



(51) International Patent Classification:
A61K 31/7076 (2006.01) *A61K 31/41* (2006.01)

(21) International Application Number:
PCT/US2011/062208

(22) International Filing Date:
28 November 2011 (28.11.2011)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
61/458,660 29 November 2010 (29.11.2010) US

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(81) Designated States (*unless otherwise indicated, for every kind of national protection available*): AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States (*unless otherwise indicated, for every kind of regional protection available*): ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:

- with international search report (Art. 21(3))
- before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments (Rule 48.2(h))
- with sequence listing part of description (Rule 5.2(a))

(54) Title: METHODS, ASSAYS AND COMPOUNDS FOR TREATING BACTERIAL INFECTIONS BY INHIBITING METHYLTHIOINOSINE PHOSPHORYLASE

(57) Abstract: The present invention discloses methods for treating bacterial infections in a subject comprising administering to the subject a sub-growth inhibiting amount of a 5'-methylthioinosine phosphorylase (MTIP) inhibitor, as well as assays for identifying such inhibitors, and compounds and pharmaceutical compositions comprising the inhibitors.



METHODS, ASSAYS AND COMPOUNDS FOR TREATING BACTERIAL INFECTIONS
BY INHIBITING METHYLTHIOINOSINE PHOSPHORYLASE

CROSS-REFERENCE TO RELATED APPLICATION

[0001] This application claims the benefit of U.S. Provisional Application No. 61/458,660, filed November 29, 2010.

STATEMENT OF GOVERNMENT SUPPORT

[0002] This invention was made with government support under Grant No. GM41916 awarded by the National Institutes of Health. The government has certain rights in the invention.

FIELD OF THE INVENTION

[0003] The present invention relates to methods, assay, compounds and compositons for treating infections caused by bacteria that use 5'-methylthioinosine phosphorylase (MTIP) in a quorum sensing pathway, such as *Pseudomonas aeruginosa*.

BACKGROUND OF THE INVENTION

[0004] Throughout this application various publications are referred to by Arabic numerals in parentheses. Full citations for these references may be found at the end of the specification immediately preceding the claims. The disclosures of these publications are hereby incorporated by reference in their entireties into the subject application to more fully describe the art to which the subject application pertains.

[0005] *Pseudomonas aeruginosa* is a Gram-negative bacterium found in a wide range of environments including water, soil, and mammals (1). It is a major opportunistic human pathogen, infecting burns, and lungs in cystic fibrosis (2). Compromised immune systems and extended hospitalization are correlated with infections, making *P. aeruginosa* the causative agent of approximately 15% of all hospital infections (2, 3). *P. aeruginosa* infections are difficult to treat since the bacterium has multiple antimicrobial resistance mechanisms (4). Chronic infections can be severe in patients with cystic fibrosis, causing high rates of morbidity and mortality (5, 6). *P. aeruginosa* infections are related to quorum sensing (QS) pathways, which regulate virulence factors and biofilm formation.

[0006] Signal molecules of QS include *N*-acyl-homoserine lactones (AHLs). The concentration of AHLs increases during bacterial growth to allow AHL binding to specific receptors and the regulation of target genes. In *P. aeruginosa*, the *las* and *rhl* QS systems use AHLs of 3-oxo-C₁₂-homoserine lactone and C₄-homoserine lactone as signal molecules, respectively. Microarray studies on *P. aeruginosa* indicated that QS regulated 3-7% of the total open reading frames (7-9). Deletion of single or multiple QS genes reduced the virulence of *P. aeruginosa* in mouse studies, indicating a strong correlation between the QS system and *P. aeruginosa* pathogenesis (10-15). Quorum sensing blockade does not affect bacterial growth and is therefore expected to attenuate the virulence of infection without causing drug resistance (16, 17).

[0007] Potential therapeutic targets in the QS system include enzymes involved in the formation of AHLs that act as signaling molecules and as virulence factors in *P. aeruginosa* (16). AHLs are synthesized from *S*-adenosylmethionine (SAM) and acylated-acyl carrier protein by AHL synthase with 5'-methylthioadenosine (MTA) as a by-product. MTA is recycled to ATP and methionine for SAM recycling (17). In bacteria, 5'-methylthioadenosine nucleosidase (MTAN) is the normal path to produce adenine and 5-methylthioribose- α -D-1-phosphate (MTR-1-P) from MTA for recycling. Transition state analogue inhibitors of *E. coli* and *V. cholerae* MTAN disrupted quorum sensing and reduced biofilm formation, supporting MTAN as a target for QS (17).

[0008] With the growing global threat of multi-drug resistance, nonconventional antibacterial discovery approaches are required that are nonlethal to bacteria where the potential to develop drug resistance is assumed to be less significant. The present invention addresses that need for infections caused by bacteria such as *Pseudomonas aeruginosa* that use 5'-methylthioinosine phosphorylase (MTIP) in a quorum sensing pathway.

SUMMARY OF THE INVENTION

[0009] The invention provides methods for treating infections caused by bacteria that use 5'-methylthioinosine phosphorylase (MTIP) in a quorum sensing pathway comprising administering to a subject having the infection a sub-growth inhibiting amount of a 5'-methylthioinosine phosphorylase (MTIP) inhibitor. The invention also provides pharmaceutical compositions comprising a sub-bacterial-growth inhibiting amount of a 5'-methylthioinosine phosphorylase (MTIP) inhibitor and a pharmaceutically acceptable carrier.

[0010] The invention also provides methods for determining whether or not a compound is a candidate for treating an infection caused by bacteria that use 5'-

methylthioinosine phosphorylase (MTIP) in a quorum sensing pathway, the method comprising determining whether or not the compound inhibits MTIP, wherein a compound that inhibits MTIP is a candidate for treating an infection caused by bacteria that use MTIP in a quorum sensing pathway and wherein a compound that does not inhibit MTIP is not a candidate for treating an infection caused by bacteria that use MTIP in a quorum sensing pathway.

BRIEF DESCRIPTION OF THE FIGURES

[0011] Figure 1. Early and late transition-state mimics as inhibitors for *Pa*MTIP and *Pf*PNP.

[0012] Figure 2. Structure-based sequence alignment. Protein are listed by PDB ID. The top 7 sequences are PNPs, the bottom 4 sequences are MTAPs, and *Pa*MTIP is 3OZB. The conserved residues in the phosphate binding site of PNPs (bold font) and MTAPs (bold font) are indicated. The conserved residues in the purine binding site of PNPs and MTAPs are underlined. The conserved Phe ("F") and His ("H") in ribose-binding site of PNPs are in italics. The conserved His and small hydrophobic amino acid in the (methylthio)ribose-binding site of MTAPs are in lower case letters. The residues in columns marked by * are conserved (but not identical) in all species. The Leu ("L") of *Pa*MTIP is not conserved in either MTAPs or PNPs and is in underlined italic. The PDB IDs are as follows: 1YR3, *E. coli* PNP II (SEQ ID NO:1); 1N3I, *Mycobacterium tuberculosis* PNP (SEQ ID NO:2); 3KHS, *Grouper Iridoviurs* PNP (SEQ ID NO:3); 2P4S, *Anopheles Gambia* PNP (SEQ ID NO:4); 1VMK, *Thermotoga maritime* PNP (SEQ ID NO:5); 1A9T, bovine PNP (SEQ ID NO:6); 1RR6, human PNP (SEQ ID NO:7); 3OZB, *Pa*MTIP (SEQ ID NO:8); 1V4N, *Sulfolobus tokodaii* MTAP (SEQ ID NO:9); 1K27, human MTAP (SEQ ID NO:10); 1WTA, *Aeropyrum pernix K1* MTAP (SEQ ID NO:11); 2A8Y, *Sulfolobus solfataricus* MTAP (SEQ ID NO:12).

[0013] Figure 3. Metabolism of [8-¹⁴C]MTA in *P. aeruginosa*. *P. aeruginosa* lysate was incubated with [8-¹⁴C]MTA for 0, 10, and 25 min, respectively. The ¹⁴C-metabolites MTA, MTI, adenine and hypoxanthine were purified using RP-HPLC and quantitated by scintillation counting. Bar graphs from left to right of figure for hypoxanthine, adenine, MTI and MTA.

DETAILED DESCRIPTION OF THE INVENTION

[0014] The invention provides a method for treating an infection caused by bacteria that use 5'-methylthioinosine phosphorylase (MTIP) in a quorum sensing pathway comprising

administering to a subject having the infection a sub-growth inhibiting amount of a 5'-methylthioinosine phosphorylase (MTIP) inhibitor.

[0015] The bacteria can be, for example, *Pseudomonas aeruginosa*, *Pseudomonas syringae* or *Xanthomonas campestris*.

[0016] The subject can be, for example, an animal, such as a mammal, such as a primate, such as a human, or a plant.

[0017] For example, in one embodiment, the bacteria is *Pseudomonas aeruginosa* and the subject is a human. In another embodiment, for example, the bacteria is *Pseudomonas syringae* or *Xanthomonas campestris* and the subject is a plant.

[0018] As used herein, to treat a bacterial infection in a subject means to reduce the virulence of the bacteria in the subject. The term “bacterial infection” shall mean any deleterious presence of bacteria in the subject.

[0019] The term “sub-growth inhibiting amount” of a MTIP inhibitor as used herein means an amount of the inhibitor, which when contacted with a population of bacteria, does not reduce the growth of the bacterial population. Preferably, the sub-growth inhibiting amount of the MTIP inhibitor inhibits quorum sensing in the bacteria. Preferably, the sub-growth inhibiting amount of the MTIP inhibitor is effective to reduce virulence of the bacteria without promoting the development of resistance by the bacteria to the MTIP inhibitor.

[0020] The term “quorum sensing” as used herein refers to the process by which bacteria produce and detect signaling molecules with which to coordinate gene expression and regulate processes beneficial to the microbial community. The term “inhibit quorum sensing” as used herein means altering this process such that coordination of gene expression and process regulation in microbial communities are impaired or prevented.

[0021] The invention also provides a pharmaceutical composition or an agrochemical composition comprising a sub-bacterial-growth inhibiting amount of a 5'-methylthioinosine phosphorylase (MTIP) inhibitor and a pharmaceutically or agrochemically acceptable carrier. Preferably, the pharmaceutical composition is formulated in dosage form. Preferably, the sub-bacterial-growth inhibiting amount of the MTAN inhibitor inhibits quorum sensing in bacteria. Preferred bacteria include, for example, *Pseudomonas aeruginosa*, *Pseudomonas syringae* and *Xanthomonas campestris*.

[0022] As used herein, “pharmaceutically acceptable carriers” are materials that (i) are compatible with the other ingredients of the composition without rendering the

composition unsuitable for its intended purpose, and (ii) are suitable for use with subjects as provided herein without undue adverse side effects (such as toxicity, irritation, and allergic response). Side effects are "undue" when their risk outweighs the benefit provided by the composition. Non-limiting examples of pharmaceutically acceptable carriers include any of the standard pharmaceutical carriers such as phosphate buffered saline solutions, water, and emulsions such as oil/water emulsions and microemulsions.

DEFINITIONS as applied to MTIP Inhibitors:

[0023] The term "alkyl" is intended to include straight- and branched-chain alkyl groups, as well as cycloalkyl groups and having up to 30 carbon atoms, and includes any C₁-C₂₅, C₁-C₂₀, C₁-C₁₅, C₁-C₁₀, or C₁-C₆ alkyl group. Cycloalkyl groups may have one or two carbon atoms substituted by a nitrogen, oxygen or sulfur atom. The same terminology applies to the non-aromatic moiety of an aralkyl radical. Examples of alkyl groups include, but are not limited to: methyl group, ethyl group, *n*-propyl group, *iso*-propyl group, *n*-butyl group, *iso*-butyl group, *sec*-butyl group, *t*-butyl group, *n*-pentyl group, 1,1-dimethylpropyl group, 1,2-dimethylpropyl group, 2,2-dimethylpropyl group, 1-ethylpropyl group, 2-ethylpropyl group, *n*-hexyl group and 1-methyl-2-ethylpropyl group. Examples of cycloalkyl group include cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, adamantyl, 2-tetrahydrofuranyl, 2-thietanyl, 3-piperidiny, 2-pyrrolidiny and 4-thiacyclohexanyl.

[0024] The term "alkenyl" means any hydrocarbon radical having at least one double bond, and having up to 30 carbon atoms, and includes any C₂-C₂₅, C₂-C₂₀, C₂-C₁₅, C₂-C₁₀, or C₂-C₆ alkenyl group, as well as cycloalkenyl groups, and is intended to include both straight- and branched-chain alkenyl groups. The same terminology applies to the non-aromatic moiety of an aralkenyl radical. Examples of alkenyl groups include but are not limited to: ethenyl group, prop-1-en-1-yl group, prop-2-en-1-yl group, prop-1-en-2-yl group, *cis*-but-2-en-1-yl group, *trans*-but-2-en-1-yl group, but-3-en-1-yl group, but-1-en-1-yl group, but-3-en-2-yl group, pent-4-en-1-yl group, 2-methyl-but-3-en-2-yl group, hex-5-en-1-yl group, cyclohex-1-enyl group, cyclohex-2-enyl group and cyclohex-3-enyl group

[0025] The term "alkynyl" means any hydrocarbon radical having at least one triple bond, and having up to 30 carbon atoms, and includes any C₂-C₂₅, C₂-C₂₀, C₂-C₁₅, C₂-C₁₀, or C₂-C₆ alkynyl group, and is intended to include both straight- and branched-chain alkynyl groups. The same terminology applies to the non-aromatic moiety of an aralkynyl radical. Examples of alkynyl groups include but are not limited to: ethynyl group, prop-1-yn-1-yl group, prop-2-yn-1-yl group, but-1-yn-1-yl group, but-2-yn-1-yl group, but-3-yn-1-yl group,

pent-1-yn-1-yl group, 3,3-dimethyl-but-1-yn-1-yl group, 2-methyl-but-3-yn-2-yl group and hex-1-yn-1-yl group.

[0026] The term “aryl” means an aromatic radical having 1 to 18 carbon atoms and includes heteroaromatic radicals. Examples include monocyclic groups, as well as fused groups such as bicyclic groups and tricyclic groups. Examples include but are not limited to: phenyl group, indenyl group, 1-naphthyl group, 2-naphthyl group, azulenyl group, heptalenyl group, biphenyl group, indacenyl group, acenaphthyl group, fluorenyl group, phenalenyl group, phenanthrenyl group, anthracenyl group, cyclopentacyclooctenyl group, and benzocyclooctenyl group, pyridyl group, pyrrolyl group, pyridazinyl group, pyrimidinyl group, pyrazinyl group, triazolyl group, tetrazolyl group, benzotriazolyl group, pyrazolyl group, imidazolyl group, benzimidazolyl group, indolyl group, isoindolyl group, indoliziny group, purinyl group, indazolyl group, furyl group, pyranyl group, benzofuryl group, isobenzofuryl group, thienyl group, thiazolyl group, isothiazolyl group, benzothiazolyl group, oxazolyl group, and isoxazolyl group.

[0027] The term “aralkyl” means an alkyl radical having an aryl substituent.

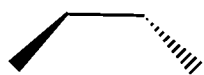
[0028] The term “aralkenyl” means an alkenyl radical having an aryl substituent.

[0029] The term “alkoxy” means an hydroxy group with the hydrogen replaced by an alkyl group.

[0030] The term “halogen” includes fluorine, chlorine, bromine and iodine.

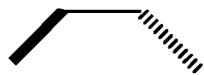
[0031] The term “prodrug” as used herein means a pharmacologically acceptable derivative of the MTIP inhibitor, such that an *in vivo* biotransformation of the derivative gives the MTIP inhibitor. Prodrugs of MTIP inhibitors may be prepared by modifying functional groups present in the compounds in such a way that the modifications are cleaved *in vivo* to give the parent compound.

[0032] As used herein, the structural formulae showing the “wedge” notation, *e.g.*:



are intended to represent pure enantiomeric forms of a trans isomer.

Similarly, the structural formulae showing the “rectangular” notation, *e.g.*:

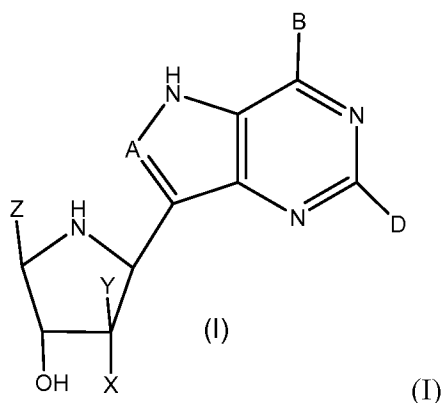


are intended to represent racemic mixtures of trans isomers.

[0033] It will be appreciated that the representation of the compound of the invention where B is a hydroxy group, is of the enol-type tautomeric form of a corresponding amide, and this will largely exist in the amide form. The enol-type tautomeric representation is

reproduced the referenced patents and patent applications simply to allow direct comparison the compounds of the invention with those in the references.

[0034] In one embodiment, as described in part in U.S. Patent No. 5,985,848 and in PCT International Patent Application Publication No. WO 99/19338, the contents of which are herein incorporated by reference, the MTIP inhibitor comprises a compound having formula (I):



wherein A is CH or N; B is OH; D is chosen from H, OH, NH₂, or SCH₃; and X and Y are independently selected from H, OH or halogen, except that when one of X and Y is hydroxy or halogen, the other is hydrogen; Z is selected from CH₂SQ, CH₂OQ or Q, where Q is an optionally substituted alkyl, alkenyl, aralkyl, aralkenyl or aryl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; provided that Q is not CH₂OH; or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

[0035] Preferably A is CH. Preferably D is H. Preferably X is OH and Y is H. Preferably Z is CH₂SQ. Preferably, when Z is CH₂SQ, Q is alkyl, preferably an optionally substituted C₁-C₆ alkyl group such as methyl, ethyl, propyl or butyl, or Q is an aryl group such as a phenyl group or an aralkyl group such as a benzyl group. It is also preferred that Q is an alkyl group substituted with one or more amino groups. Preferably Q is an alkyl group substituted with one or more amino and one or more carboxy groups.

[0036] Alternatively preferably, Z is Q. Preferably, when Z is Q, Q is an optionally substituted alkyl group, e.g. a C₁-C₆ alkyl group such as a methyl, ethyl, propyl or butyl group. It is also preferred that Q is an alkyl group substituted with one or more amino groups. Preferably Q is an alkyl group substituted with one or more amino and one or more carboxy groups.

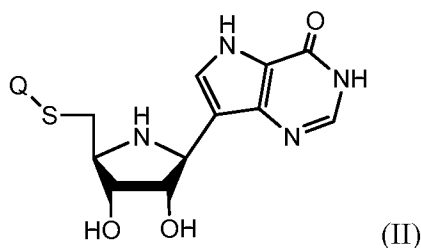
[0037] Alternatively, preferably, Z is Q and Q is an optionally substituted aralkyl group, e.g. a phenylethyl group. In still other preferred embodiments, Q is an optionally substituted aralkenyl group, e.g. a phenylethenyl group, e.g. a *cis*-phenylethenyl group or a *trans*-phenylethenyl group.

[0038] It is preferred that Z is selected from the group consisting of: phenylthiomethyl, p-chlorophenylthiomethyl, methylthiomethyl, ethylthiomethyl, propylthiomethyl, e.g. n-propylthiomethyl or isopropylthiomethyl, butylthiomethyl, -CH₂S(CH₂)₂CH(NH₂)COOH, methyl, ethyl, propyl, butyl, benzyl, *cis*-phenylethenyl, *trans*-phenylethenyl and -(CH₂)₄CH₃C(NH₂)COOH.

[0039] Preferably when one or more halogens are present they are chosen from chlorine and fluorine.

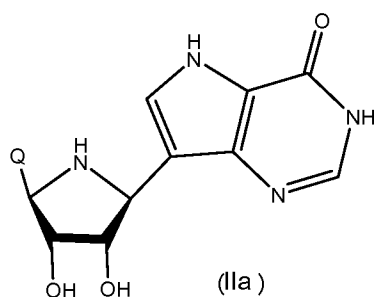
[0040] In one embodiment, A is CH or N; B is OH; D is chosen from H, OH, NH₂, or SCH₃; and X is OH; Y is H; Z is Q, where Q is a methyl group optionally substituted with a halogen, a methoxy, an amino or a carboxy group, or Q is an optionally substituted aralkyl, aryl or C₂-C₁₀ alkyl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

[0041] Preferred compounds include those having the formula (II):



where Q is aryl, aralkyl or alkyl, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; or a tautomer thereof; or a pharmaceutically acceptable salt thereof, or an ester thereof. Preferably, Q is alkyl, preferably a C₁-C₆ alkyl group such as methyl, ethyl, propyl or butyl, or Q is an aryl group such as a phenyl group or an aralkyl group such as a benzyl group. Preferably Q is an alkyl group substituted with one or more amino and one or more carboxy groups. Preferred compounds include those where Q is methyl or optionally substituted phenyl.

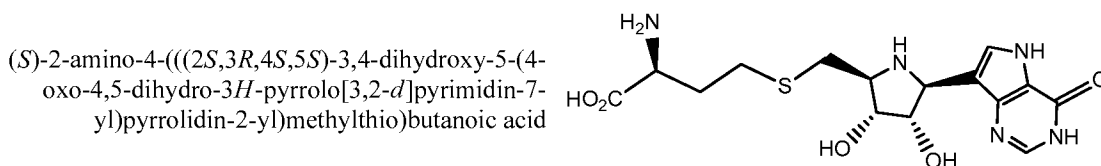
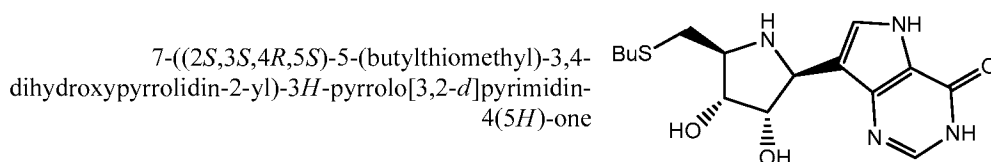
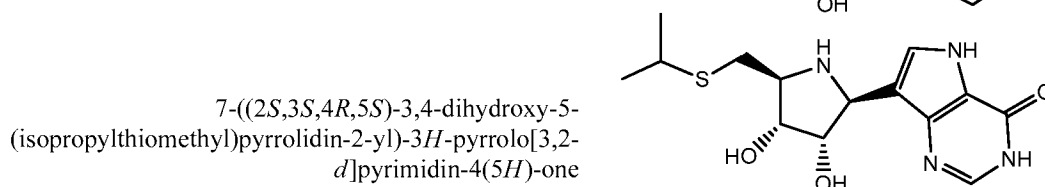
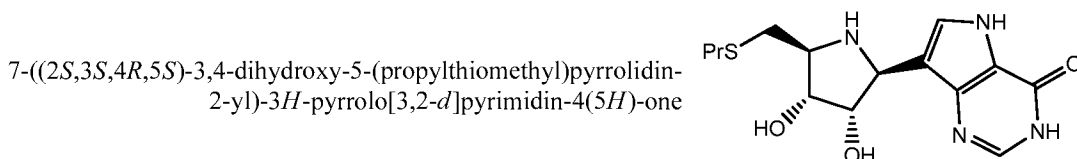
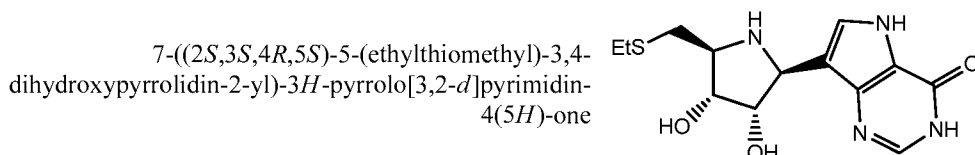
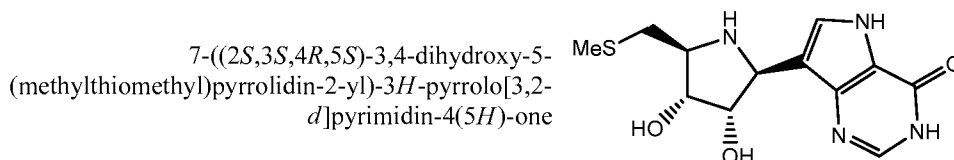
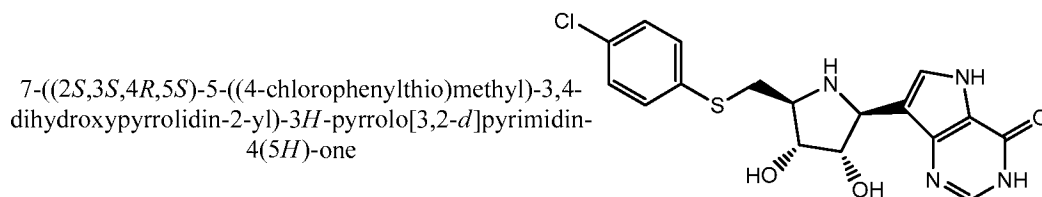
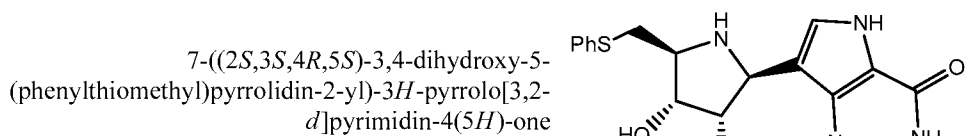
[0042] Additional preferred compounds include those having the formula (IIa):



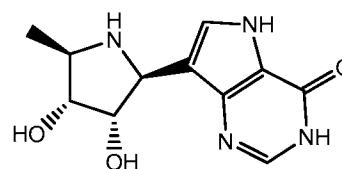
where Q is a methyl group optionally substituted with a halogen, a methoxy, an amino or a carboxy group, or Q is an optionally substituted aralkyl, aryl or C₂-C₁₀ alkyl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; or a tautomer thereof; or a pharmaceutically acceptable salt thereof, or an ester thereof. Preferably, Q is alkyl, preferably a C₁-C₆ alkyl group such as methyl, ethyl, propyl or butyl, or Q is an aryl group such as a phenyl group or an aralkyl group such as a benzyl group or an aralkenyl group such as a phenylethenyl group e.g. a *cis*-phenylethenyl group or a *trans*-phenylethenyl group. Preferably Q is an alkyl group substituted with one or more amino and one or more carboxy groups.

