k_{cat} value for PSI-353222 was significantly reduced, approximately 240-fold lower than that for GMP. No activity was observed when PSI-353224 was used as substrate at a concentration of $500 \mu\text{M}$, the highest concentration tested. These results strongly suggest that the hydrolysis of the O^6 -methyl group of PSI-353224 occurs before phosphorylation to the diphosphate takes place.

Phosphorylation of PSI-353579 (2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine-5'-diphosphate) to PSI-352666 can be carried out by at least three enzymes: nucleoside diphosphate kinase (NDPK), pyruvate kinase (PK), and 3-phosphoglycerate kinase (3-PGK). Of the three enzymes NDPK was the most efficient in catalyzing the reaction. The apparent steady-state parameters for PSI-353579 and the natural substrate, guanosine-5'-diphosphate (GDP), with NDPK are summarized in Table 8. The K_m for PSI-353579 was slightly higher compared to the K_m for GDP, while the k_{cat} for both was similar. Overall the catalytic efficiency (k_{cat}/K_m) for PSI-353579 was similar to that of GDP. Phosphorylation of PSI-353579 was also observed with PK and 3-PGK; however, the kinetic parameters were unable to be determined due to the poor affinity (K_m too high) of PSI-353579 for these enzymes.

3.7. Inhibition of HCV NS5B polymerase

The active triphosphate form, PSI-352666, acts as an alternate substrate for HCV RdRp to inhibit RNA synthesis. The ability of PSI-352666 to inhibit NS5B polymerase from different genotypes was tested in the presence of $1 \mu\text{M}$ NTPs. PSI-352666 was found to be active against recombinant NS5B from genotypes 1b, 2a, 3a,

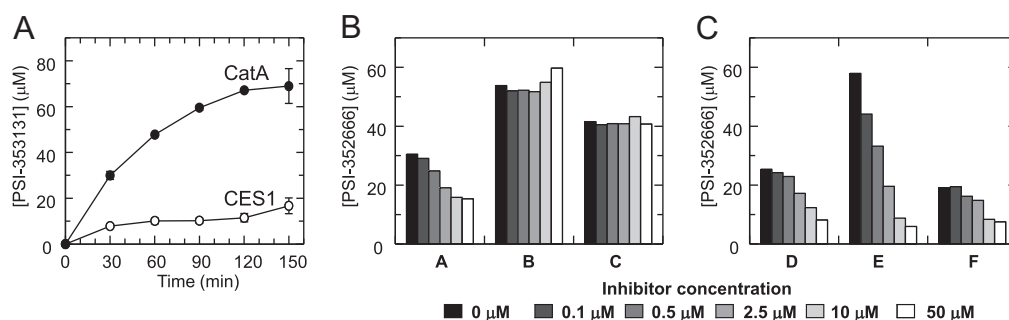


Fig. 5. Hydrolysis of PSI-353661 by CatA and CES1. CatA and CES1 reactions were performed in the presence of $100 \mu\text{M}$ PSI-353661 (A). Error bars represent standard deviation from three independent experiments. Metabolism of PSI-353661 was studied in primary human hepatocytes from different donors by following formation of the triphosphate, PSI-352666 in the presence of a CatA inhibitor, telaprevir, (Panel B; Donors A, B, and C) or a CES1 inhibitor, BNPP (Panel C; Donors D, E, and F).

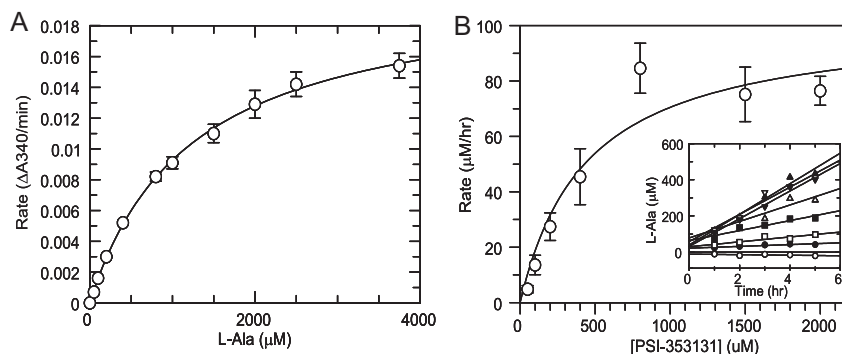


Fig. 6. Hint1 activity assay. A standard curve for L-alanine dehydrogenase (L-AlaDH) was generated by plotting the initial rates against the L-Ala concentrations (A). L-Ala formation was monitored for Hint1-mediated deamination of PSI-353131 (B). Rates were calculated from the individual time courses (inset). Bars in the graphs represent the standard error of the linear regression analysis from the individual time courses.

Table 6

Kinetic parameters for ADAL1-mediated demethylation.