[0043] Examples of compounds of formula (I) include:

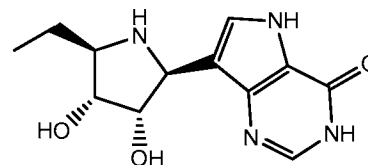
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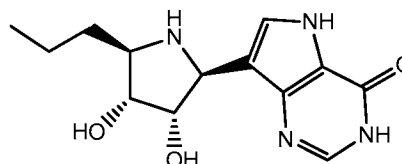
7-((2*S*,3*S*,4*R*,5*R*)-3,4-dihydroxy-5-methylpyrrolidin-2-yl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



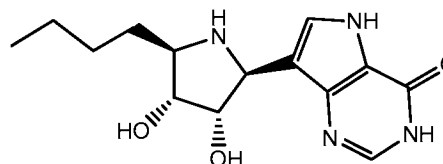
7-((2*S*,3*S*,4*R*,5*R*)-5-ethyl-3,4-dihydroxypyrrolidin-2-yl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



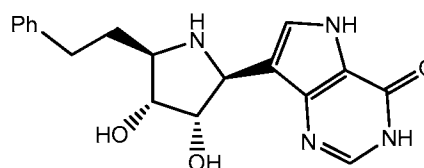
7-((2*S*,3*S*,4*R*,5*R*)-3,4-dihydroxy-5-propylpyrrolidin-2-yl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



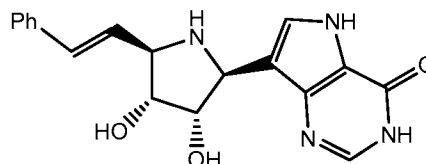
7-((2*S*,3*S*,4*R*,5*R*)-5-butyl-3,4-dihydroxypyrrolidin-2-yl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



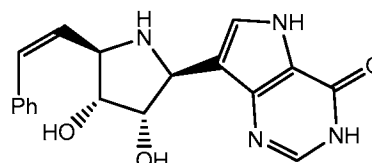
7-((2*S*,3*S*,4*R*,5*R*)-3,4-dihydroxy-5-phenethylpyrrolidin-2-yl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



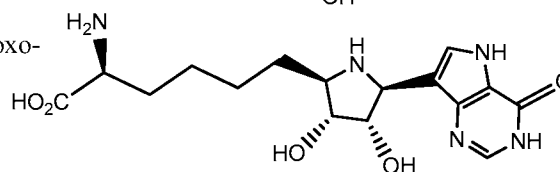
7-((2*S*,3*S*,4*R*,5*R*)-*cis*-3,4-dihydroxy-5-styrylpyrrolidin-2-yl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



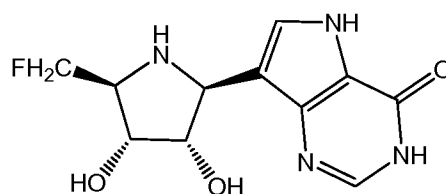
7-((2*S*,3*S*,4*R*,5*R*)-*trans*-3,4-dihydroxy-5-styrylpyrrolidin-2-yl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



(*S*)-2-amino-6-((2*R*,3*R*,4*S*,5*S*)-3,4-dihydroxy-5-(4-oxo-4,5-dihydro-3*H*-pyrrolo[3,2-*d*]pyrimidin-7-yl)pyrrolidin-2-yl)hexanoic acid

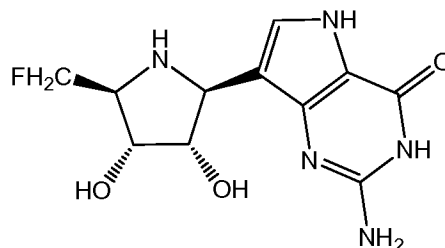


7-((2*S*,3*S*,4*R*,5*S*)-5-(fluoromethyl)-3,4-dihydroxypyrrolidin-2-yl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



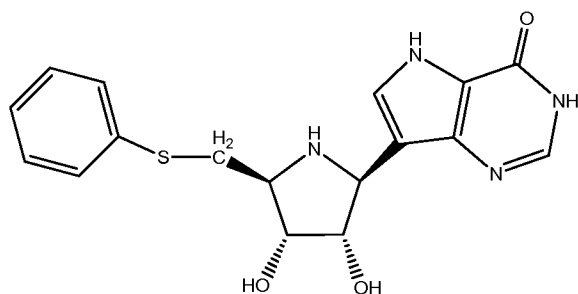
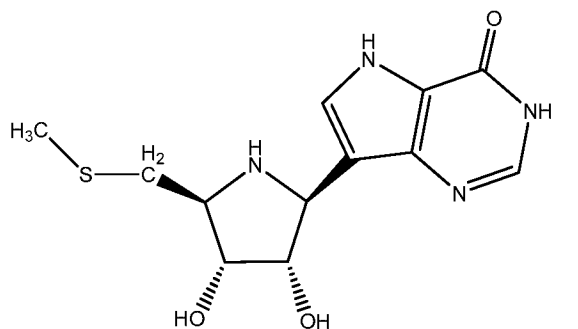
and

2-amino-7-((2*S*,3*S*,4*R*,5*S*)-5-(fluoromethyl)-3,4-dihydroxypyrrolidin-2-yl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

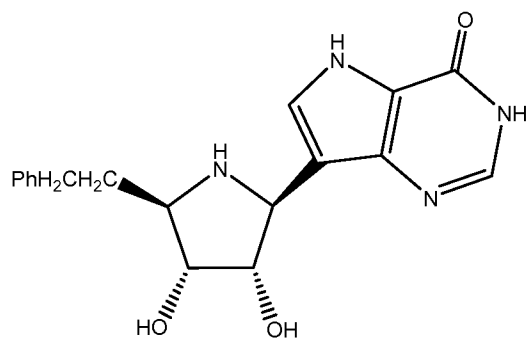


or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

[0044] Preferred compounds include those having the structure:

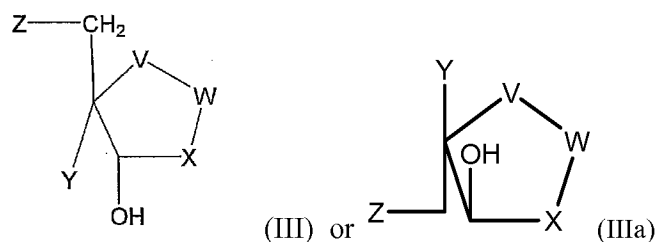


or

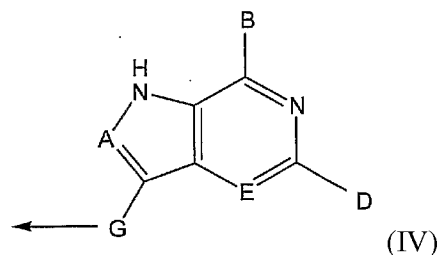


[0045] In another embodiment, as described in U.S. Patent No. 7,553,839 and in PCT International Patent Application Publication No. WO 04/018496, and in WO 2007/069924,

the contents of which are herein incorporated by reference, the MTIP inhibitor comprises a compound having formula (III) or (IIIa):



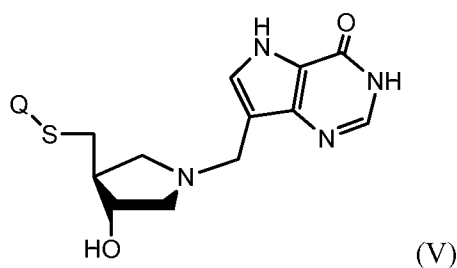
wherein V is CH₂ and W is NR¹; X is CH₂; Y is selected from hydrogen, halogen and hydroxyl; Z is selected from hydrogen, halogen, SQ, OQ and Q, where Q is an optionally substituted alkyl, aralkyl or aryl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; R¹ is a radical of the formula (IV)



A is selected from N, CH and CR, where R is selected from halogen or optionally substituted alkyl, aralkyl or aryl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; B is OH; D is selected from OH, NH₂, SCH₃ and hydrogen; E is N; G is CH₂; or a tautomer thereof or a pharmaceutically acceptable salt thereof; or an ester thereof.

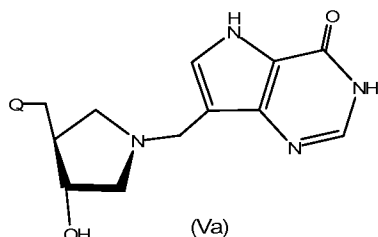
[0046] Preferably Y is H. Preferably Z is SQ. Preferably A is CH. Preferably D is H. Preferably Q is alkyl, preferably a C₁-C₆ alkyl group such as methyl, ethyl, propyl, butyl, pentyl or cyclohexyl or Q is an aryl group such as a phenyl group or an aralkyl group such as a benzyl group. It is also preferred that Q is an alkyl group substituted with one or more amino and one or more carboxy groups.

[0047] Preferred compounds include those having the formula (V):



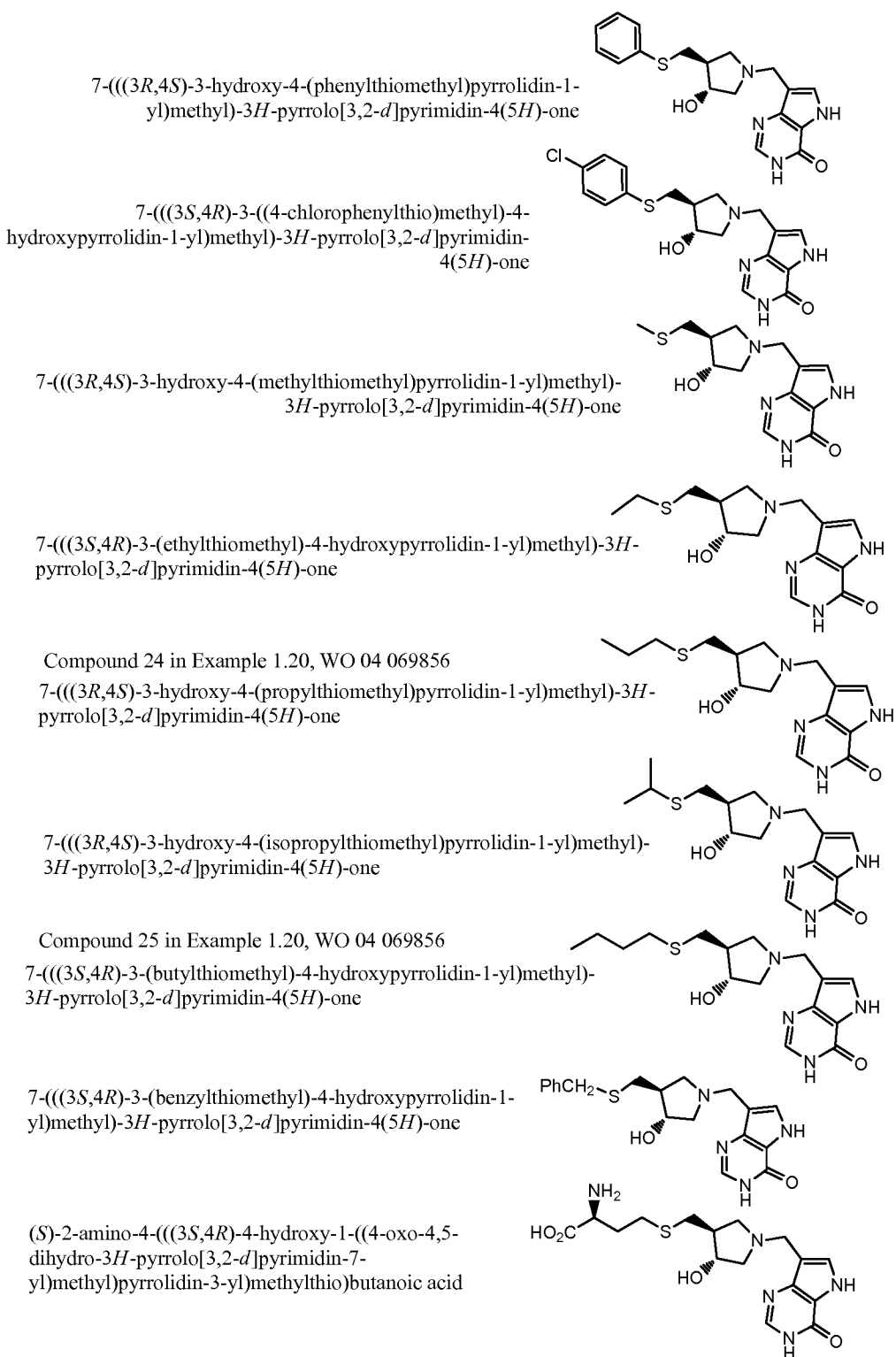
where Q is aryl, aralkyl or alkyl, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, and straight- or branched-chain C₁-C₆ alkyl; or a tautomer thereof; or an ester thereof; or a pharmaceutically acceptable salt thereof, or a prodrug thereof. Preferably, Q is optionally substituted alkyl, preferably a C₁-C₆ alkyl group such as methyl, ethyl, propyl or butyl, or Q is an optionally substituted aryl group such as a phenyl group or an optionally substituted aralkyl group such as a benzyl group. Preferred compounds include those where Q is methyl or phenyl. It is also preferred that Q is an alkyl group substituted with one or more amino groups. Preferably Q is an alkyl group substituted with one or more amino and one or more carboxy groups.

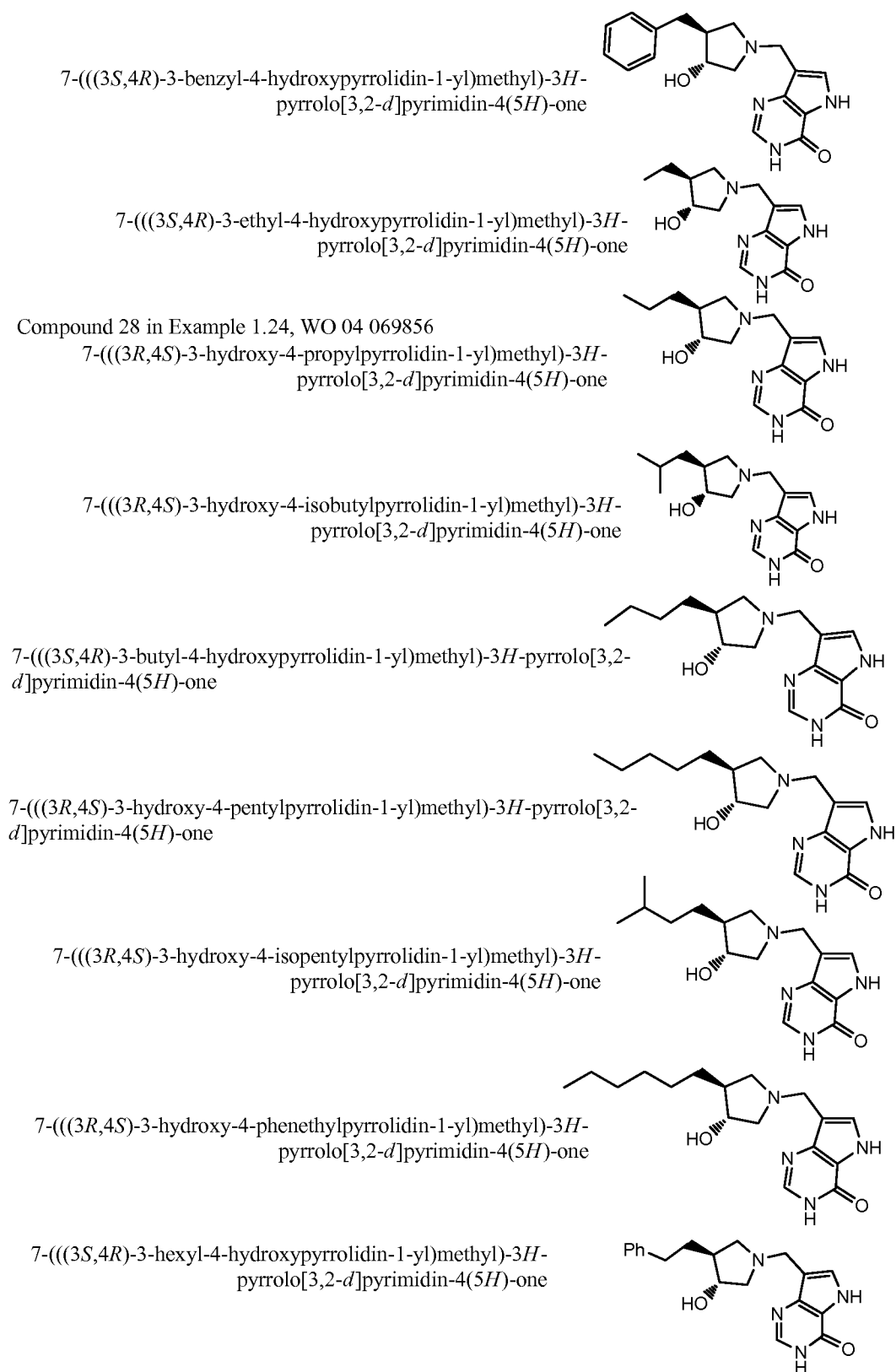
[0048] Other preferred compounds include those having the formula (Va):

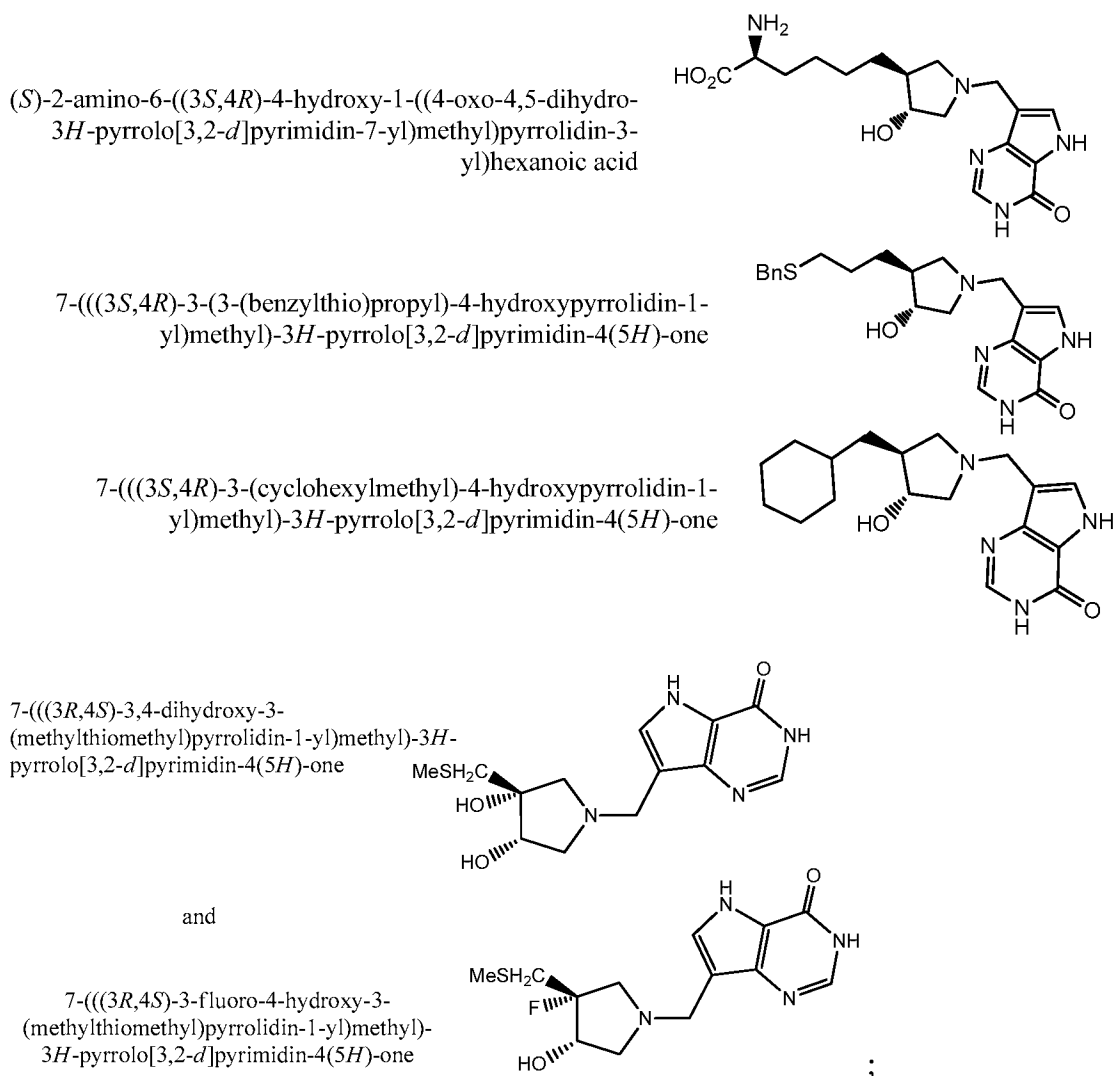


where Q is aryl, aralkyl or alkyl, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, and straight- or branched-chain C₁-C₆ alkyl; or a tautomer thereof; or an ester thereof; or a pharmaceutically acceptable salt thereof, or a prodrug thereof. Preferably, Q is optionally substituted alkyl, preferably a C₁-C₆ alkyl group such as methyl, ethyl, propyl or butyl, or Q is an optionally substituted aryl group such as a phenyl group or an aralkyl group such as a benzyl group. Preferred compounds include those where Q is methyl, ethyl or phenyl. It is also preferred that Q is an alkyl group substituted with one or more amino groups. Preferably Q is an alkyl group substituted with one or more amino and one or more carboxy groups.

[0049] Examples of compounds of formula (III) include:

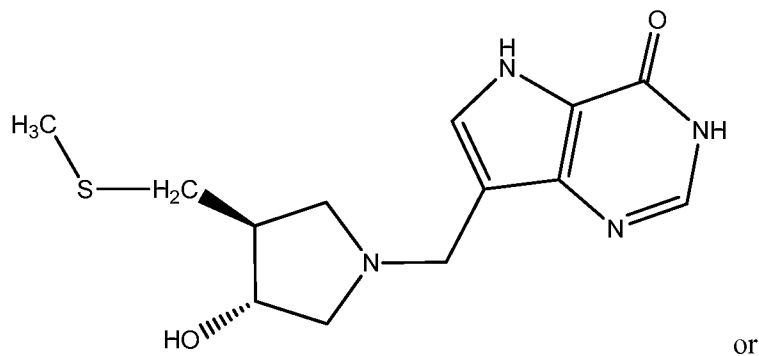


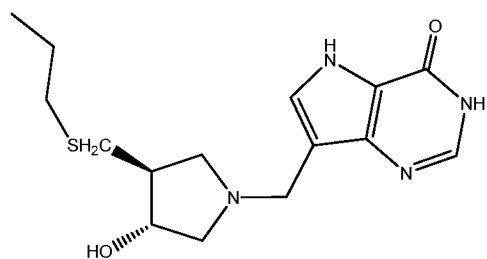




or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

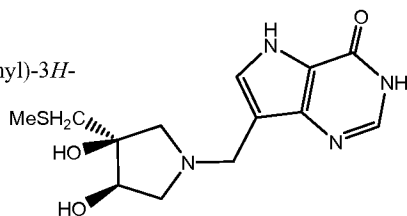
[0050] Preferred compounds include those having the structure:



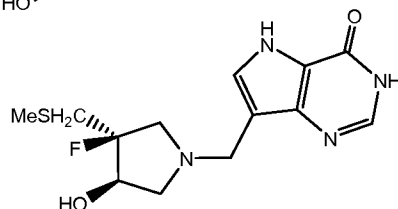


[0051] Examples of compounds of formula (IIIa) include:

7-(((3*S*,4*R*)-3,4-dihydroxy-3-(methylthiomethyl)pyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



7-(((3*S*,4*R*)-3-fluoro-4-hydroxy-3-(methylthiomethyl)pyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



7-(((3*S*,4*R*)-3-hydroxy-4-(phenylthiomethyl)pyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-(((3*R*,4*S*)-3-((4-chlorophenylthio)methyl)-4-hydroxypyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-(((3*S*,4*R*)-3-hydroxy-4-(methylthiomethyl)pyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-(((3*R*,4*S*)-3-(ethylthiomethyl)-4-hydroxypyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

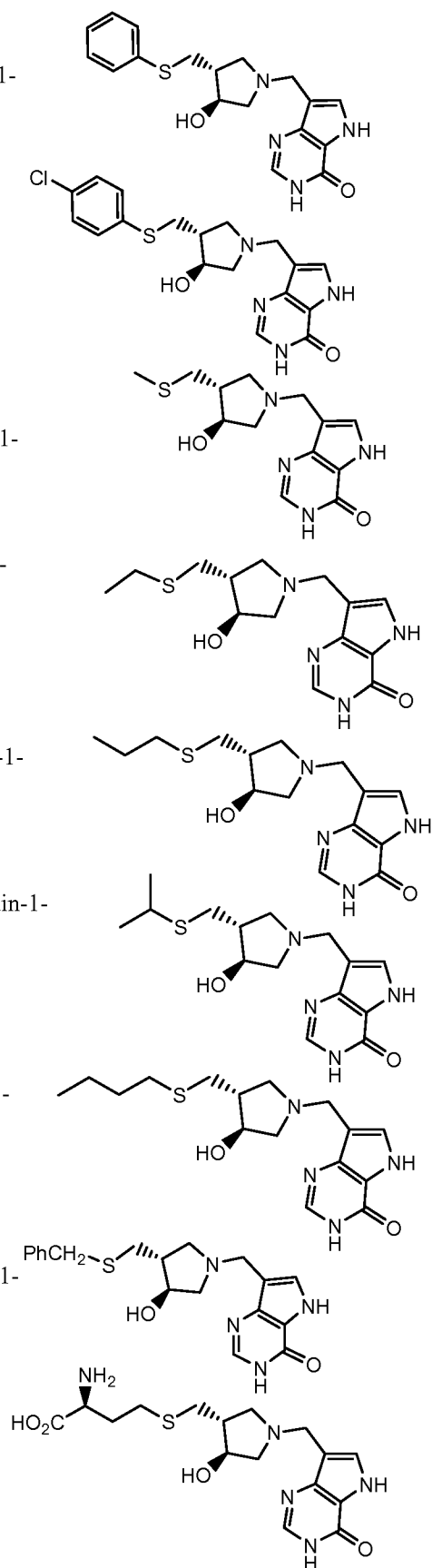
7-(((3*S*,4*R*)-3-hydroxy-4-(propylthiomethyl)pyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-(((3*S*,4*R*)-3-hydroxy-4-(isopropylthiomethyl)pyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-(((3*R*,4*S*)-3-(butylthiomethyl)-4-hydroxypyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-(((3*R*,4*S*)-3-(benzylthiomethyl)-4-hydroxypyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

(*S*)-2-amino-4-(((3*R*,4*S*)-4-hydroxy-1-((4-oxo-4,5-dihydro-3*H*-pyrrolo[3,2-*d*]pyrimidin-7-yl)methyl)pyrrolidin-3-yl)methylthio)butanoic acid



7-(((3*R*,4*S*)-3-benzyl-4-hydroxypyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-(((3*S*,4*R*)-3-hydroxy-4-propylpyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-(((3*R*,4*S*)-3-ethyl-4-hydroxypyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-(((3*S*,4*R*)-3-hydroxy-4-isobutylpyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

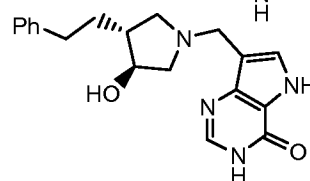
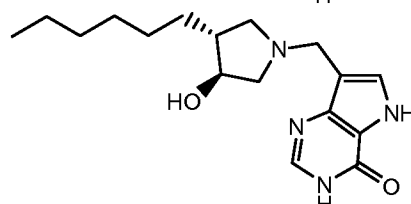
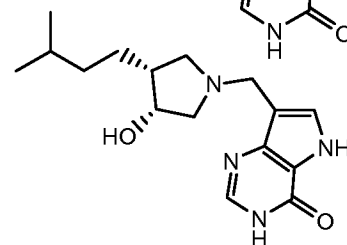
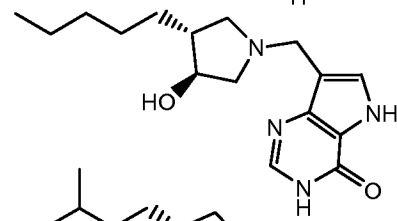
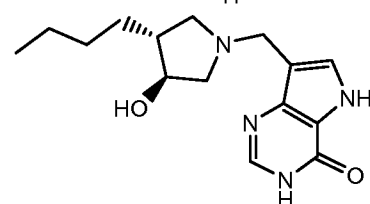
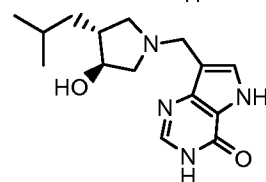
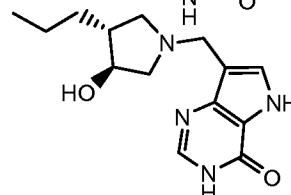
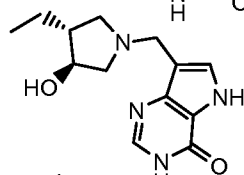
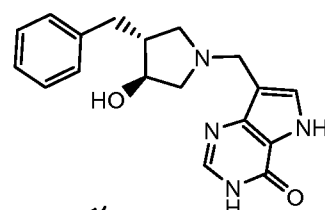
7-(((3*R*,4*S*)-3-butyl-4-hydroxypyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

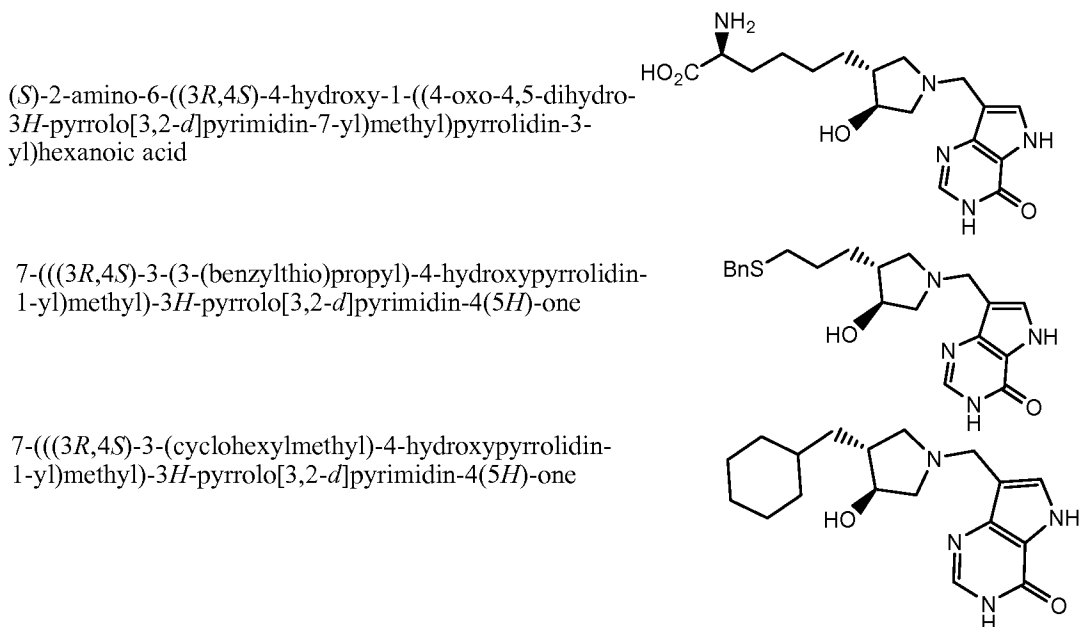
7-(((3*S*,4*R*)-3-hydroxy-4-pentylpyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-(((3*R*,4*R*)-3-hydroxy-4-isopentylpyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-(((3*R*,4*S*)-3-hexyl-4-hydroxypyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

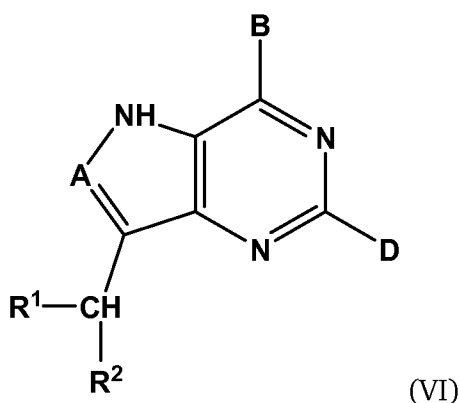
7-(((3*S*,4*R*)-3-hydroxy-4-phenethylpyrrolidin-1-yl)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one





or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

[0052] In another embodiment, as described in PCT International Patent Application Publication No. WO 2008/030119, the contents of which are herein incorporated by reference, the MTIP inhibitor comprises a compound having formula (VI):



wherein R^1 is H or NR^3R^4 ; R^2 is H or an alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group, each of which is optionally substituted with one or more hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, or NR^3R^4 groups, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; provided that when R^1 is H, R^2 is an alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group which is substituted with at least one NR^3R^4 group, and optionally substituted with one or more hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester or nitro group; R^3 and R^4 , independently of each other, are H or an

alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group, each of which is optionally substituted with one or more hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, or NR^3R^4 groups, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; A is N or CH; B is OH; and D is H, OH, NH_2 , or SCH_3 ; or a tautomer thereof, or a pharmaceutically acceptable salt thereof, or an ester prodrug form thereof.

[0053] Preferably, R^1 is NR^3R^4 and R^2 is H. It is further preferred that R^1 is NR^3R^4 and one of R^3 and R^4 is H, a lower alkyl group, e.g. a methyl group, or an aralkyl group, e.g. a benzyl group. Preferably R^1 is NR^3R^4 and one of R^3 and R^4 is H.

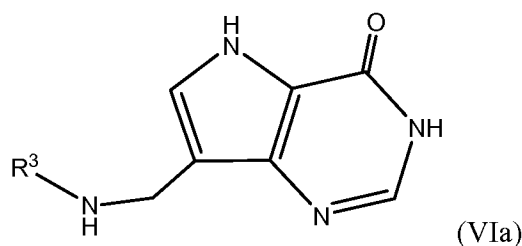
[0054] Preferably R^3 or R^4 is alkyl substituted by one or more hydroxy groups and/or one or more optionally substituted thiol, alkylthio, arylthio, or aralkylthio groups. For example, R^3 or R^4 may be, methylthioethyl, methylthiopropyl, methylthiohydroxypropyl, methylthiobutyl, methylthiohydroxybutyl, methylthiodihydroxybutyl, methylthiopentyl, methylthiohydroxypentyl, methylthiodihydroxypentyl, or methylthiotrihydroxypentyl.

[0055] Preferably R^1 is NR^3R^4 and one of R^3 and R^4 is H and the other is selected from the group consisting of methylthioethyl, methylthiopropyl, methylthiohydroxypropyl, methylthiobutyl, methylthiohydroxybutyl, methylthiodihydroxybutyl, methylthiopentyl, methylthiohydroxypentyl, methylthiodihydroxypentyl, and methylthiotrihydroxypentyl.

[0056] In addition, preferably R^2 is an optionally substituted alkyl group, more preferably an optionally substituted $\text{C}_1\text{-C}_5$ alkyl group, for example, hydroxymethyl, hydroxyethyl, hydroxypropyl, dihydroxypropyl, hydroxybutyl, dihydroxybutyl, trihydroxybutyl, hydroxypentyl, dihydroxypentyl, trihydroxypentyl, methylthiomethyl, methylthioethyl, methylthiopropyl, methylthiohydroxypropyl, methylthiobutyl, methylthiohydroxybutyl, methylthiodihydroxybutyl, methylthiopentyl, methylthiohydroxypentyl, methylthiodihydroxypentyl, or methylthiotrihydroxypentyl.

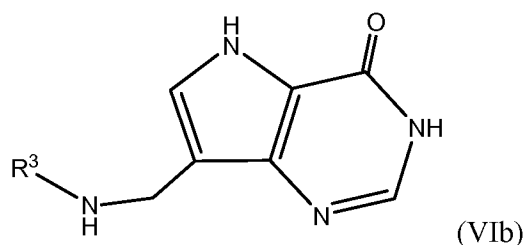
[0057] Preferably A is CH. It is further preferred that D is H.

[0058] Preferred compounds include those having the formula (VIa):



wherein R^3 is an alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group, each of which is optionally substituted with one or more hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, or nitro group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; or a tautomer thereof, or a pharmaceutically acceptable salt thereof, or an ester prodrug form thereof.

[0059] Other preferred compounds include those having the formula (VIb):



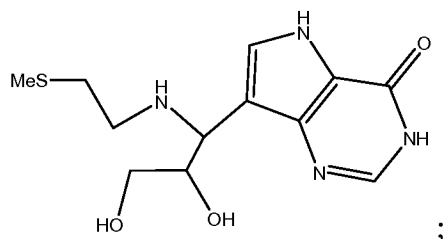
wherein R^3 is an alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group, each of which is substituted with at least one alkoxy, thiol, alkylthio, arylthio, or aralkylthio group and optionally substituted with one or more hydroxy, halogen, carboxylic acid, carboxylate alkyl ester, or nitro group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; or a tautomer thereof, or a pharmaceutically acceptable salt thereof, or an ester prodrug form thereof.

[0060] Preferably R^3 in the above formulae (VIa) and (VIb) is selected from the group consisting of, methylthioethyl, methylthiopropyl, methylthiohydroxypropyl, methylthiobutyl, methylthiohydroxybutyl, methylthiodihydroxybutyl, methylthiopentyl, methylthiohydroxypentyl, methylthiodihydroxypentyl, or methylthiotrihydroxypentyl.

[0061] Examples of compounds of formula (VI), as described in WO 2008/030119, include:

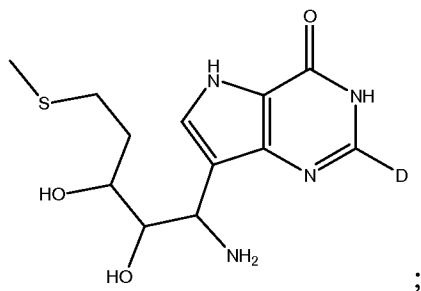
2-amino-7-(2,3-dihydroxy-1-(2-(methylthio)ethylamino)propyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(2,3-dihydroxy-1-(2-(methylthio)ethylamino)propyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



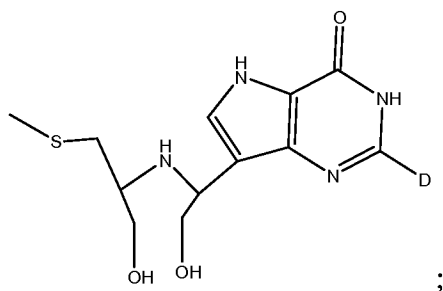
2-amino-7-(1-amino-2,3-dihydroxy-5-(methylthio)pentyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(1-amino-2,3-dihydroxy-5-(methylthio)pentyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



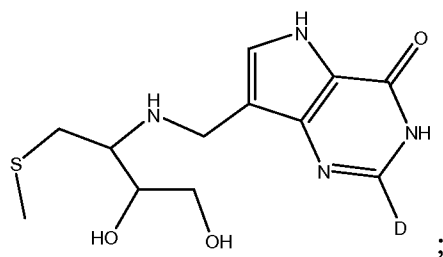
2-amino-7-(2-hydroxy-1-(1-hydroxy-3-(methylthio)propan-2-ylamino)ethyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(2-hydroxy-1-(1-hydroxy-3-(methylthio)propan-2-ylamino)ethyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



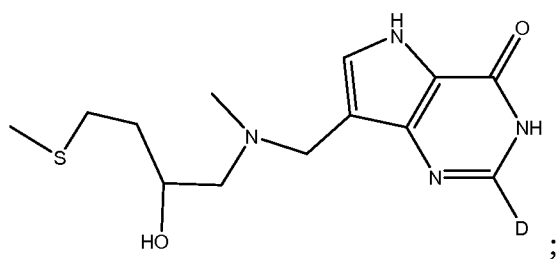
2-amino-7-((3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-((3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one (each enantiomer);



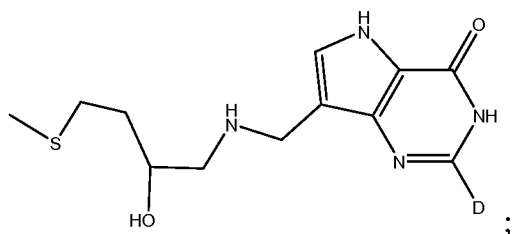
2-amino-7-(((2-hydroxy-4-(methylthio)butyl)(methyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(((2-hydroxy-4-(methylthio)butyl)(methyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



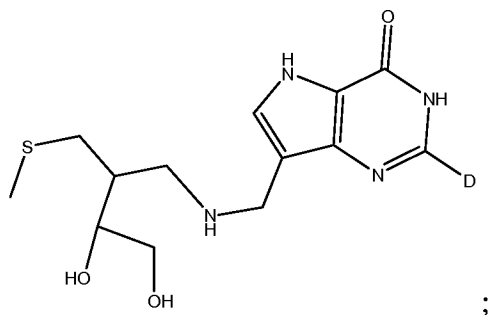
2-amino-7-((2-hydroxy-4-(methylthio)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-((2-hydroxy-4-(methylthio)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



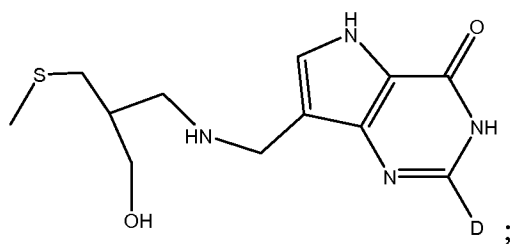
2-amino-7-((3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-((3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one (DL-erythro, DL-threo);



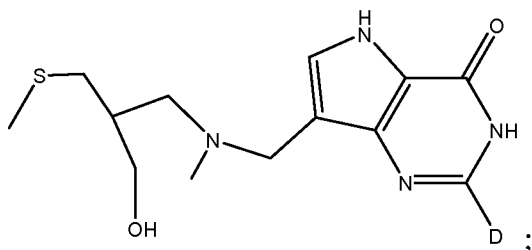
2-amino-7-((3-hydroxy-2-(methylthiomethyl)propylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-((3-hydroxy-2-(methylthiomethyl)propylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



7-(((3-hydroxy-2-(methylthiomethyl)propyl)(methyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

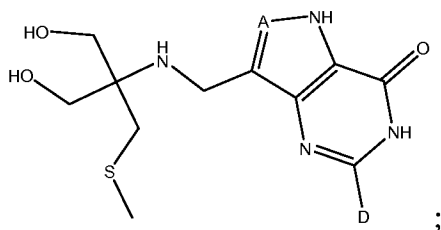
2-amino-7-(((3-hydroxy-2-(methylthiomethyl)propyl)(methyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



7-((1,3-dihydroxy-2-(methylthiomethyl)propan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-((1,3-dihydroxy-2-(methylthiomethyl)propan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

5-amino-3-((1,3-dihydroxy-2-(methylthiomethyl)propan-2-ylamino)methyl)-1H-pyrazolo[4,3-d]pyrimidin-7(6H)-one;



7-((benzyl((2R,3R)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-((benzyl((2S,3S)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-((benzyl((2R,3S)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-((benzyl((2S,3R)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

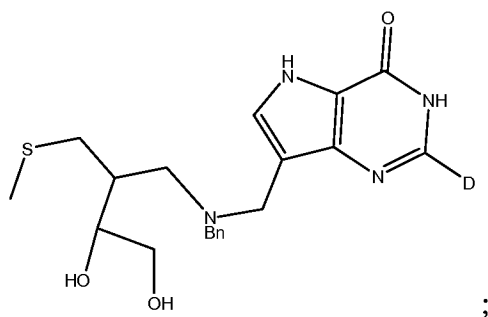
(Racemic mixture of above two as DL-erythro);

2-amino-7-((benzyl((2R,3R)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-((benzyl((2S,3S)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-((benzyl((2*R*,3*S*)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-((benzyl((2*S*,3*R*)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;



7-(((2*R*,3*R*)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

7-(((2*S*,3*S*)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

7-(((2*R*,3*S*)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

7-(((2*S*,3*R*)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

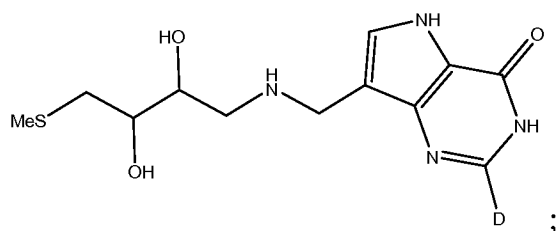
(Racemic mixture of above two as DL-threo);

2-amino-7-(((2*R*,3*R*)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-(((2*S*,3*S*)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-(((2*R*,3*S*)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-(((2*S*,3*R*)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;



7-(((2*R*,3*R*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

7-(((2*S*,3*S*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

7-(((2*R*,3*S*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

7-(((2*S*,3*R*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-(((2*R*,3*R*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-(((2*S*,3*S*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-(((2*R*,3*S*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

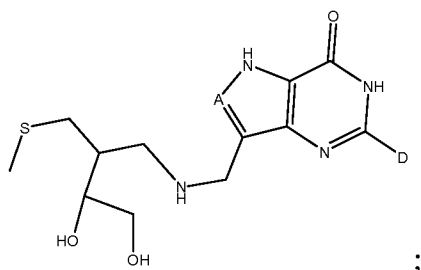
2-amino-7-(((2*S*,3*R*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

3-(((2*R*,3*R*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;

3-(((2*S*,3*S*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;

3-(((2*R*,3*S*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;

3-(((2*S*,3*R*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;



3-(((2*R*,3*R*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;

3-(((2*S*,3*S*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;

3-(((2R,3S)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-1H-pyrazolo[4,3-d]pyrimidin-7(6H)-one;

3-(((2S,3R)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-1H-pyrazolo[4,3-d]pyrimidin-7(6H)-one;

2-amino-7-(((2R,3R)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-(((2S,3S)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-(((2R,3S)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

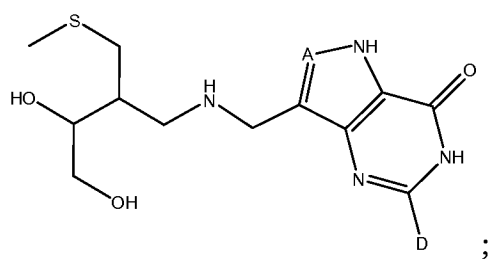
2-amino-7-(((2S,3R)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(((2R,3R)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