	K_m (μM)	k_{cat} (s^{-1})	k_{cat}/K_m ($\mu\text{M}^{-1} \text{s}^{-1}$)
N^6 -meAMP	12.5 ± 1.6	1.58 ± 0.12	0.13
O^6 -meGMP	5.2 ± 0.9	2.33 ± 0.07	0.45
PSI-353661	No activity		
PSI-353131	No activity		
PSI-353224	9.0 ± 2.1	5.84 ± 0.40	0.65

Values are reported as the mean \pm SD from three independent experiments.

Table 7

Kinetic parameters for hGUK1-mediated phosphorylation.

	K_m (μM)	k_{cat} (s^{-1})	k_{cat}/K_m ($\mu\text{M}^{-1} \text{s}^{-1}$)
GMP	19.1 ± 4.5	74.7 ± 17.6	3.9
PSI-353222	64.9 ± 1.6	0.31 ± 0.03	0.005
PSI-353224	No activity		

Values are reported as the mean \pm SD from three independent experiments.

Table 8

Apparent kinetic parameters for NDPK-mediated phosphorylation.^a

	K_m (μM)	k_{cat} (s^{-1})	k_{cat}/K_m ($\mu\text{M}^{-1} \text{s}^{-1}$)
GDP	56.2 ± 4.9	38.8 ± 5.0	0.69
PSI-353579	66.3 ± 2.2	44.6 ± 4.9	0.67

Values are reported as the mean \pm SD from three independent experiments.

^a Kinetic parameters are apparent as PK contributed in the phosphorylation.

and 4a with average IC_{50} values of 1.0 ± 0.2 , 4.7 ± 0.6 , 1.3 ± 0.5 , and 4.2 ± 0.8 μM , respectively.

Additionally, we compared the ability of PSI-353666 and 2'- α -OH-2'- β -C-meGTP to inhibit wild-type and S282T mutant genotype 1b RdRp in the presence of 5 μM NTPs (Fig. 7A and B). With PSI-352666, there was approximately a 2-fold difference in IC_{50} values between wild-type ($\text{IC}_{50} = 4.0 \pm 1.1$ μM) and S282T mutant ($\text{IC}_{50} = 9.2 \pm 1.4$ μM) (Fig. 7A). When the reaction was performed with 2'- α -OH-2'- β -C-meGTP as the inhibitor, the S282T mutant RdRp showed a more than 10-fold reduction in sensitivity when compared with wild-type NS5B (Fig. 7B). The IC_{50} values for 2'- α -OH-2'- β -C-meGTP with the wild-type and the S282T RdRp were 6.8 ± 1.7 and 103 ± 18 μM , respectively.

4. Discussion

PSI-353661 is a single diastereoisomer of a phosphoramidate prodrug of 2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine-5'-

monophosphate currently undergoing preclinical evaluation. In addition to the phosphoramidate motif, PSI-353661 contains an O^6 -methyl modification on the guanine base. Our results demonstrated that PSI-353661 has potent activity against HCV in both the replicon assay and the infectious virus assay. We compared the activity of PSI-353661 with that of the recently reported phosphoramidate prodrug INX-08189 (McGuigan et al., 2010). Both PSI-353661 and INX-08189 showed similar activity in the replicon assay. However, INX-08189 was significantly more cytotoxic than PSI-353661.

Intracellular phosphorylation studies with PSI-353661 and primary human hepatocytes clearly show that PSI-353661 was transported into cells and phosphorylated to the active triphosphate form, PSI-352666. PSI-352666 was formed rapidly and achieved a high concentration (>50 μM) after 4 hours of exposure before declining. The first step in the metabolism of PSI-353661 involves hydrolysis of the carboxyl ester bond between the alaninyl moiety and the isopropyl alcohol (McGuigan et al., 1998a; McGuigan et al., 1998b). Recently, we demonstrated that PSI-7977 was a substrate for both CatA and CES1 (Murakami et al., 2010). Since PSI-353661 and PSI-7977 are both Sp isomers and have the identical phosphoramidate moiety in common, we tested CatA and CES1 for their ability to catalyze this hydrolytic reaction. As was observed with PSI-7977, PSI-353661 was a substrate for both enzymes. However, as we previously reported for PSI-7977 (Murakami et al., 2010), incubating CES1 with PSI-353661 resulted in unusual kinetics. Only a small fraction of PSI-353661 ($\sim 10\%$) was converted to product, PSI-353131. Although we do not completely understand the mechanism of this behavior, it is our hypothesis that PSI-353661 bind in an orientation that is essentially nonproductive. Further studies are ongoing to better understand this mechanism.

The effect of telaprevir and BNPP on the formation of the active triphosphate, PSI-352666, provides further evidence supporting the role of CatA and CES1 in the metabolism of PSI-353661. While telaprevir was an inhibitor of CatA, inhibition of the formation PSI-352666 by telaprevir was observed in hepatocytes from only one of three different donors. This may be due to differences in the level of expression of CatA in the hepatocytes from the different donors. It was previously demonstrated by Western blot analysis that the expression level of both CatA and CES1 varies substantially from one donor to another (Murakami et al., 2010). Therefore the lack of inhibition of PSI-353661 metabolism by telaprevir in hepatocytes from two of the three donors may be because CES1 is predominantly contributing to PSI-353661 hydrolysis in the cells from these two donors due to the differential expression of CatA and CES1.