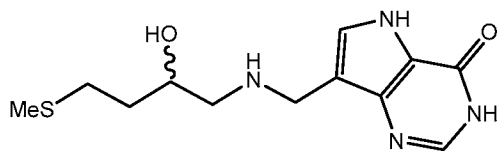
7-(((2S,3S)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(((2R,3S)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(((2S,3R)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



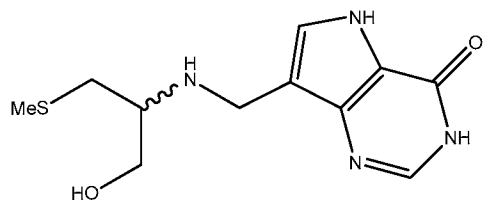
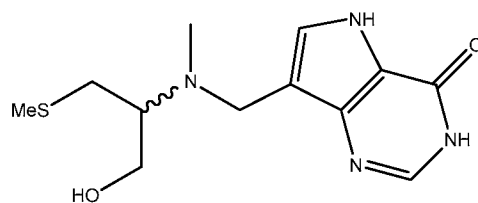
(*RS*)-7-((2-hydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



(*RS*)-7-(((1-hydroxy-3-(methylthio)propan-2-yl)(methyl)amino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

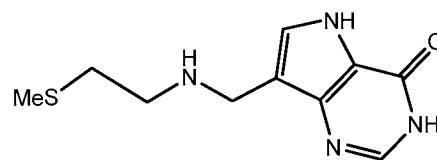
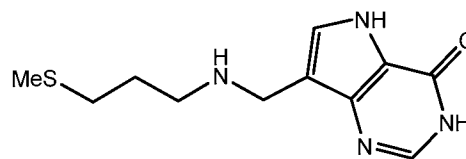
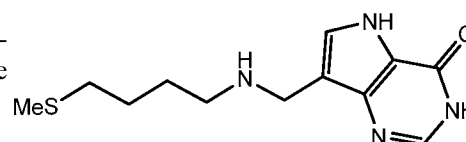
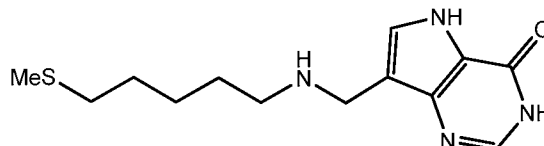
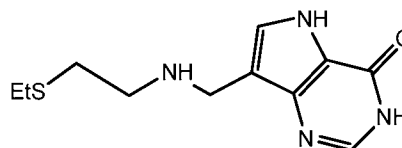
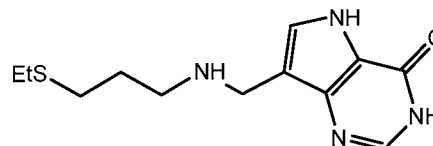
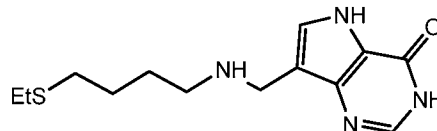
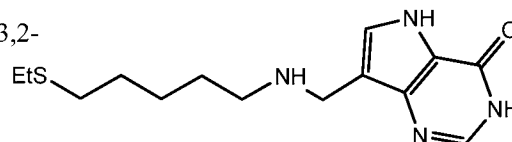
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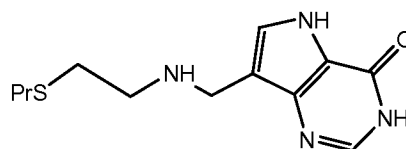
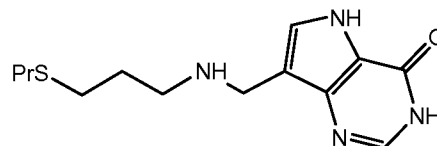
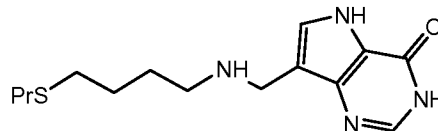
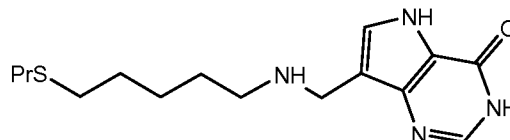
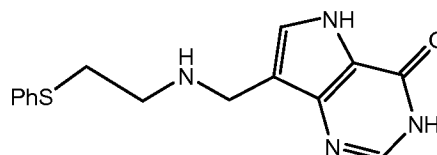
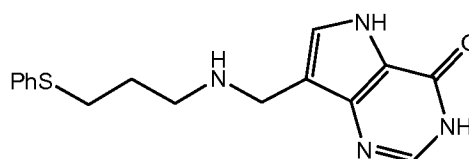
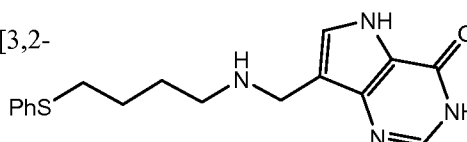
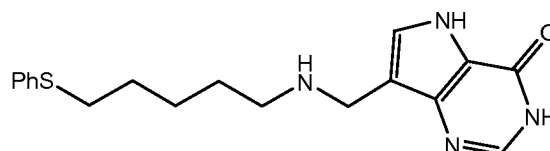
(*RS*)-7-(((1-hydroxy-3-(methylthio)propan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



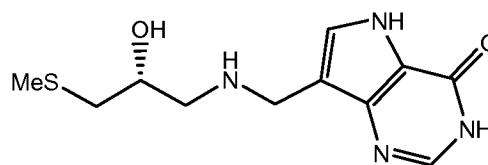
or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

[0062] Additional examples of compounds of formula (VI) include:

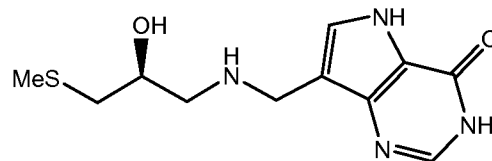
7-((2-(methylthio)ethylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((3-(methylthio)propylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((5-(methylthio)pentylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((2-(ethylthio)ethylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((3-(ethylthio)propylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((4-(ethylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((5-(ethylthio)pentylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-((2-(propylthio)ethylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((3-(propylthio)propylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((4-(propylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((5-(propylthio)pentylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((2-(phenylthio)ethylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((3-(phenylthio)propylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((4-(phenylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one7-((5-(phenylthio)pentylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

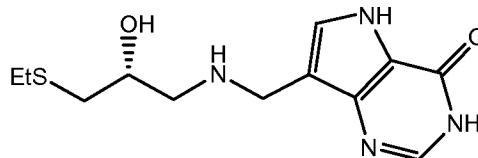
(*R*)-7-((2-hydroxy-3-(methylthio)propylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



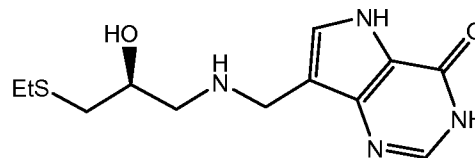
(*S*)-7-((2-hydroxy-3-(methylthio)propylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



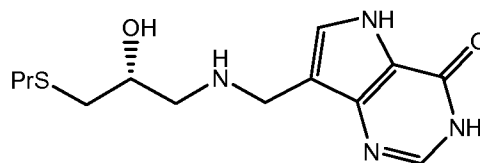
(*R*)-7-((3-(ethylthio)-2-hydroxypropylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



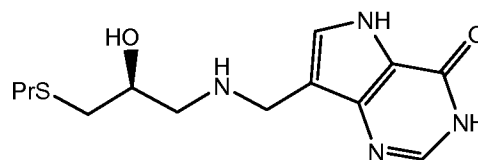
(*S*)-7-((3-(ethylthio)-2-hydroxypropylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



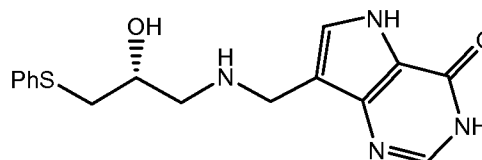
(*R*)-7-((2-hydroxy-3-(propylthio)propylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



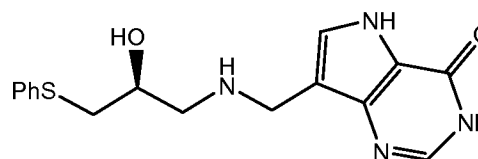
(*S*)-7-((2-hydroxy-3-(propylthio)propylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



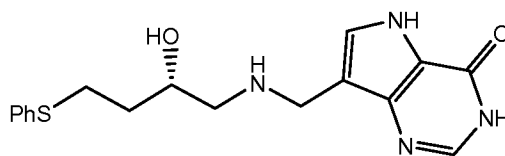
(*R*)-7-((2-hydroxy-3-(phenylthio)propylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



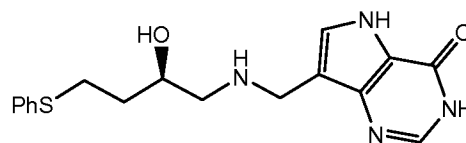
(*S*)-7-((2-hydroxy-3-(phenylthio)propylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



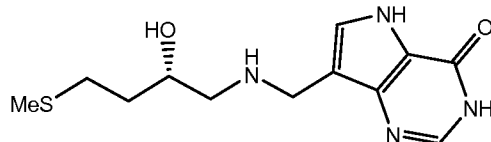
(*S*)-7-((2-hydroxy-4-(phenylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



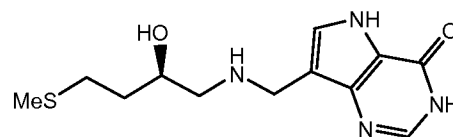
(*R*)-7-((2-hydroxy-4-(phenylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



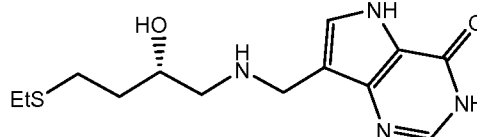
(*S*)-7-((2-hydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



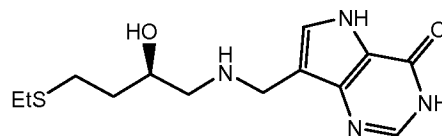
(*R*)-7-((2-hydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



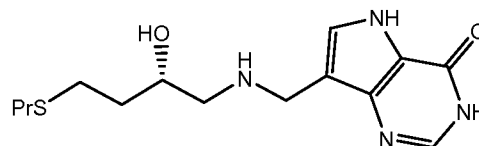
(*S*)-7-((4-(ethylthio)-2-hydroxybutylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



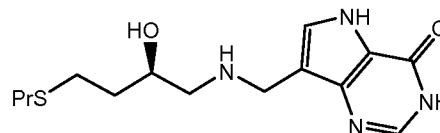
(*R*)-7-((4-(ethylthio)-2-hydroxybutylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

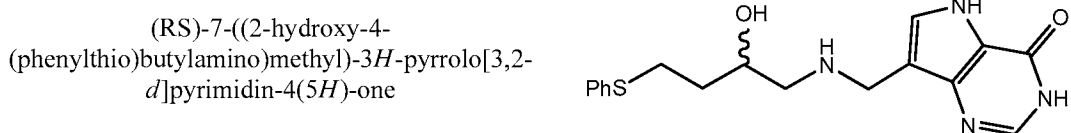
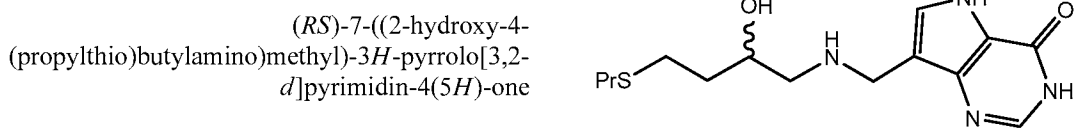
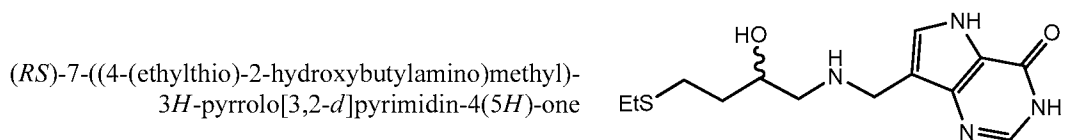
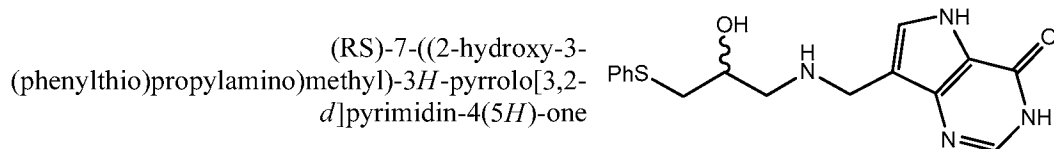
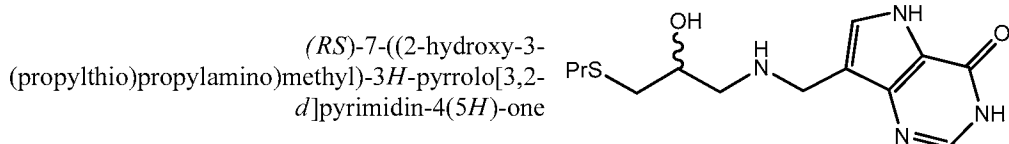
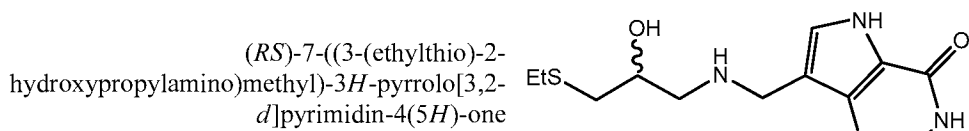
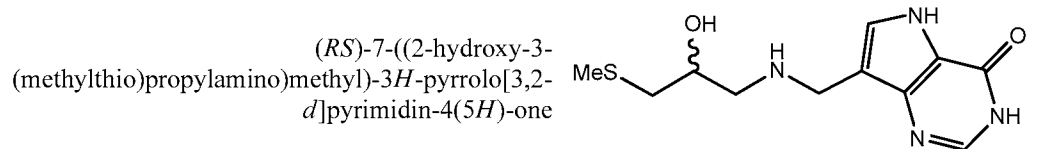


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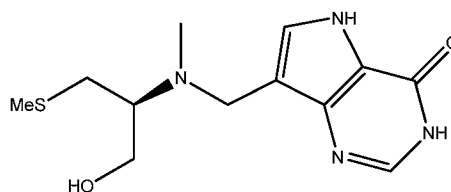


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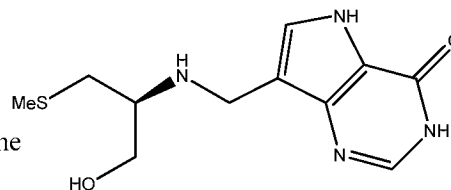




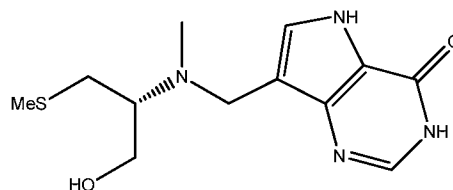
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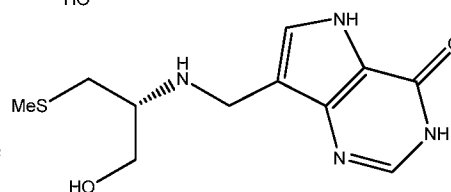
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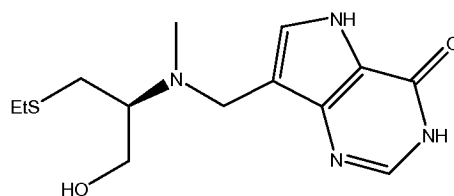
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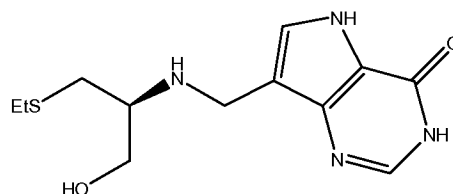
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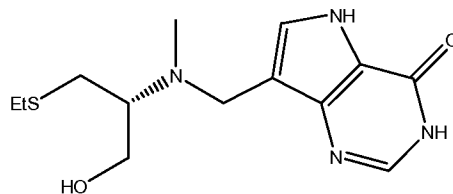
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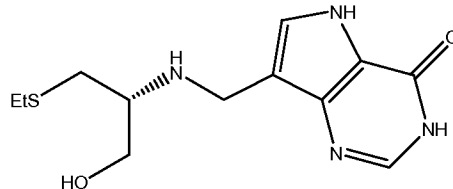
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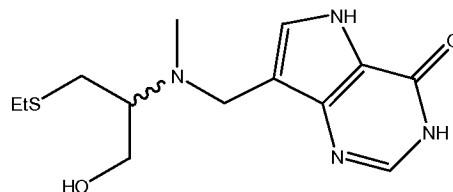
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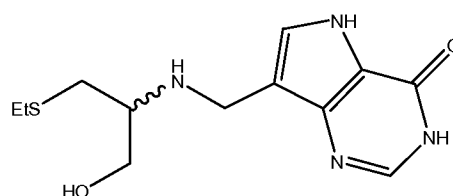
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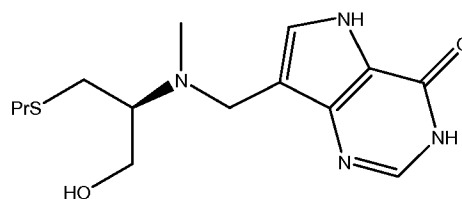
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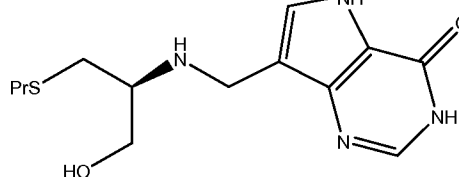
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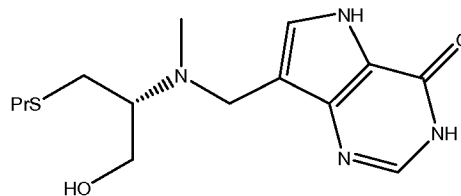
(*S*)-7-(((1-hydroxy-3-(propylthio)propan-2-yl)(methyl)amino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



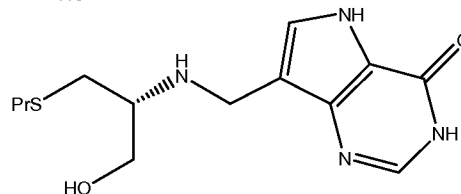
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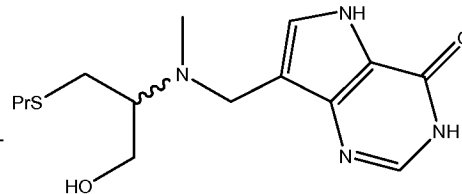
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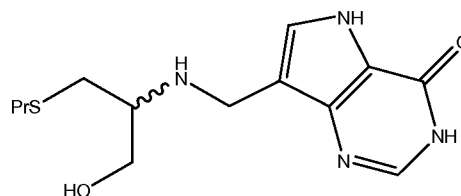
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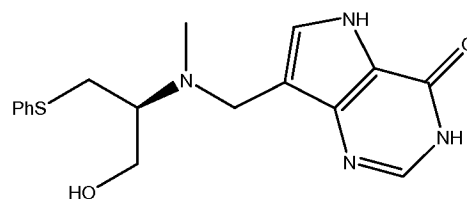
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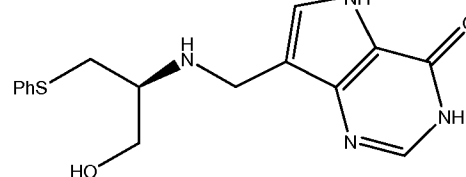
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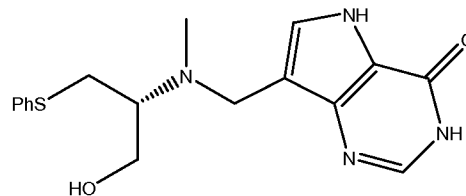
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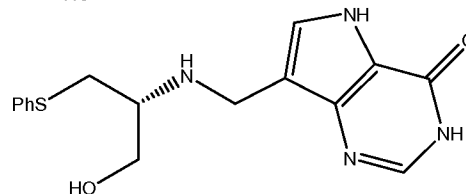
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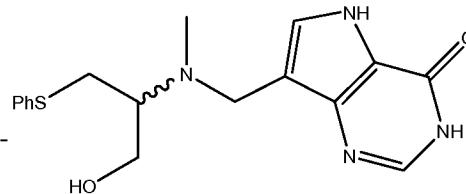
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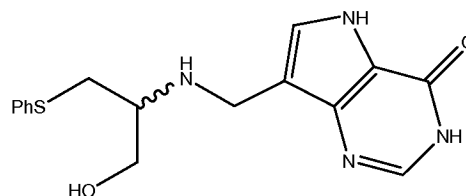
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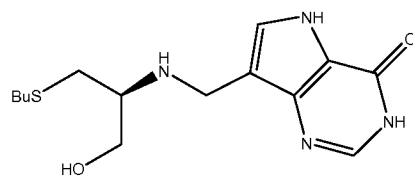
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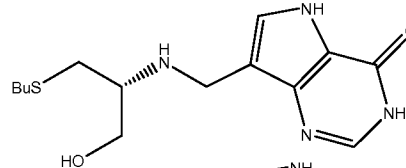
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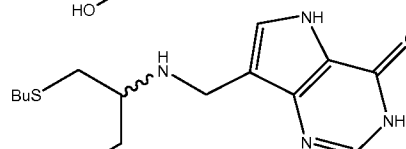
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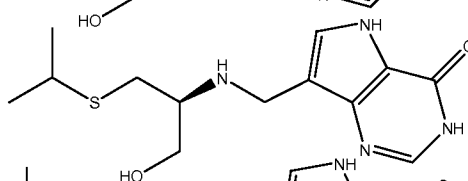
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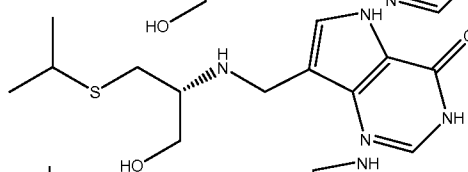
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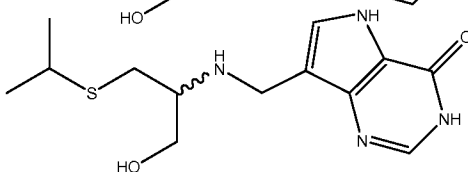
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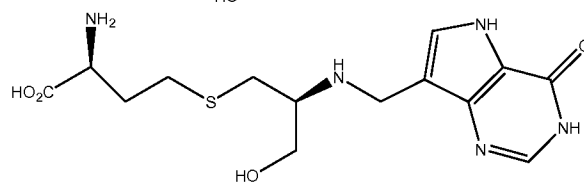
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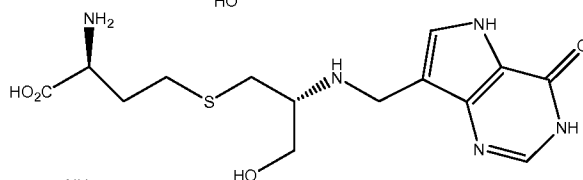
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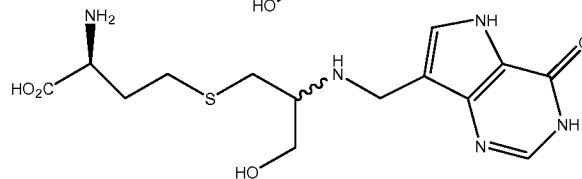
(*S*)-2-amino-4-((*S*)-3-hydroxy-2-((4-oxo-4,5-dihydro-3*H*-pyrrolo[3,2-*d*]pyrimidin-7-yl)methylamino)propylthio)butanoic acid



(*S*)-2-amino-4-((*R*)-3-hydroxy-2-((4-oxo-4,5-dihydro-3*H*-pyrrolo[3,2-*d*]pyrimidin-7-yl)methylamino)propylthio)butanoic acid



(*S*)-2-amino-4-((*RS*)-3-hydroxy-2-((4-oxo-4,5-dihydro-3*H*-pyrrolo[3,2-*d*]pyrimidin-7-yl)methylamino)propylthio)butanoic acid



(*S*)-7-((1-hydroxy-4-phenylbutan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

(*S*)-7-((1-hydroxybutan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

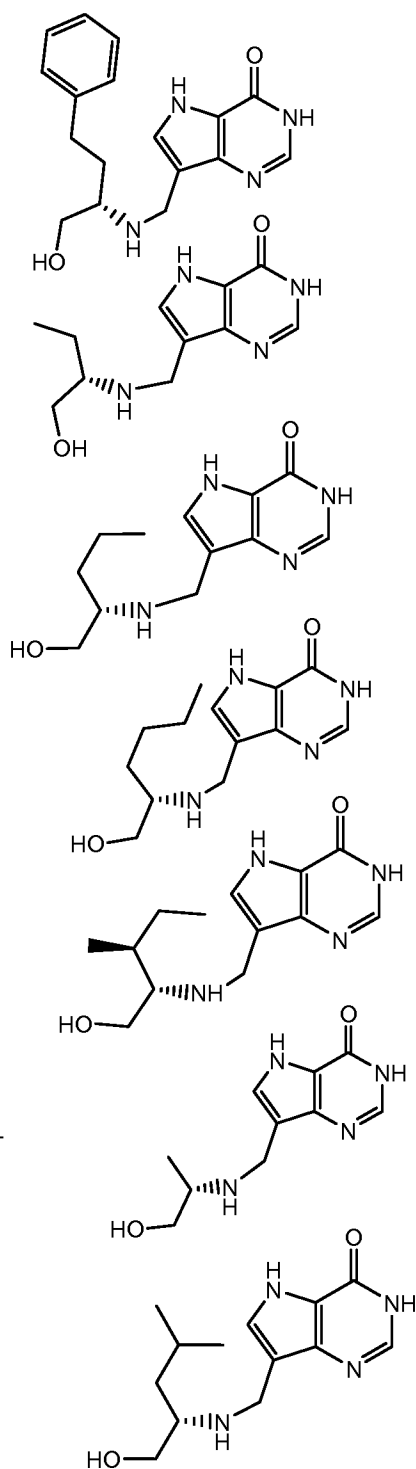
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(*S*)-7-((1-hydroxyhexan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

7-(((2*S*,3*S*)-1-hydroxy-3-methylpentan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

(*S*)-7-((1-hydroxypropan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

(*S*)-7-((1-hydroxy-4-methylpentan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one



(*R*)-7-((1-hydroxy-4-phenylbutan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

(*R*)-7-((1-hydroxybutan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

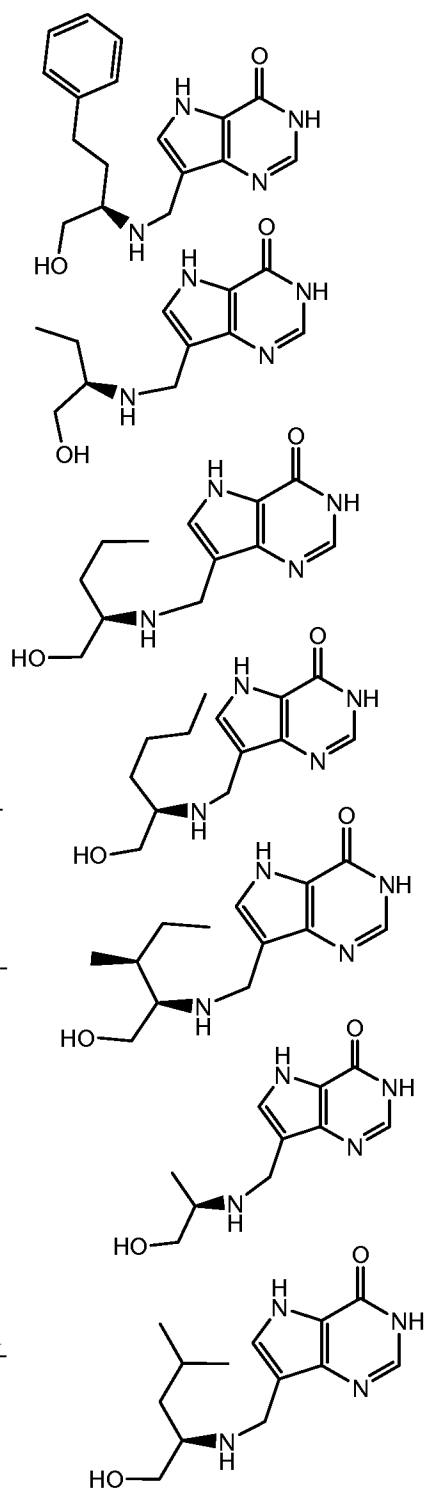
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(*R*)-7-((1-hydroxyhexan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

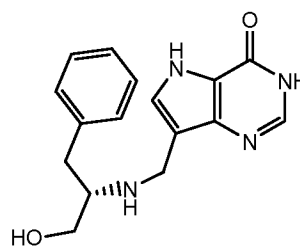
7-(((2*R*,3*S*)-1-hydroxy-3-methylpentan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

(*R*)-7-((1-hydroxypropan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one

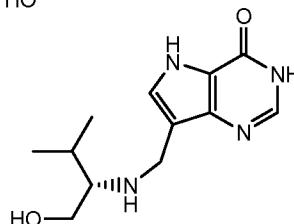
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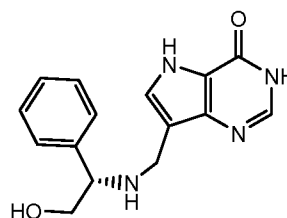
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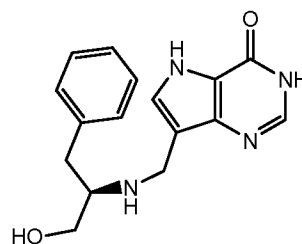
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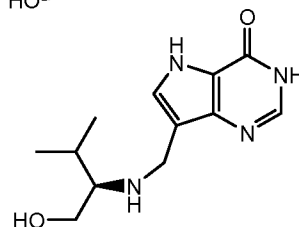
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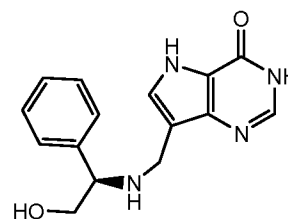
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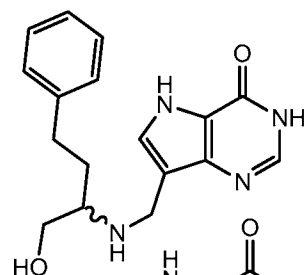
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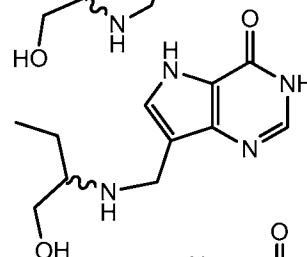
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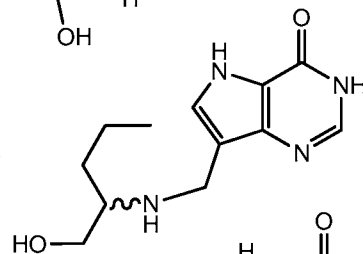
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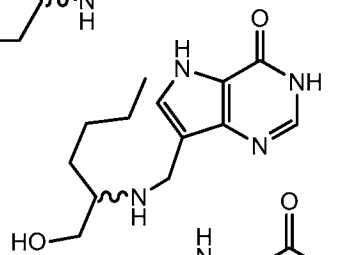
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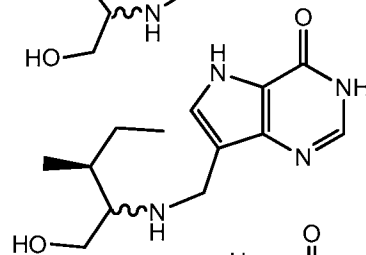
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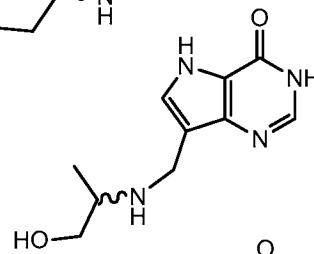
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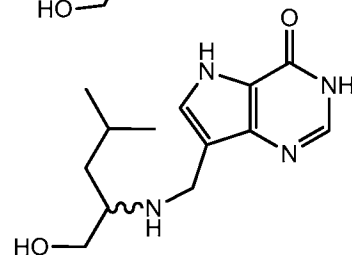
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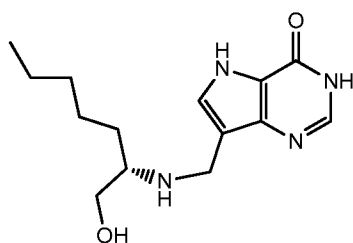


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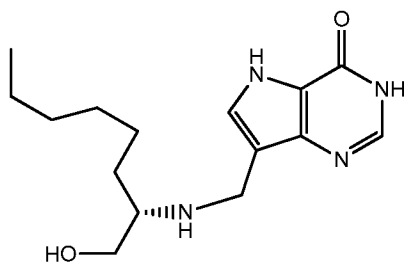
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(*S*)-7-((1-hydroxyheptan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one ,

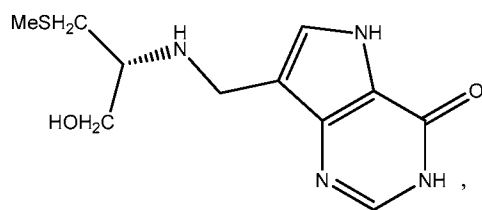
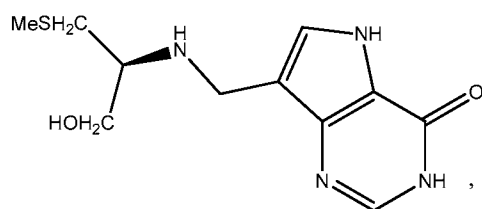
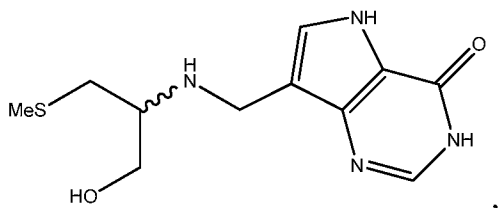
and

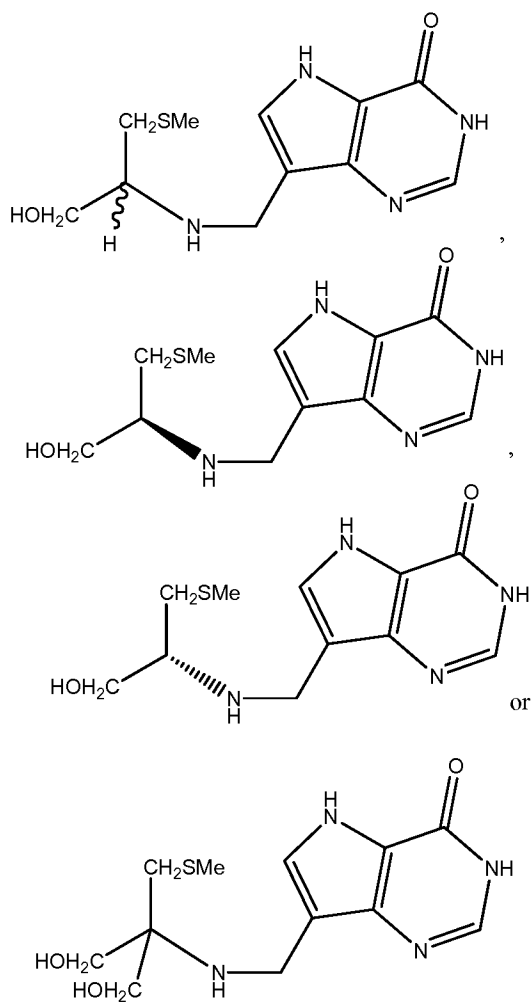


(*S*)-7-((1-hydroxyoctan-2-ylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one ;

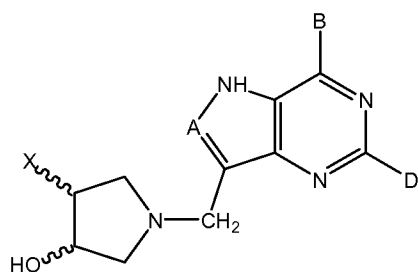
or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

[0063] Preferred compounds include those having the structure:





[0064] In another embodiment, the MTIP inhibitor comprises a compound having formula (VII):

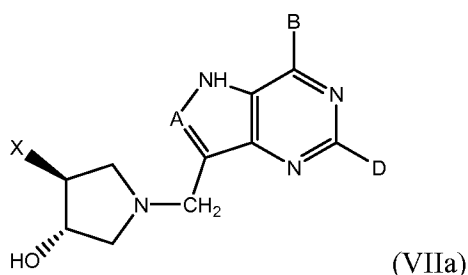


(VII)

wherein X is an alkyl, cycloalkyl, aralkyl, aralkenyl, alkenyl, alkynyl or aryl group, each of which is optionally substituted with one or more substituents selected from the group consisting of hydroxy, alkoxy, cycloalkyl, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, cyano, thiazole and NR^2R^3 group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; or X is SR^1 ; or X is NR^2R^3 ; R^1 , R^2 and R^3 are

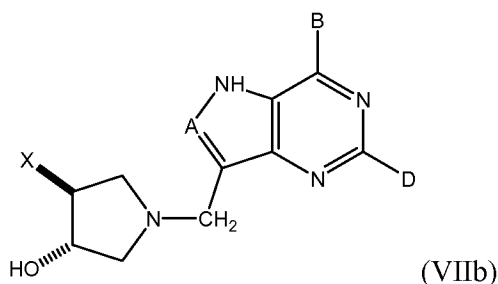
independently selected from the group consisting of alkyl, alkenyl, alkynyl, aralkyl or aryl, each of which is optionally substituted with one or more substituents selected from the group consisting of hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, cyano and NR^2R^3 group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; A is N or CH; B is OH; D is H, OH, NH_2 , or SCH_3 ; provided that X is not CH_2Z , where Z is selected from OH, hydrogen, halogen, SQ^1 , OQ^2 and Q^3 , where Q^1 is an optionally substituted alkyl, aralkyl or aryl group, Q^2 is an optionally substituted alkyl group and Q^3 is an optionally substituted alkyl group; or a tautomer thereof, or a pharmaceutically acceptable salt thereof, or an ester prodrug form thereof.

[0065] Preferred compounds include those of formula (VIIa):



wherein A, B, D and X are as defined above.

[0066] Alternatively, preferably the compound is a compound of formula (VIIb):



wherein A, B, D and X are as defined above.

[0067] Preferably, in the above formulae (VII), (VIIa) and (VIIb), Q^1 is an optionally substituted alkyl, aralkyl or aryl group, Q^2 is an optionally substituted alkyl group and Q^3 is an optionally substituted alkyl group. For example, Q^1 , Q^2 or Q^3 may be optionally substituted with one or more: halogens, e.g. chlorine or fluorine; alkyl groups, e.g. methyl or cyclohexylmethyl; COOH ; or NH_2 .

[0068] Preferably A is CH. It is further preferred that D is H.

[0069] Preferably, X is an alkenyl or alkynyl group each of which is optionally substituted with one or more substituents selected from the group consisting of hydroxy,

alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, or NR^2R^3 group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups. X may be, for example, a lower alkenyl group, e.g. a vinyl, allyl or prop-1-en-2-yl group. X may be, for example, a lower alkynyl group, e.g. an ethynyl group or a propyn-3-yl group.

[0070] Alternatively, preferably, X is an alkyl group which is optionally substituted with one or more substituents selected from the group consisting of hydroxy, alkoxy, thiol, alkylthio, arylthio, or aralkylthio, e.g. benzylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro or NR^2R^3 group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy or alkoxy groups. X may be, for example, a lower alkyl group, e.g. ethyl, propyl, butyl, isobutyl, pent-3-yl or an alkyl group substituted with a cycloalkyl group, e.g. cyclohexyl group, e.g. X may be cyclohexanemethyl. Alternatively, X may be, for example, an alkyl group which is substituted with an aralkylthio group, e.g. X may be 3-benzylthiopropyl group.

[0071] Alternatively, preferably, X is an alkyl group which is optionally substituted with one or more substituents selected from the group consisting of cycloalkyl, e.g. cycloalkyl in which one or more of the ring carbon atoms is substituted by a heteroatom chosen from nitrogen, oxygen or sulfur. In some examples, X may be cyclopropanemethyl, 2-tetrahydrofuranmethyl, 2-thietanemethyl, 3-piperidinemethyl, 2-pyrrolidinemethyl or 4-thiacyclohexanemethyl.

[0072] Alternatively, preferably, X is a cycloalkyl group which is optionally substituted with one or more substituents selected from the group consisting of hydroxy, alkoxy, cycloalkyl, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, cyano, thiazole and NR^2R^3 group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy or alkoxy groups. X may be, for example, a cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl or adamantyl group.

[0073] Alternatively, preferably, X may be a cycloalkyl group where one or more of the ring atoms is a heteroatom, e.g. a nitrogen, sulfur or oxygen atom. X may be, for example, 2-tetrahydrofuranyl, 2-tetrahydrothienyl, 1,2-dithian-3-yl, piperidin-3-yl, thietan-2-yl, 2-pyrrolidinyl or 4-thiacyclohexyl.

[0074] Alternatively, preferably, X is an aryl group which is optionally substituted with one or more substituents selected from the group consisting of hydroxy, alkoxy,

cycloalkyl, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, cyano, thiazole and NR^2R^3 group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups. The aryl group may be a heteroaryl group, where one or more of the ring carbon atoms is a heteroatom, e.g. a nitrogen, sulfur or oxygen atom. X may be, for example, a phenyl group or an optionally substituted triazole group. Where X is an optionally substituted triazole group the triazole ring may optionally be substituted with one or more substituents selected from the group consisting of aryl group, e.g. phenyl; alkyl group, e.g. a lower alkyl group, e.g. a propyl group which may optionally be substituted with one or more substituents selected from aryl, hydroxyl, or alkoxy; aralkyl group, e.g. benzyl; or cycloalkyl group. Where X is an optionally substituted triazole group, the triazole ring may be attached to the pyrrolidine ring via either a triazole ring nitrogen or a triazole ring carbon atom.

[0075] Alternatively, preferably, X is SR^1 , where R^1 is alkyl, alkenyl, alkynyl, aralkyl or aryl, each of which is optionally substituted with one or more substituents selected from the group consisting of hydroxy, alkoxy, cycloalkyl, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, cyano, thiazole and NR^2R^3 group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy or alkoxy groups. For example, X may be phenylthio, 4-chlorophenylthio, 4-fluorophenylthio, 3-fluorophenylthio, 4-methylphenylthio, ethylthio, propylthio, butylthio, pentylthio, 3-fluoropropylthio, 2,3-dihydroxypropylthio, 3-hydroxypropylthio, 2-hydroxyethylthio, allylthio or 4-chlorobutylthio.

[0076] Alternatively, preferably, X is NR^2R^3 , where R^2 and R^3 are independently selected from alkyl, alkenyl, alkynyl, aralkyl or aryl, each of which is optionally substituted with one or more substituents selected from the group consisting of hydroxy, alkoxy, cycloalkyl, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, cyano, thiazole and NR^2R^3 group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups. For example, X may be diethylamino, ethylamino, propylamino, butylamino, 2-hydroxyethylamino, 3-hydroxypropylamino, 2,3-dihydroxypropylamino, 3-fluoroethylamino, trifluoroethylamino, bis(2-hydroxyethyl)amino, 3-butenylamino, benzylamino, 4-fluorobenzylamino, 4-chlorobenzylamino, or N-methyl-benzylamino.

[0077] Examples of compounds of formula (VII) include:
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-phenylpyrrolidine;

(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-vinylpyrrolidine;
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-4-ethynyl-3-hydroxypyrrolidine;
(±)-*trans*-4-Allyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-4-Cyclopropyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-4-Cyclohexyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-4-Cyclohexylmethyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(prop-1-en-2-yl)-pyrrolidine;
(±)-*trans*-4-Butyl-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(pent-3-yl)-pyrrolidine;
(±)-*trans*-4-Cyclopentyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(1*H*-1,2,3-triazol-1-yl)pyrrolidine;
(±)-*trans*-4-(1-Benzyl-1*H*-1,2,3-triazol-4-yl)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-4-(3-Benzylthiopropyl)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*cis*-1-[(9-Deazahypoxanthin-9-yl)methyl]-4-ethyl-3-hydroxypyrrolidine;
(3*R*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-phenylpyrrolidine;
(3*R*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-vinylpyrrolidine;
(3*R*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-4-ethynyl-3-hydroxy-pyrrolidine;
(3*R*,4*S*)-4-Allyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(3*R*,4*S*)-4-Cyclopropyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(3*R*,4*S*)-4-Cyclohexyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(3*R*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(prop-1-en-2-yl)-pyrrolidine;
(3*R*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(pent-3-yl)-pyrrolidine;
(3*R*,4*S*)-4-Cyclopentyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(3*S*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(1*H*-1,2,3-triazol-1-yl)pyrrolidine;
(3*R*,4*R*)-4-(1-Benzyl-1*H*-1,2,3-triazol-4-yl)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(2-methylpropyl)pyrrolidine;
(±)-*trans*-4-Butyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(thiazol-2-yl)-pyrrolidine;
(3*R*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(thiazol-2-yl)-pyrrolidine;

(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(tetrazol-5-yl)-pyrrolidine; (3R,4R)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(tetrazol-5-yl)-pyrrolidine; and (3S,4R)-1-[(9-Deaza-adenin-9-yl)methyl]-4-ethyl-3-hydroxypyrrolidine; or a tautomer thereof, or a pharmaceutically acceptable salt thereof, or an ester thereof.

[0078] Preferably, the MTIP inhibitor has an IC₅₀ value greater than or equal to 50 nanomolar for human purine nucleoside phosphorylase (PNP), as determined by the method described in Bantia, *et al.*, Immunopharmacology 35, p. 54, paragraph 2.1 (1996) (52).

Preferably, the MTIP inhibitor has an IC₅₀ value greater than or equal to 50 nanomolar for 5'-methylthioadenosine nucleosidase (MTAN). Preferably, the MTIP inhibitor has an IC₅₀ value greater than or equal to 50 nanomolar for 5'-methylthioadenosine phosphorylase (MTAP). "IC₅₀" means the molar concentration of an inhibitor needed to reduce the rate of product formation by 50% from the uninhibited reaction in an assay mixture containing a target enzyme.

[0079] The active compounds may be administered to a subject, such as a human, by a variety of routes, including orally, parenterally, by inhalation spray, topically, rectally, nasally, buccally or via an implanted reservoir. The specific dosage required for any particular patient will depend upon a variety of factors, including the patient's age and body weight.

[0080] For oral administration the compounds can be formulated into solid or liquid preparations, for example tablets, capsules, powders, solutions, suspensions and dispersions. Such preparations are well known in the art as are other oral dosage regimes not listed here. In the tablet form the compounds may be tableted with conventional tablet bases such as lactose, sucrose and corn starch, together with a binder, a disintegration agent and a lubricant. The binder may be, for example, corn starch or gelatin, the disintegrating agent may be potato starch or alginic acid, and the lubricant may be magnesium stearate. For oral administration in the form of capsules, diluents such as lactose and dried cornstarch may be employed. Other components such as colourings, sweeteners or flavourings may be added.

[0081] When aqueous suspensions are required for oral use, the active ingredient may be combined with carriers such as water and ethanol, and emulsifying agents, suspending agents and/or surfactants may be used. Colourings, sweeteners or flavourings may also be added.

[0082] The compounds may also be administered by injection in a physiologically acceptable diluent such as water or saline. The diluent may comprise one or more other

ingredients such as ethanol, propylene glycol, an oil or a pharmaceutically acceptable surfactant.