Once the carboxyl ester is hydrolyzed, the phenol on the phosphate moiety is released via a non-enzymatic chemical reaction

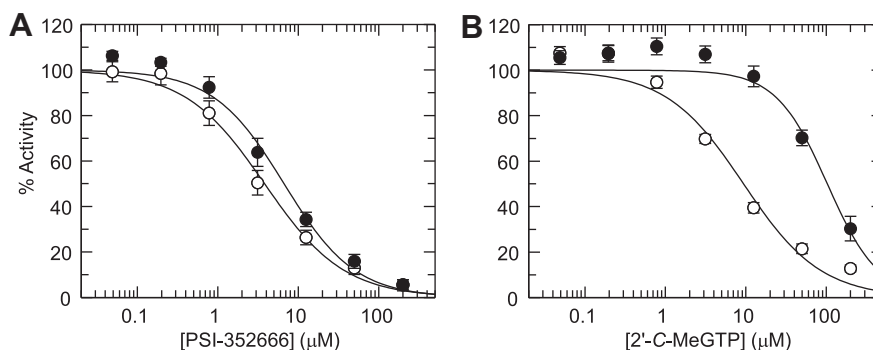


Fig. 7. Inhibition of wild type and S282T HCV NS5B polymerase with PSI-352666 and 2'- α -OH-2'- β -C-methylGTP. Activity of NS5B polymerase activity was plotted against concentrations of PSI-352666 (A) or 2'- α -OH-2'- β -C-methylGTP (B). The experiments were performed using wild type (open circles) and S282T mutant (closed circles) enzymes. Bars represent the standard deviation from at least three independent assays performed in duplicate.

that involves a nucleophilic attack of the carboxyl group at the phosphate affording the alaninyl phosphate intermediate, PSI-353131. As has been demonstrated by others, this chemical step is followed by the enzymatic removal of the amino acid moiety from the phosphate by Hint 1 (Chou et al., 2007; Murakami et al., 2010). While other enzymes may be involved in removing the amino acid moiety, our studies show that Hint1 is capable of converting PSI-353131 to PSI-353224.

Based on our metabolism and enzyme studies, the next step in the metabolism of PSI-353661 involved the demethylation of the compound resulting in the formation of 2'-deoxy-2'- α -fluoro-2'- β -C-methylguanosine-5'-monophosphate (PSI-353222). ADAL1 was identified as an enzyme capable of catalyzing this reaction. The last two steps in the metabolic pathway of PSI-353661 were catalyzed by hGUK1 and NDPK (the enzyme predominately involved in converting the diphosphate to the triphosphate), PK and 3-PGK.

The 5'-triphosphate, PSI-352666, was found to be an inhibitor of recombinant NS5B RdRp from genotype 1b, 2a, 3a, and 4a with IC_{50} values ranging from 1.0 ± 0.2 to 4.7 ± 0.6 μ M. Given that PSI-352666 was a potent inhibitor of the NS5B of HCV genotypes 1–4 and that PSI-353661 demonstrated potent activity against genotype 1a, 1b, and 2a replicons and in the H77 (genotype 1a) and JFH1 (genotype 2a) infectious virus assays suggests that PSI-353661 would provide broad genotype coverage.

We assessed the activity profile of PSI-353661 against replicons containing amino acid alterations in NS5B known to confer resistance to nucleoside analogs, S282T and S96T/N142T. Replicons containing either the S282T or the S96T/N142T amino acid alteration showed no change in sensitivity to PSI-353661 compared to the wild-type control replicon. As yet we have been unable to select for a mutation(s) that confer resistance to PSI-353661. However the S282T amino acid alteration did confer resistance to INX-08189. This was not surprising since the S282T amino acid alteration also confers resistance to the parent nucleoside, 2'-C-methylguanosine (Migliaccio et al., 2003). The active form of both prodrugs, the corresponding 5'-triphosphate, was tested against both recombinant wild-type NS5B and NS5B containing the S282T amino acid alteration. It is clear that there is a marked distinction between a purine having a 2'- β -methyl substitution with a 2'- α -fluoro versus a 2'- α -hydroxy substitution. Unlike the pyrimidines where the S282T amino acid alteration confers resistance to both the 2'- α -fluoro and the 2'- α -hydroxy substituted 2'- β -methyl analogs, the combination of the 2'- β -methyl group and the 2'- α -fluorine substitution seems to impart unique characteristics to the guanine nucleoside, which clearly has a dramatic effect on the resistance profile. It is not yet clear what it is about the 2'- α -fluorine relative to the α -hydroxyl substitution that

imparts this differentiating characteristic. The unique effect conveyed by the 2'- α -fluorine group could be the result of an electronic effect imparted on the nucleoside or some as yet unknown interaction with the polymerase.

Overall, these studies characterize the *in vitro* activity and safety and the mechanism of action of PSI-353661. Based on its preclinical *in vitro* activity and safety profile, PSI-353661 warrants further investigation.

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