[0083] The compounds may also be administered topically. Carriers for topical administration of the compounds of include mineral oil, liquid petrolatum, white petrolatum, propylene glycol, polyoxyethylene, polyoxypropylene compound, emulsifying wax and water. The compounds may be present as ingredients in lotions or creams, for topical administration to skin or mucous membranes. Such creams may contain the active compounds suspended or dissolved in one or more pharmaceutically acceptable carriers. Suitable carriers include mineral oil, sorbitan monostearate, polysorbate 60, cetyl ester wax, cetearyl alcohol, 2-octyldodecanol, benzyl alcohol and water.

[0084] The compounds may further be administered by means of sustained release systems. For example, they may be incorporated into a slowly dissolving tablet or capsule.

[0085] The compounds can be administered to plants, for example, by spraying the plant surfaces with the compound and a carrier of the type normally used in agricultural applications, such as, for example, water or an oil. Typical concentrations can be, for example, 1 - 100 grams of inhibitor per 1000 gallons of plant spray.

[0086] The present invention also provides for the use of a subgrowth inhibiting amount of an MTIP inhibitor for treating bacterial infections in a subject. The present invention further provides for the use of a subgrowth inhibiting amount of an MTIP inhibitor for the preparation of a composition for treating bacterial infections in a subject.

[0087] The invention also provides a method for determining whether or not a compound is a candidate for treating an infection caused by bacteria that use 5'-methylthioinosine phosphorylase (MTIP) in a quorum sensing pathway, the method comprising determining whether or not the compound inhibits MTIP, wherein a compound that inhibits MTIP is a candidate for treating an infection caused by bacteria that use MTIP in a quorum sensing pathway and wherein a compound that does not inhibit MTIP is not a candidate for treating an infection caused by bacteria that use MTIP in a quorum sensing pathway.

[0088] Preferably, the MTIP inhibitor has an IC₅₀ value less than 50 nanomolar for MTIP. Preferably, the MTIP inhibitor has an IC₅₀ value greater than or equal to 50 nanomolar for human purine nucleoside phosphorylase (PNP), as determined by the method described in Bantia, *et al.*, Immunopharmacology 35, p. 54, paragraph 2.1 (1996) (52). Preferably, the MTIP inhibitor has an IC₅₀ value greater than or equal to 50 nanomolar for

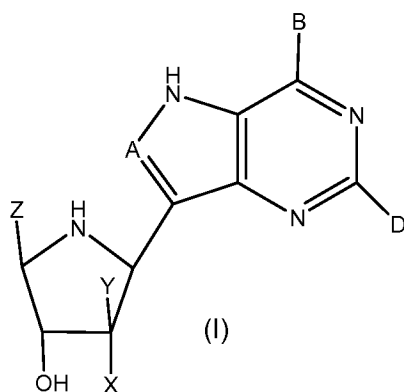
5'-methylthioadenosine nucleosidase (MTAN). Preferably, the MTIP inhibitor has an IC50 value greater than or equal to 50 nanomolar for 5'-methylthioadenosine phosphorylase (MTAP). "IC50" means the molar concentration of an inhibitor needed to reduce the rate of product formation by 50% from the uninhibited reaction in an assay mixture containing a target enzyme.

[0089] An "inhibitor" is a substance that when added to an assay mixture for a target enzyme causes a decrease in the rate of product formation by interaction with the enzyme.

[0090] The bacteria can be, for example, *Pseudomonas aeruginosa*, *Pseudomonas syringae* or *Xanthomonas campestris*.

[0091] Enzymatic assays can be carried out, for example, as described herein in Experimental Details.

[0092] The invention also provides a chemical compound of formula (I)



wherein A is CH or N; B is OH; D is chosen from H, OH, NH₂, or SCH₃; X is OH; Y is H; and Z is Q, where Q is a methyl group which is substituted with one or more substituents selected from the group consisting of methoxy, amino and carboxy, or Q is an optionally substituted, alkenyl, aralkyl, aralkenyl, aryl group or C₂-C₁₀ alkyl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

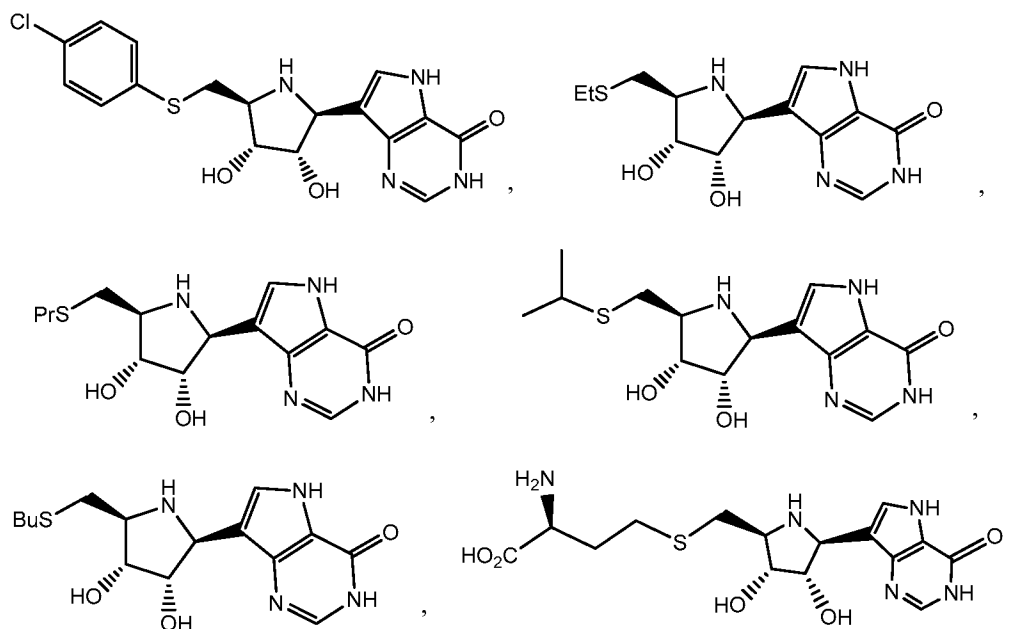
[0093] Preferably, D is H. It is also preferred that A is CH. It is further preferred that D is H and A is CH.

[0094] Preferably, Q is an optionally substituted alkyl group, e.g. a C₂-C₆ alkyl group such as an ethyl, propyl or butyl group. It is also preferred that Q is an alkyl group substituted with one or more amino groups. Preferably Q is an alkyl group substituted with one or more amino and one or more carboxy groups, e.g. -(CH₂)₄CH(NH₂)COOH.

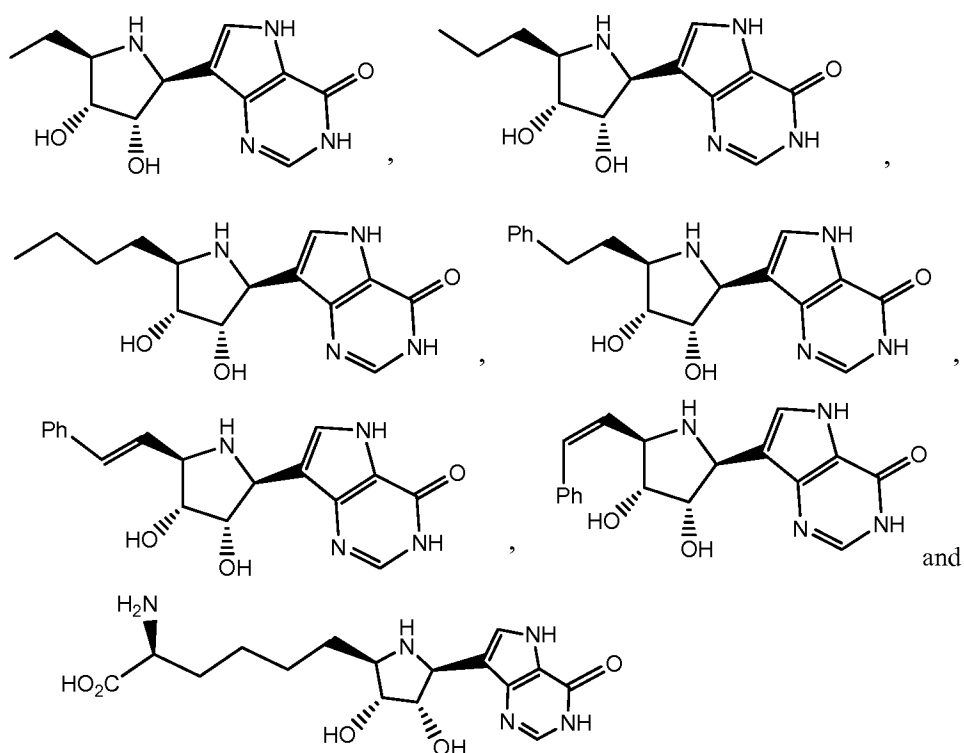
[0095] Alternatively, preferably, Q is an optionally substituted aralkyl group, e.g. a phenylethyl or benzyl group. In still other preferred embodiments, Q is an optionally substituted aralkenyl group, e.g. a phenylethenyl group, e.g. a *cis*-phenylethenyl group or a *trans*-phenylethenyl group.

[0096] Preferably, the compound has an IC₅₀ value greater than or equal to 50 nanomolar for human purine nucleoside phosphorylase (PNP), as determined by the method described in Bantia, *et al.*, Immunopharmacology 35, p. 54, paragraph 2.1 (1996) (52). Preferably, the compound has an IC₅₀ value greater than or equal to 50 nanomolar for 5'-methylthioadenosine nucleosidase (MTAN). Preferably, the compound has an IC₅₀ value greater than or equal to 50 nanomolar for 5'-methylthioadenosine phosphorylase (MTAP).

[0097] The invention also provides a compound selected from the group consisting of

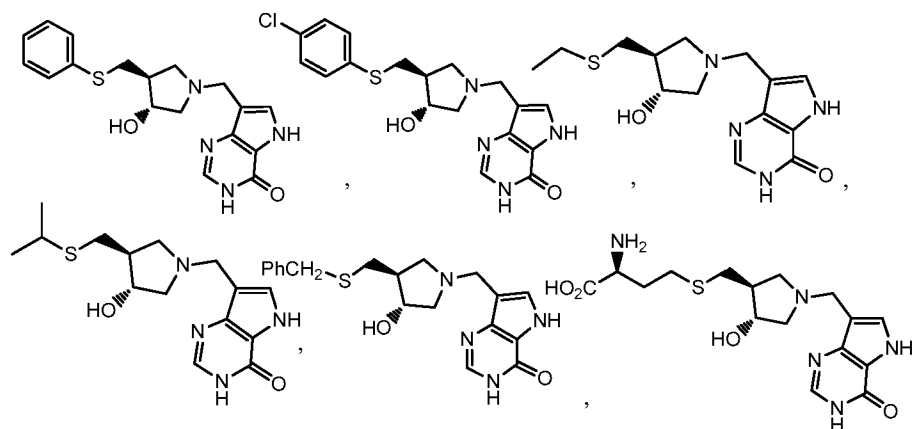


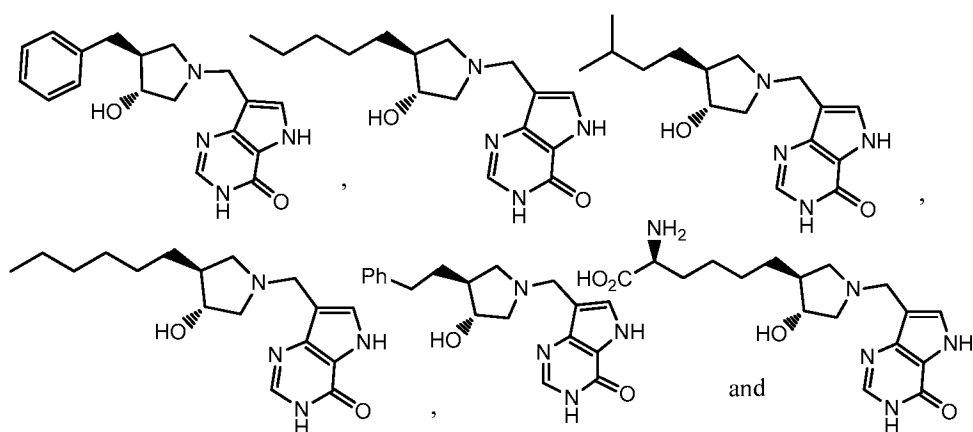
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or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

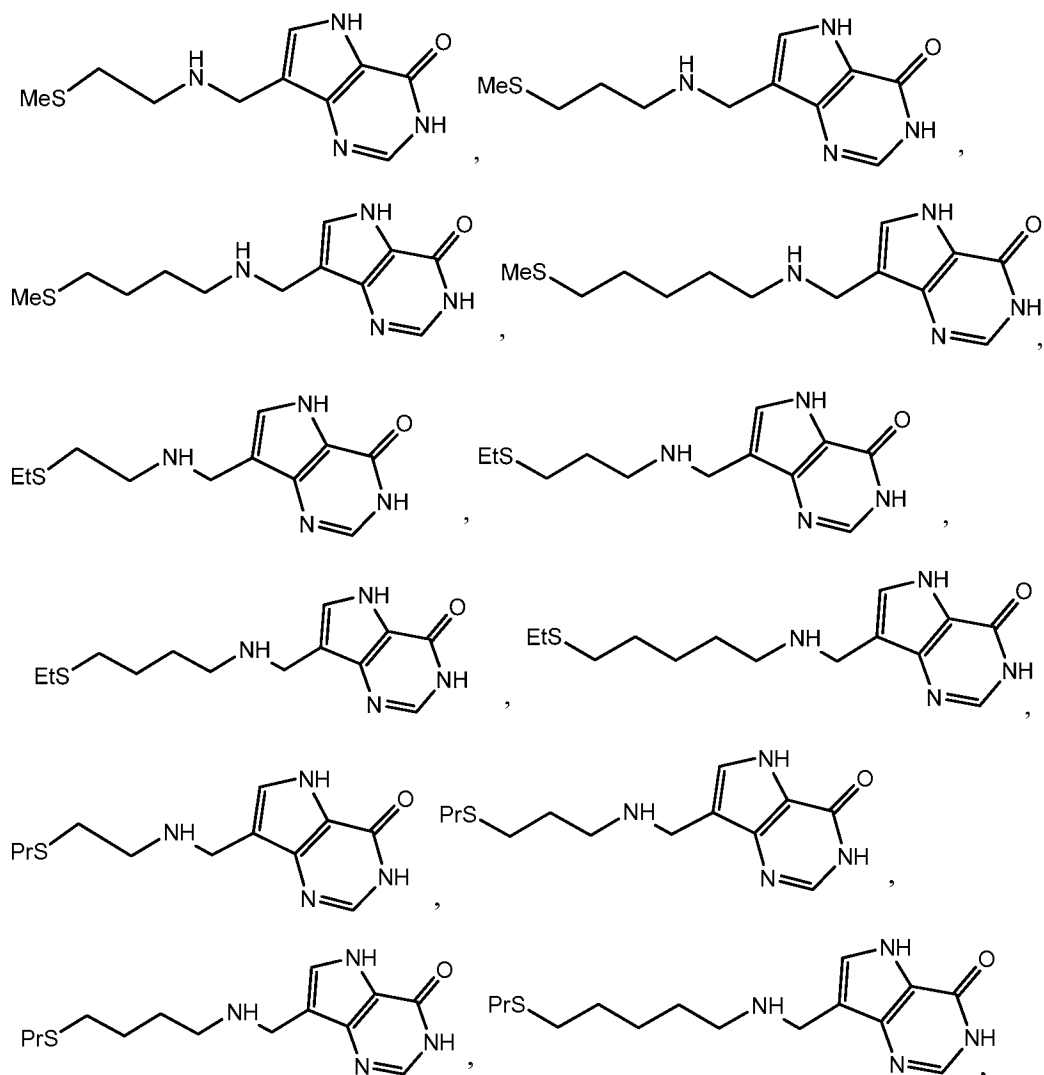
[0098] The invention further provides a compound selected from the group consisting of

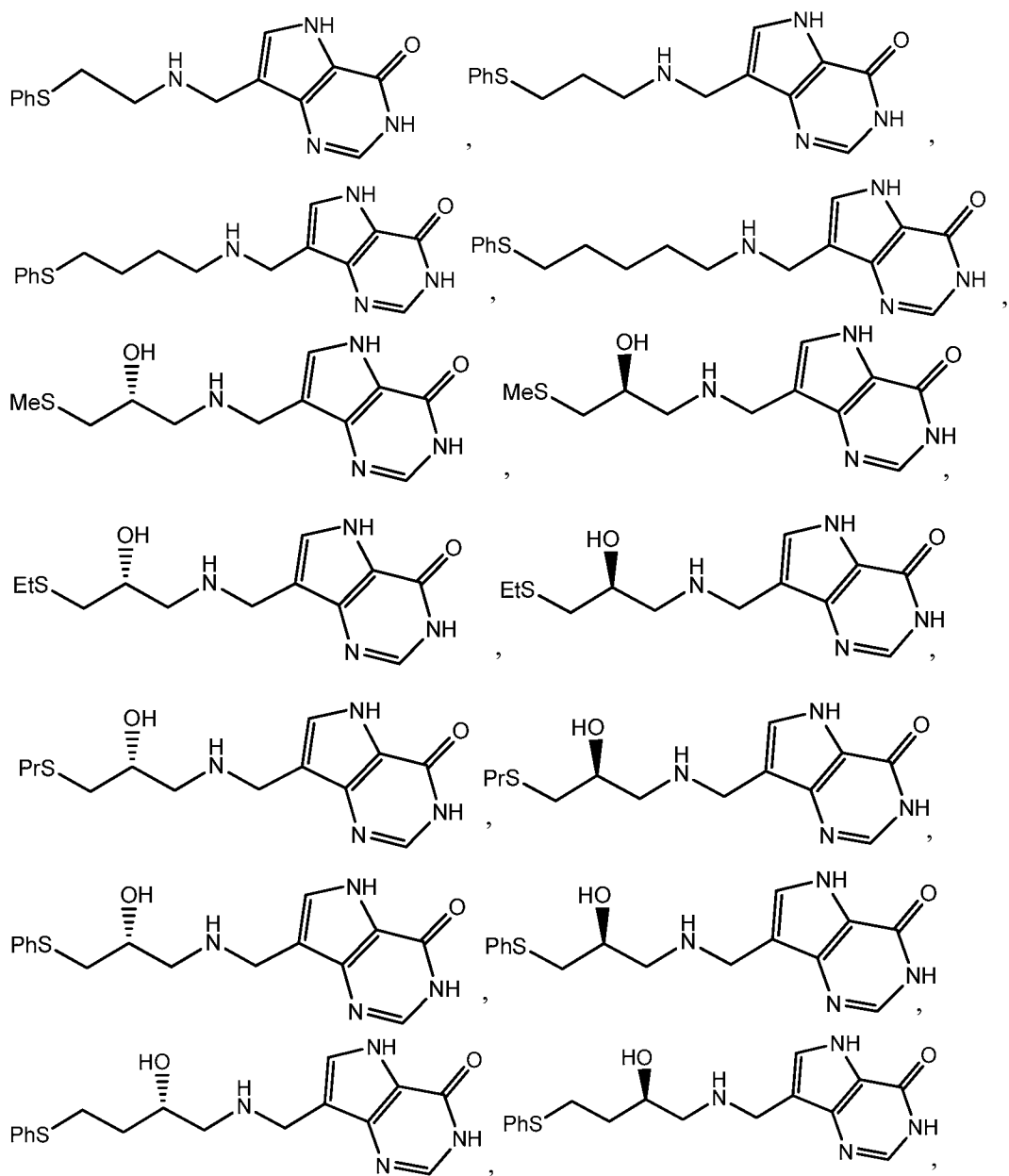


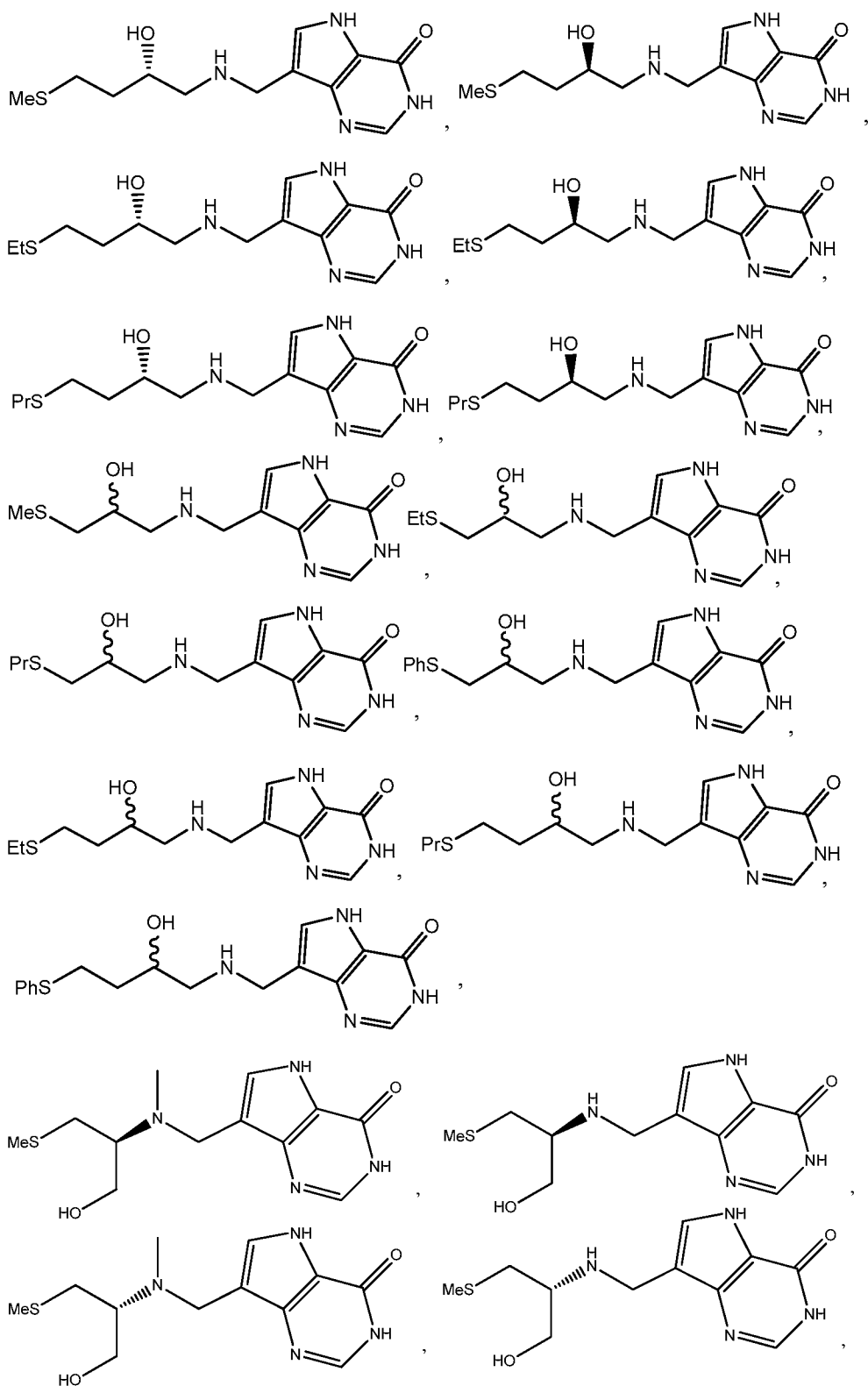


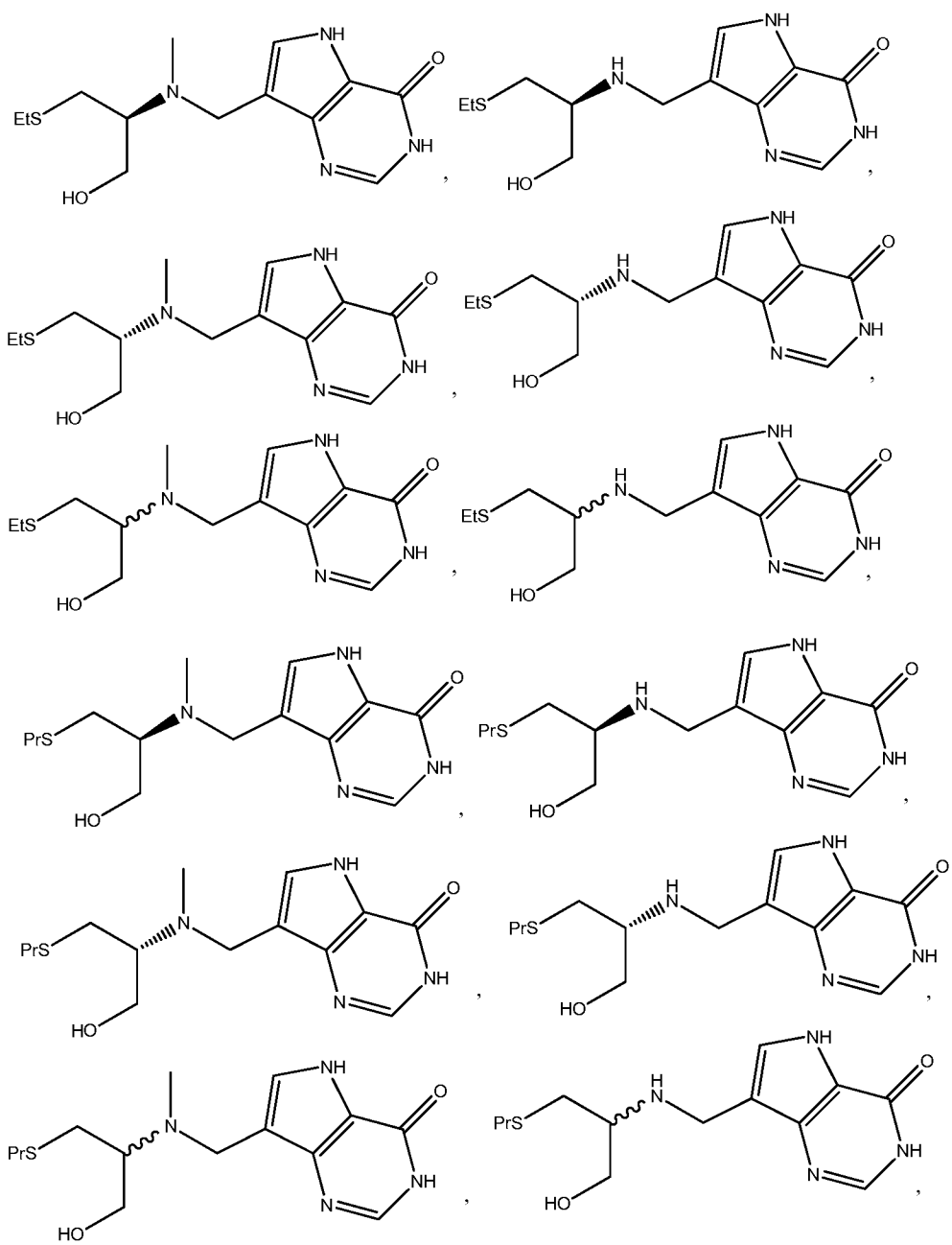
or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

[0099] The invention still further provides a compound selected from the group consisting of:

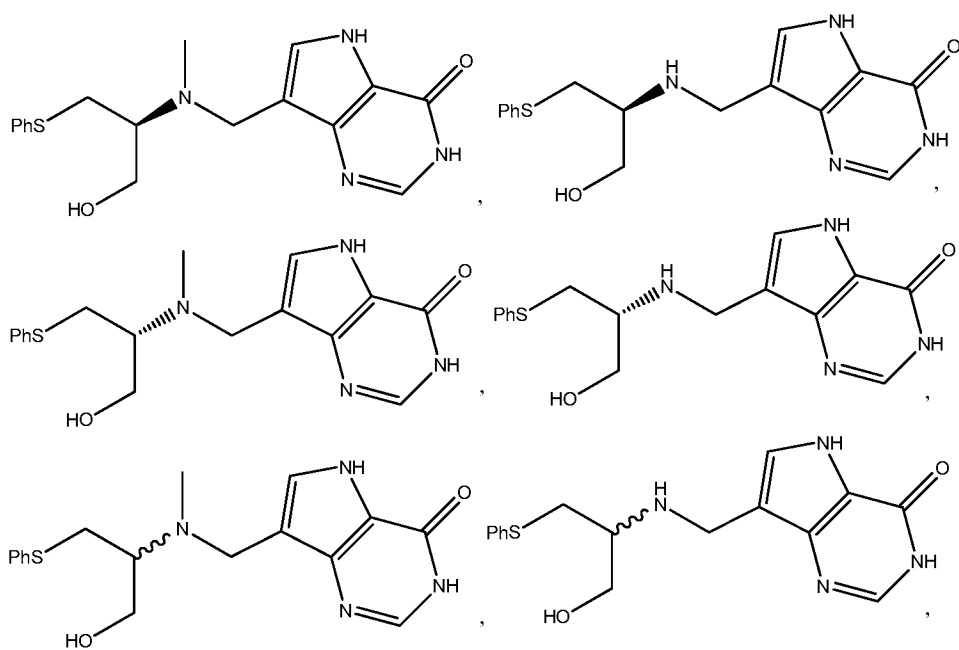


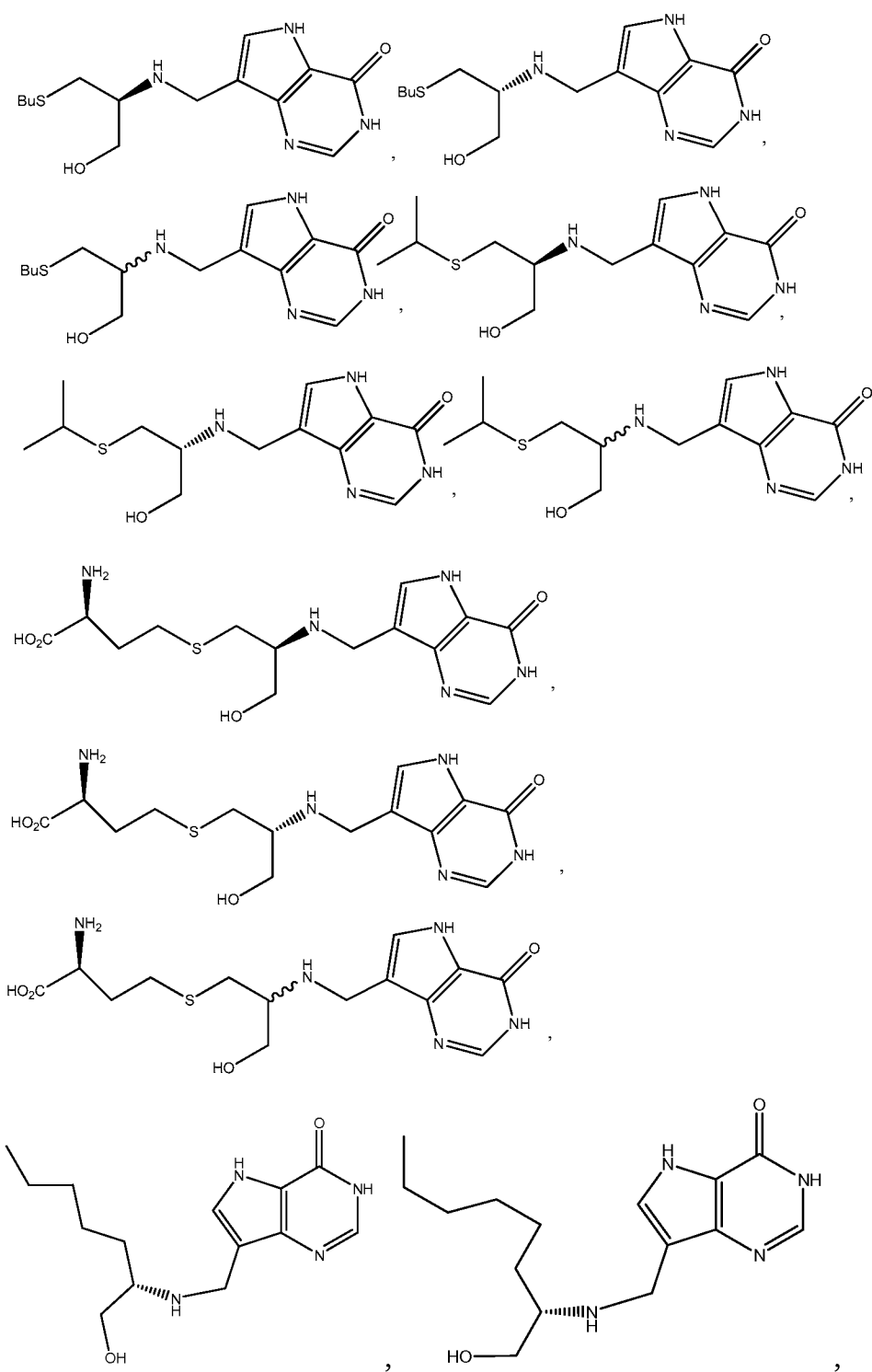


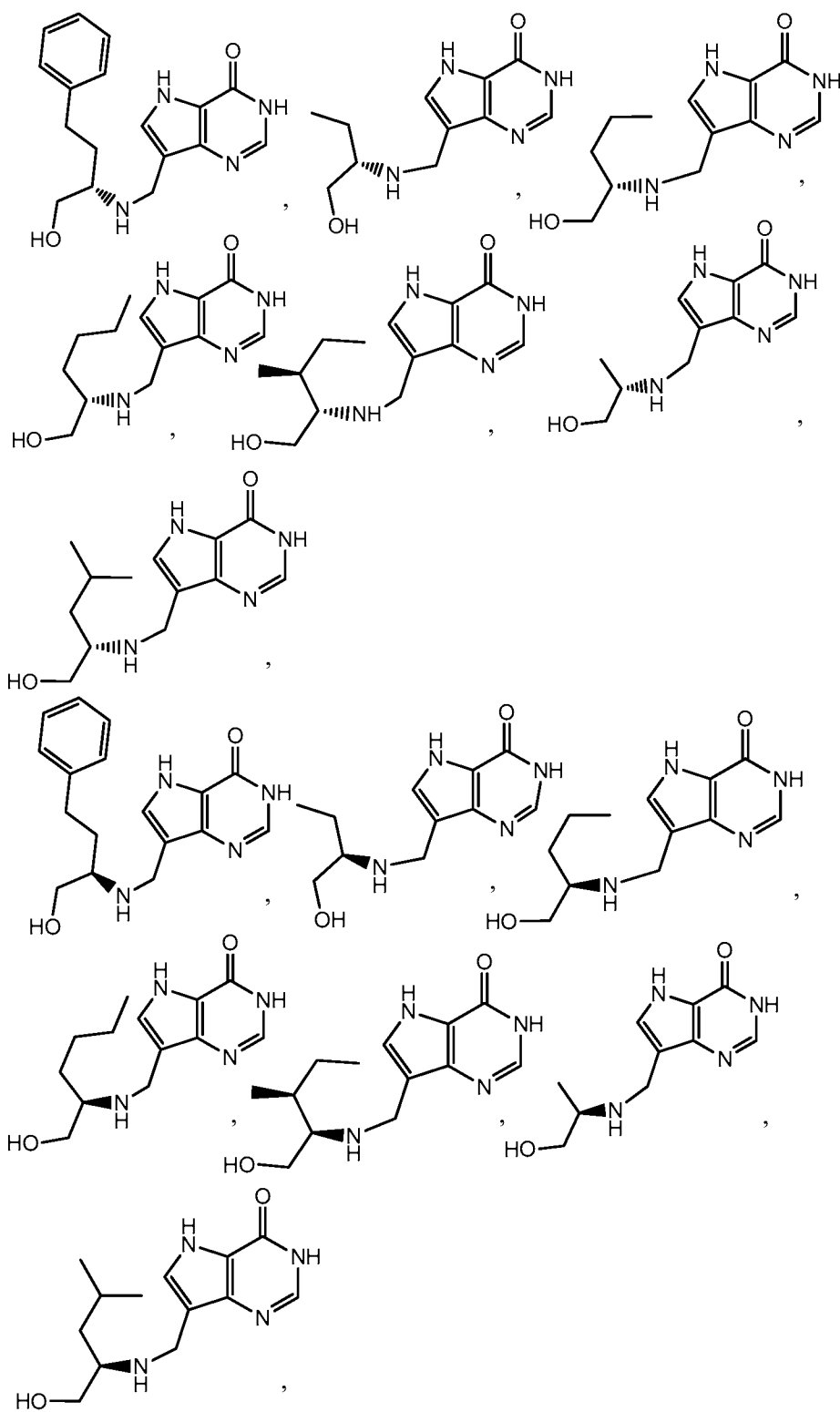


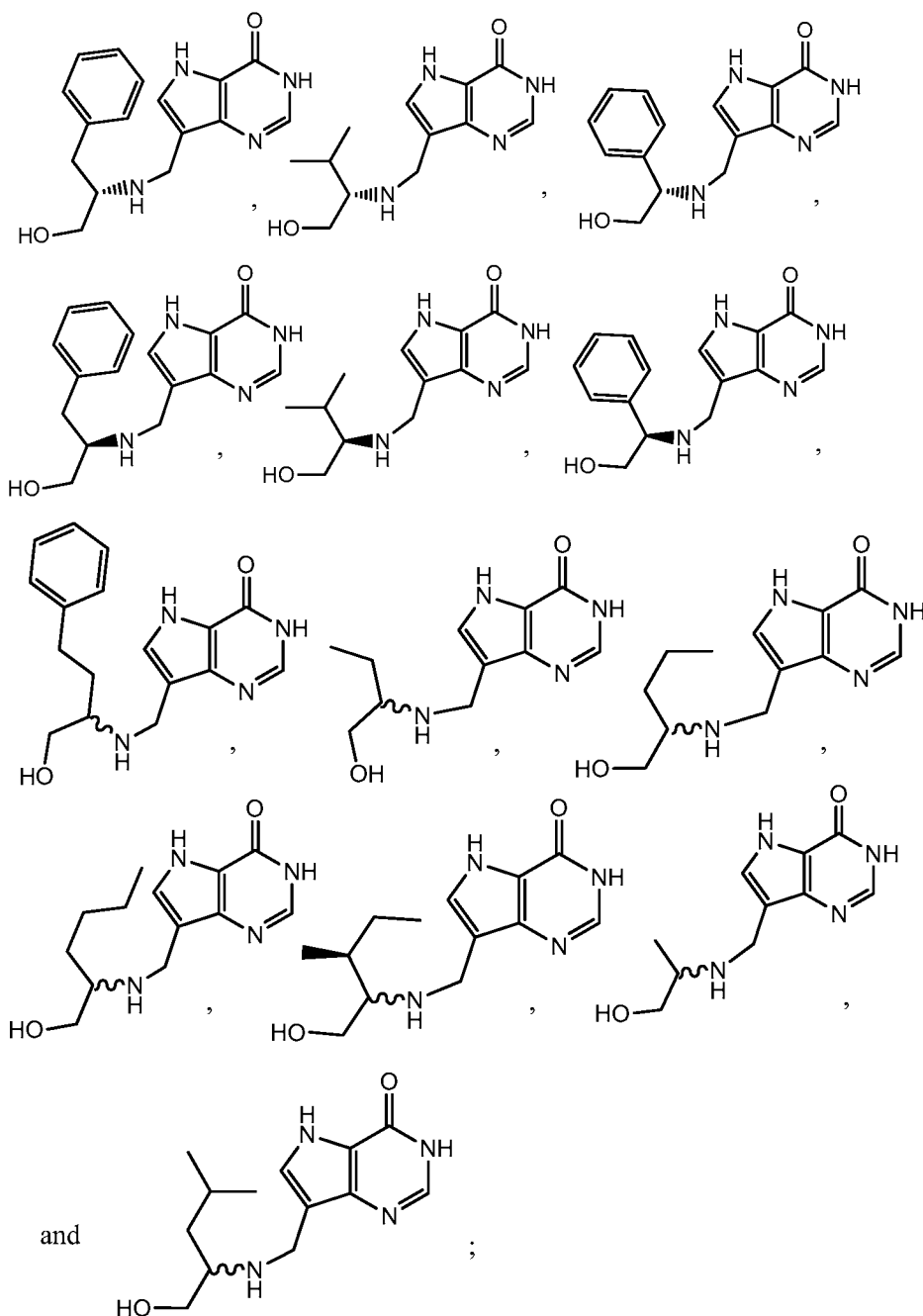


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or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

[00100] The invention also provides a pharmaceutical composition comprising any one or more of the compounds disclosed herein and a pharmaceutically acceptable salt.

[00101] The term “pharmaceutically acceptable salt” is intended to apply to non-toxic salts derived from inorganic or organic acids, including, for example, the following acid salts: acetate, adipate, alginate, aspartate, benzoate, benzenesulfonate, bisulfate, butyrate, citrate, camphorate, camphorsulfonate, cyclopentanepropionate, digluconate, dodecylsulfate, ethanesulfonate, formate, fumarate, glucoheptanoate, glycerophosphate, glycolate,

hemisulfate, heptanoate, hexanoate, hydrochloride, hydrobromide, hydroiodide, 2-hydroxyethanesulfonate, lactate, maleate, malonate, methanesulfonate, 2-naphthalenesulfonate, nicotinate, nitrate, oxalate, palmoate, pectinate, persulfate, 3-phenylpropionate, phosphate, picrate, pivalate, propionate, *p*-toluenesulfonate, salicylate, succinate, sulfate, tartrate, thiocyanate, and undecanoate.

[00102] The invention also provides a pharmaceutical composition or an agrochemical composition comprising one or more of the compounds disclosed herein and a pharmaceutically or agrochemically acceptable carrier. The compound can be present in a concentration that is a sub-bacterial-growth inhibiting amount of a 5'-methylthioinosine phosphorylase (MTIP) inhibitor.

[00103] The invention also provides a pharmaceutical composition or an agrochemical composition comprising any one or more of the compounds disclosed herein and one or more additional compounds, such as 5'-methylthio-inosine or 5'-methylthioadenosine.

[00104] The invention also provides methods for treating infections caused by bacteria that use 5'-methylthioinosine phosphorylase (MTIP) in a quorum sensing pathway comprising coadministering to a subject having the infection a sub-growth inhibiting amount of a 5'-methylthioinosine phosphorylase (MTIP) inhibitor, and 5'-methylthioinosine and/or 5'-methylthioadenosine.

[00105] This invention will be better understood from the Experimental Details, which follow. However, one skilled in the art will readily appreciate that the specific methods and results discussed are merely illustrative of the invention as described more fully in the claims that follow thereafter.

EXPERIMENTAL DETAILS

Introduction

[00106] *P. aeruginosa* is an unusual bacterium as it possesses a putative 5'-methylthioadenosine phosphorylase (MTAP: PA3004 gene) instead of MTAN. MTAP is rare in bacteria and common in mammals, while MTAN is not found in mammals. The action of MTAP on MTA would be functionally similar to that of MTAN by relieving MTA product inhibition of AHL synthase and SAM recycling in *P. aeruginosa* (18). The PA3004 gene in *P. aeruginosa* PAO1 was originally annotated from sequence homology, followed by metabolic studies in PA3004 mutants that mistakenly supported the MTAP assignment (19, 20). The present study establishes the PA3004 encoded protein to be 5'-methylthioinosine phosphorylase (MTIP) rather than MTA phosphorylase (MTAP). Examination of MTA

metabolism in *P. aeruginosa* using [8-¹⁴C]MTA confirmed that the pathway involves MTIP. Transition state analogue inhibitors with picomolar K_i values for *Pa*MTIP are described.

Materials and Methods

[00107] *Chemicals.* MT-ImmH, PhT-ImmH, and MT-DADMe-ImmH were synthesized by the Carbohydrate Chemistry Team of Industrial Research Ltd, Lower Hutt, New Zealand (Figure 1). [8-¹⁴C]MTA was synthesized as described previously (21). All other chemicals and reagents were obtained from Sigma or Fisher Scientific, and were of reagent grade.

[00108] *Plasmid construction.* A synthetic gene was designed based on the predicated protein sequence of NP_251694.1 in NCBI, annotated as MTAP of *Pseudomonas aeruginosa* PAO1. The synthetic gene was obtained in a pJexpress414 expression vector from DNA2.0 Inc. This construct encodes an additional 14 amino acids at the N-terminus which includes a His₆ tag and a TEV cleavage site.

[00109] *Enzyme purification and preparation.* BL21-CodonPlus(DE3)-RIPL *E. coli* were transformed with the above plasmid and grown overnight at 37 °C in 50 mL of LB medium with 100 µg/mL Ampicillin. The culture was transferred into 1 L of LB/Ampicillin medium and growth continued at 37 °C to an O.D.₆₀₀ of 0.7. Expression was induced by addition of 1 mM IPTG. After 4 hours at 37 °C, the cells were harvested by centrifugation at 4500 g for 30 min. The cell pellet was suspended in 20 mL of 15 mM imidazole, 300 mM NaCl, and 50 mM phosphate, pH 8.0 (lysis buffer), with 2 tablets of EDTA-free protease inhibitor (from Roche Diagnostics) and lysozyme (from chicken egg) added to the mixture. Cells were disrupted twice with a French Press and centrifuged at 20,000 g for 30 min. The supernatant was loaded onto a 4 mL column of Ni-NTA Superflow resin previously equilibrated with 5 columns of lysis buffer. The column was washed with 5 volumes of 80 mM imidazole, 300 mM NaCl, and 50 mM phosphate, pH 8.0 (wash buffer), and enzyme was eluted with 3 volumes of 250 mM imidazole, 300 mM NaCl, and 50 mM phosphate, pH 8.0 (elution buffer). The purified enzyme (> 95% purity on the basis of SDS-PAGE) was dialyzed against 50 mM Hepes, pH 7.4 and concentrated to 8 mg/ml. Enzyme was stored at -80 °C. The extinction coefficient of target protein is 20.4 mM⁻¹cm⁻¹ at 280 nm, determined by ProtParam program from ExPASy.

[00110] *Enzymatic assays.* Product formation was monitored by conversion of hypoxanthine to uric acid by xanthine oxidase (22). The extinction coefficient of this assay was 12.9 mM⁻¹cm⁻¹ at 293 nm. Enzyme activity with adenosine or MTA as substrates was

determined by conversion of adenine to 2,8-dihydroxyadenine using the same coupling enzyme (23). Its extinction coefficient at 293 nm was $15.2 \text{ mM}^{-1}\text{cm}^{-1}$. Reactions were carried out at 25°C in 1 cm cuvettes, 1 mL volumes of 100 mM Hepes, pH 7.4, 100 mM phosphate, pH 7.4, variable concentrations of nucleoside substrate, 0.5 unit of xanthine oxidase, 5 mM DTT, and appropriate amounts of purified MTIP. Reactions were initiated by addition of enzyme and the initial rates were monitored with a CARY 300 UV-Visible spectrophotometer. Control rates (no *Pa*MTIP) were subtracted from initial rates. K_m and k_{cat} values for *Pa*MTIP were obtained by fitting initial rates to the Michaelis-Menten equation using GraFit 5 (Erithacus Software). Phosphate was found to be near saturation when present at 100 mM.

[00111] *Inhibition assays.* Assays for slow-onset inhibitors were carried out by adding 1 nM *Pa*MTIP into reaction mixtures at 25°C containing 100 mM Hepes, pH 7.4, 100 mM phosphate, pH 7.4, 2 mM MTI, 5 mM DTT, 0.5 unit of xanthine oxidase and variable inhibitor concentration. Inhibitors were present at ≥ 10 times the enzyme concentration, required to simplify data analysis (24). Assays for MTA inhibition used 200 μM MTI. Controls having no enzyme and no inhibitor were included in all of the inhibition assays. Inhibition constants were obtained by fitting initial rates with variable inhibitor concentrations to equation (1) using GraFit 5 (Erithacus Software):

$$\frac{v_i}{v_o} = \frac{[S]}{K_m + [S] + \frac{K_m[I]}{K_i}} \quad (1)$$

where v_i is the initial rate in the presence of inhibitor, v_o is the initial rate in the absence of inhibitor, K_m is the Michaelis constant for MTI, $[S]$ and $[I]$ are MTI and inhibitor concentrations, respectively, and K_i is the inhibition constant. Slow-onset inhibitors display a second phase of tighter binding after reaching a thermodynamic equilibrium with the enzyme. The equilibrium constant for the second binding phase is indicated as K_i^* . This constant was obtained by fitting initial final rates and inhibitor concentrations to equation (1) using GraFit 5 (22).

[00112] *Protein crystallization and data collection.* Recombinant *Pa*MTIP (9 mg/ml) in 50 mM HEPES, pH 7.4 crystallized in 30% polyethylene glycol monomethyl ether 2000, and 0.1 M potassium thiocyanate in the presence of 5 mM MTI and 5 mM sulfate by sitting-drop vapor diffusion. Crystals were transferred to a fresh drop of crystallization solution supplemented with 20% glycerol and flash-cooled in liquid nitrogen. X-ray diffraction data

were collected at Beamline X29A, Brookhaven National Laboratory and processed with the HKL2000 program suite (Table 1).

[00113] *Structure determination and refinement.* The crystal structure of *PaMTIP* was determined by molecular replacement with Molrep using the published structure of *Sulfolobus tokodaii* MTAP (PDB:1V4N) as the search model (25). A model without catalytic site ligands was built by Phenix (26), followed by iterative rounds of manual model building and refinement in COOT and REFMAC5 (27, 28). Although *PaMTIP* was co-crystallized in the presence of sulfate and MTI to mimic the Michaelis complex of *PaMTIP*, based on ligand-omitted $F_o - F_c$ maps (contoured at 3σ) electron density was consistent with the presence of only a purine ring in the active site. *PaMTIP* was later confirmed to hydrolyze MTI under these conditions. Hypoxanthine was modeled in the active site of *PaMTIP* (Table 1).

[00114] *MTA catabolism using [8- ^{14} C]MTA.* *P. aeruginosa* PAO1 (ATCC number: 15692) was grown at 37 °C in LB medium for 16 hours. Cells were collected by centrifugation at 16100 g and washed twice with 100 mM phosphate, pH 7.4. Washed cells were lysed using BugBuster (Novagen). Cleared lysate (53 μ L) was incubated with [8- ^{14} C]MTA (10 μ L containing approximately 0.1 μ Ci ^{14} C) in 100 mM phosphate, pH 7.4, for 10 and 25 min. Reaction mixtures were quenched with perchloric acid (1.8 M final concentration) and neutralized with potassium hydroxide. Precipitates were removed by centrifugation and carrier hypoxanthine, adenine, MTI, and MTA were added to the cleared supernatant. Metabolites were resolved on a C₁₈ Luna HPLC column (Phenomenex) with a gradient of 5-52.8% acetonitrile in 20 mM ammonium acetate, pH 5.2. UV absorbance was detected at 260 nm and the retention times were 5.1 min for hypoxanthine, 7.5 min for adenine, 20.4 min for MTI, and 21.9 min for MTA. Fractions were collected in scintillation vials, dried, reconstituted in 200 μ L deionized water prior to addition of 10 mL ULTIMA GOLD LSC-Cocktail and ^{14} C was counted for three cycles at 20 minutes per cycle using a Tri-Carb 2910TR liquid scintillation analyzer. Control experiments replaced cell lysate by lysis buffer in reaction mixtures.

Results and Discussion

[00115] *Annotation of PA3004 as a MTAP.* The PA3004 gene of *P. aeruginosa* PAO1 encodes a protein (NCBI ID of NP_251694.1) annotated as a "probable nucleoside phosphorylase" in PseudoCAP (29). It was later proposed to be an MTAP based on catabolism studies in mutant strains of *P. aeruginosa* (19, 20). PAO503 is a *P. aeruginosa*

methionine-auxotroph. A new strain (PAO6422) was created by inactivating the PA3004 gene of PAO503. While PAO503 was complemented for growth on minimum medium with methionine, homocysteine or MTA, PAO6422 responded to methionine and homocysteine but was not complemented by MTA (20). These results supported an MTAP activity for the PA3004 encoded protein. The results also support a pathway of $\text{MTA} \rightarrow \text{MTI} \rightarrow \text{hypoxanthine} + \text{MTR-1-P} \rightarrow \text{methionine}$, but this was not considered.

[00116] *PA3004 encodes a MTI phosphorylase.* Here, the recombinant protein from PA3004 was purified and tested for substrate specificity (Table 2). The recombinant protein could not utilize MTA in the presence or absence of phosphate. The most favorable reaction was phosphorolysis of MTI with a k_{cat} of 4.8 s^{-1} , K_{m} of $2.6 \text{ }\mu\text{M}$ ($k_{\text{cat}}/K_{\text{m}}$ of $1.8 \times 10^6 \text{ M}^{-1}\text{s}^{-1}$). Enzyme was less active with inosine with a k_{cat} of 0.57 s^{-1} and a K_{m} of $90 \text{ }\mu\text{M}$ ($k_{\text{cat}}/K_{\text{m}}$ of $6.3 \times 10^3 \text{ M}^{-1}\text{s}^{-1}$) and depended on phosphate for significant activity. The methylthio- group of MTI is important for both substrate binding and catalysis. Adenosine is a weak substrate with a k_{cat} of 0.0549 s^{-1} and a K_{m} of $23 \text{ }\mu\text{M}$ ($k_{\text{cat}}/K_{\text{m}}$ of $2.4 \times 10^3 \text{ M}^{-1}\text{s}^{-1}$). Thus the protein encoded by PA3004 is a relatively specific MTI phosphorylase. The catalytic efficiency ($k_{\text{cat}}/K_{\text{m}}$) for MTI is 290 times larger than for inosine, the second best substrate. For comparison, the MTI phosphorylase activity of *Pf*PNP (see below) has a $k_{\text{cat}}/K_{\text{m}}$ of $9.4 \times 10^4 \text{ M}^{-1}\text{s}^{-1}$ for MTI and a $k_{\text{cat}}/K_{\text{m}}$ of $3.6 \times 10^5 \text{ M}^{-1}\text{s}^{-1}$ for inosine.

[00117] MTI phosphorylase activity has been documented in *Caldariella acidophilan* MTAP, human MTAP, human PNP, and *P. falciparum* PNP, but in these, MTI is a relatively poor substrate (30-33). MTIP of *P. aeruginosa* is the only reported MTIP with high specificity for MTI. MTA is not a substrate (less than the detection level of $10^{-4} k_{\text{cat}}$) but is a competitive inhibitor of MTI, with a K_{i} value of $70 \text{ }\mu\text{M}$, three times greater than the K_{m} of adenosine. Thus, MTA binds to the active site of the enzyme but is not catalytically competent.

[00118] Recombinant MTIP was expressed with a 14 amino acid extension at the N-terminus. Incubation with TEV protease removed 13 of these, leaving one additional glycine at the N-terminus. This construct exhibited identical substrate specificity as the original recombinant protein. The crystal structure (see below) shows the N-terminal extension to be remote from the active site. Unchanged substrate specificity with or without the 13 amino acid extension supports activity of the native enzyme to be *Pa*MTIP.

[00119] *Crystal structure of PaMTIP:hypoxanthine.* The crystal structure of *Pa*MTIP in complex with hypoxanthine was determined to $2.0 \text{ }\text{\AA}$ resolution with two homotrimers in

the asymmetric unit. Residues 2 to 54 and 60 to 243 of each *Pa*MTIP monomer are ordered in the electron density map. The N terminal His₆ tag and TEV protease site are disordered and distant from the active site. The *Pa*MTIP monomer exhibits an α/β -fold consisting of a 7-stranded β -sheet and 5 α -helices. The active sites of *Pa*MTIP are located near the interfaces formed between monomers in the trimer. Each trimeric *Pa*MTIP forms three active sites. Although *Pa*MTIP was co-crystallized with MTI and sulfate (5 mM) to mimic the Michaelis complex, the ligand-omitted difference Fourier map showed only the presence of hypoxanthine. Kinetic experiments demonstrated slow hydrolysis of MTI ($3.8 \times 10^{-5} \text{ s}^{-1}$) to generate hypoxanthine and 5-methylthioribose. Crystallization attempts with apo-*Pa*MTIP or with *Pa*MTIP in complex with hypoxanthine and phosphate failed to yield crystals. In the crystal structure, hypoxanthine is wedged between the backbone of a 5-stranded β sheet and the side chains of Leu180 and Met190. Structure-based sequence alignment revealed that Met190 is conserved and Leu180 is replaced by Phe in human PNP and human MTAP (Fig. 2). Hypoxanthine N7 and O6 form hydrogen bond with the side chain of Asn223 in *Pa*MTIP and N1 forms a hydrogen bond with the side chain of Asp 181.

[00120] *Comparison with Other Purine N-Ribosylphosphorylases.* *Pa*MTIP, PNPs and MTAPs have similar functions in the phosphorolysis of nucleosides, but show different substrate specificities. The structural architecture of *Pa*MTIP is similar to the four MTAPs and seven trimeric PNPs in the Protein Data Bank (r.m.s.d is in the range of 0.8 to 1.2 Å for the monomers). Structure-based sequence alignments with *Pa*MTIP show a 28-40% identity with the four MTAPs and 20-32% with the seven trimeric PNPs (Fig. 2). However, *Pa*MTIP shows no significant similarity in amino acid sequence or quaternary structures with the hexameric PNPs including that from *Plasmodium* species. Because of this structural difference, *Plasmodium* PNPs are not included in the following analysis even though most *P. falciparum* PNPs also catalyze the phosphorolysis of MTI (34). The active sites of *Pa*MTIP can be compared to those of PNP and MTAP by considering three distinct regions corresponding to the purine-, (methylthio)ribose- and phosphate-binding sites. The Glu201 and Asn243 (human PNP numbering) are conserved in the purine binding site of *Pa*MTIP and PNPs (Fig. 2), and have an important role in 6-oxopurine specificity by hydrogen bonding to N1, O6 and N7 of the 6-oxopurine (35). Human PNP exhibits a 350,000-fold catalytic preference for 6-oxopurines compared to 6-aminopurines (36). The Asn243Asp mutant and Glu201Gln:Asn243Asp double mutant in human PNP is known to shift the substrate preference in favor of adenosine, a 6-aminopurine substrate (36). In contrast, MTAPs prefer 6-aminopurine (31). Human MTAP Ser178 (via a water molecule), Asp220

and Asp222 are conserved for the MTAPs, forming hydrogen bonds to N1, O6 and N7 of 6-aminopurine, respectively (37).

[00121] Human PNP is efficient for phosphorolysis of purine nucleosides with a 5'-hydroxyl group but not for purine nucleosides with a 5'-methylthio group. The conserved His257 and Phe159 (from the adjacent monomer) of human PNP are important in binding the 5'-hydroxyl group, whereas in MTAP and *Pa*MTIP, a small hydrophobic amino acid corresponding to His257 and a His corresponding to Phe159 (human PNP numbering), provides space to accommodate the 5'-methylthio group. Consistent with these observations, the His257Gly mutant of human PNP binds 5'-methylthio- inhibitors tighter than the corresponding 5'-hydroxyl- inhibitors (38). Superposition of the liganded human MTAP and PNP structures reveals a 1 Å shift of phosphate toward the purine in human MTAP. Phosphate forms favorable hydrogen bonds with the 2'- and 3'- hydroxyl groups of (5'-methylthio)ribose, and thereby anchors (5'-methylthio)ribose to the active site. The spatial shift of the phosphate of human MTAP relative to that of human PNP places the 5'-methylthioribose closer to the purine binding site and this shift provides more room to accommodate the 5'-methylthio group in the active site of human MTAP. Structure-based sequence alignment shows that *Pa*MTIP and MTAP share key residues in the phosphate and (methylthio)ribose- binding sites; however, *Pa*MTIP has a purine binding site more similar to the PNPs, consistent with its preference for 6-oxypurines.

[00122] *P. aeruginosa* Catabolism [8-¹⁴C]MTA. Substrate specificity and structural data of PA3004 support a physiological function as MTIP. But this role requires production of MTI in *P. aeruginosa*; however, there is no previous report of MTI as a metabolite in *P. aeruginosa* and the published genetic approach supported MTAP activity (20). Catabolism of [8-¹⁴C]MTA in lysates of *P. aeruginosa* was followed by analysis of hypoxanthine, adenine, MTI, and MTA. If MTA is first deaminated to MTI followed by *Pa*MTIP action, the sequential conversion to [8-¹⁴C]MTI and hypoxanthine would occur without adenine formation. After 10 min with lysate, 77% of the MTA was converted to MTI (52%) and hypoxanthine (25%) without significant formation of adenine (Fig. 3). As 98% of the total ¹⁴C-label was recovered, the results establish MTA conversion to MTI and hypoxanthine but not to adenine. At 25 min incubation, over 97% of the [8-¹⁴C] MTA was recovered as MTI (45%) and hypoxanthine (53%). Continuous conversion of MTA → MTI → hypoxanthine without significant MTAP or MTAN activity is supported by these results, highlighting the requirement for an MTA deaminase to catalyze the conversion of MTA to MTI. There is no significant MTA phosphorylase or MTA nucleosidase activity in *P. aeruginosa* abstracts.

[00123] *MTI Formation in P. falciparum.* A similar pathway of MTA catabolism is found in *Plasmodium* species (34). The adenosine deaminase of *P. falciparum* (*PfADA*) deaminates adenosine and MTA as substrates with similar k_{cat}/K_m values (6.2×10^4 and 8.8×10^4 , $\text{M}^{-1}\text{s}^{-1}$ respectively) (34). Its purine nucleoside phosphorylase (*PfPNP*) also degrades inosine and MTI to hypoxanthine with similar catalytic efficiency (3.6×10^5 and 9.4×10^4 , $\text{M}^{-1}\text{s}^{-1}$ respectively) (33). Human PNP is a poor catalyst for MTI phosphorolysis and mammalian ADA does not deaminate MTA (34). The substrate specificities of the *P. falciparum* enzymes permit conversion of MTA to hypoxanthine via MTI. This process was reconstituted *in vitro* with purified *PfADA* and *PfPNP* (34). Metabolic and genomic studies have thus established that neither *P. aeruginosa* nor *P. falciparum* encode MTAN or MTAP activities.

[00124] *MTI in Other Organisms.* Although MTI has been used as an MTA analogue, its function as a metabolite has been documented only in *Plasmodium* (30,31, 39-42). Metabolism studies on *Saccharomyces cereviceae* suggested *in vivo* conversion of MTA to MTI, but the corresponding enzyme(s) and fate of MTI have not been determined (43). In humans, MTAP catalyzes the conversion of MTA to adenine and 5-methylthioribose- α -D-1-phosphate. And no other fate is known for MTA although it appears in human urine (44). Excess MTA added to cultured human cells is known to form some MTI through the reverse reaction of PNP using hypoxanthine and MTR-1-P, but it is not physiologically significant (45-46). The physiological fate of $[8\text{-}^{14}\text{C}]\text{MTI}$ in *P. falciparum* has been established by following its incorporation into nucleic acids, where it was shown to be equivalent to inosine as a nucleic acid precursor (34).

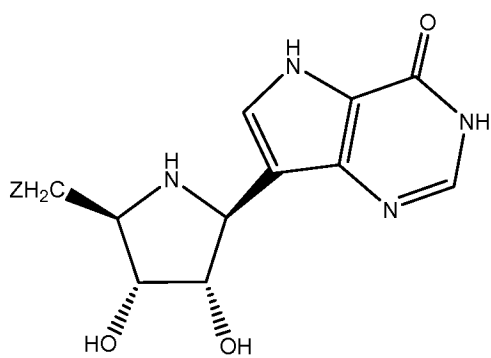
[00125] The surprising discovery of MTIP catalytic activity in *Psuedomonas aeruginosa* led to a search of the genome data bases for other microbial species that could be treated in the same manner as *P. aeruginosa* (Table 4). Of particular interest are *Pseudomonas syringae* and *Xanthomonas campestris*, which are important plant pathogens. *P. syringae* produces ice nucleation-active (Ina) proteins that cause water to freeze at fairly high temperatures resulting in surface frost damage to plants (51). It has been estimated that frost accounts for approximately \$1 billion in crop damage each year in the United States alone. *Xanthomonas campestris* causes a variety of plant diseases.

[00126] *Picomolar inhibitors of PaMTIP.* Immucillins are transition-state analogues developed for *N*-ribosyl transferases that have ribocation character in their transition states (47). ImmH is a first generation Immucillin, representing early transition states exemplified

by bovine PNP (Fig. 1). DADMe-ImmH is a second generation Immucillin and mimics the fully dissociated transition states of human and *P. falciparum* PNPs (48). 5'-Alkylthio- and arylthio- derivatives of two generations of Immucillins (MT-ImmH, PhT-ImmH, and MT-DADMe-ImmH) have been synthesized to target the transition-state features of *Pf*PNP with MTI as the substrate (34). The inhibitors exhibited slow-onset inhibition of *Pa*MTIP, suggesting a slow conversion of initial enzyme-inhibitor complex to a more stable conformation.

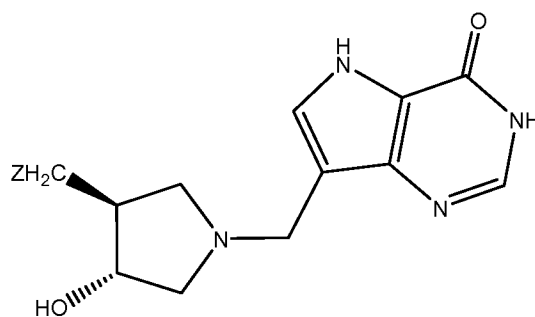
[00127] MT-ImmH inhibited *Pa*MTIP with a K_i^* value of 76 pM, binding 4-fold more tightly than MT-DADMe-ImmH, suggesting that the first generation Immucillins more closely resemble the transition state (Table 3). PhT-ImmH was the most tightly bound inhibitor with a K_i^* value of 35 pM and demonstrating the importance of hydrophobic interactions at the 5'-position. Increasing the lipophilicity at the equivalent position in the second generation inhibitors by extending the alkyl chain length in going from MT-DADMe-ImmH to PrT-DADMe-ImmH similarly enhanced potency, in this instance by a factor of 4, confirming that both first and second generation Immucillins can provide highly effective inhibitors. Introduction of the 4'-C-hydroxy group (4'-HO-MT-DADMe-ImmH) provides a nanomolar inhibitor of *Pa*MTIP. The acyclic Immucillin, MT-SerMe-ImmH, inhibited *Pa*MTIP with a K_i^* value of 96 pM, confirming that this class of Immucillin is also able to inhibit MTIP enzymes effectively. The MT-TrisMe-ImmH analogue, has a K_i value of 19 nM. The K_{MTI}/K_i values were 34,000 for MT-ImmH, 7,600 for MT-DADMe-ImmH, and 74,000 for PhT-ImmH, emphasizing the potency of these transition state analogue inhibitors. Inhibitor specificity can be compared to that for *Pf*PNP, which catalyzes the same reaction. Thus, MT-ImmH, PhT-ImmH, and MT-DADMe-ImmH bind more weakly to *Pf*PNP with dissociation constants of 2.7 nM, 150 nM, and 0.9 nM, respectively (Table 3) (33, 49). Binding of the bulky, hydrophobic 5'-PhT-group is preferred by *Pa*MTIP where $K_i^* = 35$ pM. This inhibitor induces slow-onset, tight-binding inhibition. In contrast, the same inhibitor has $K_i = 150$ nM with *Pf*PNP, where it binds 4,300-fold more weakly and does not induce slow-onset inhibition. The K_i^* values for certain MTIP inhibitors are indicated below.

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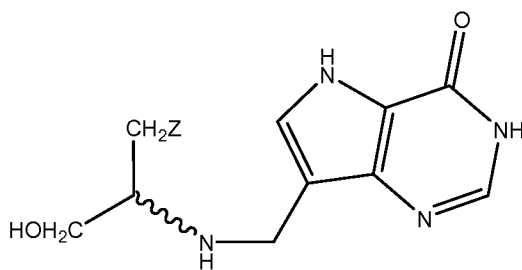


(A) Z = SMe $K_i^* = 76$ pM

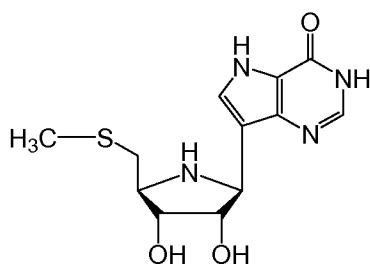
(B) Z = SPh $K_i^* = 35$ pM



(C) Z = SMe $K_i^* = 340$ pM



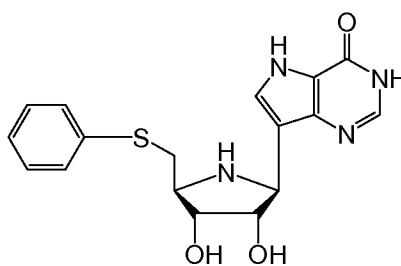
(D) Z = SMe $K_i^* = 96$ pM



MT-ImmH

$$K_i^* = 76 \pm 5 \text{ pM}$$

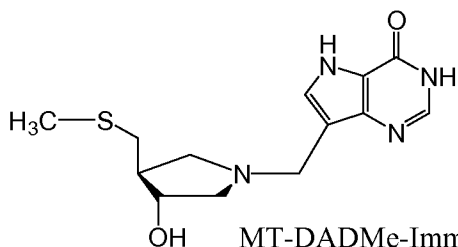
$$K_i = 840 \pm 50 \text{ pM}$$



PT-ImmH

$$K_i^* = 35 \pm 3 \text{ pM}$$

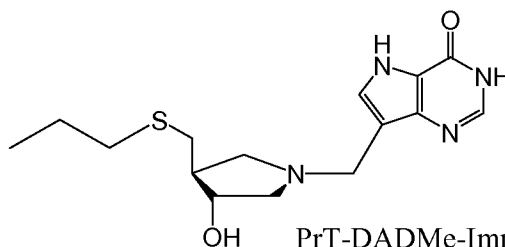
$$K_i = 660 \pm 90 \text{ pM}$$



MT-DADMe-ImmH

$$K_i^* = 340 \pm 20 \text{ pM}$$

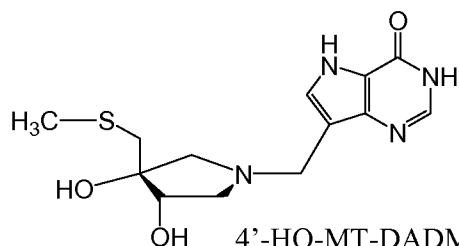
$$K_i = 800 \pm 100 \text{ pM}$$



PrT-DADMe-ImmH

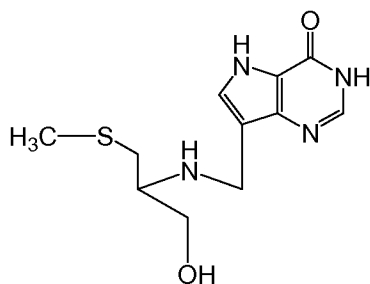
$$K_i^* = 72 \pm 2 \text{ pM}$$

$$K_i = 380 \pm 40 \text{ pM}$$



4'-HO-MT-DADMe-ImmH

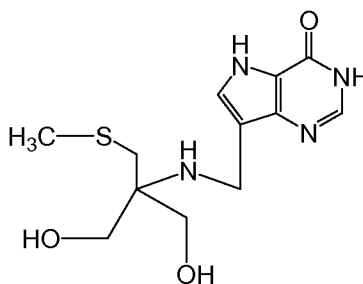
$$K_i = 1.21 \pm 0.01 \text{ nM}$$



MT-SerMe-ImmH

$$K_i^* = 96 \pm 2 \text{ pM}$$

$$K_i = 720 \pm 30 \text{ pM}$$



MT-TrisMe-ImmH

$$K_i = 19 \pm 1 \text{ nM}$$

[00128] *The Nature of the PaMTIP Transition State.* PfpNP has a fully-dissociated ribocation transition state with approximately 3 Å between the C1' ribocation and N9 and a similar separation between C1' and the attacking phosphate oxygen. Thus, it prefers to bind

MT-DADMe-ImmH rather than MT-ImmH (50). *Pa*MTIP shows the opposite pattern, consistent with an early, dissociative transition state.

[00129] *Implications for Quorum Sensing.* Inhibition studies of *Pa*MTIP have identified inhibitors with K_i^* values in the picomolar range. These are candidates for blocking *Pa*MTIP activities. In most bacteria, MTAN inhibition blocks quorum sensing, but the lack of MTAN in *P. aeruginosa* indicates that *Pa*MTIP becomes an equivalent target. This study has revealed the pathway of MTA metabolism in *P. aeruginosa* and provided new tools to explore this unusual bacterial pathway.

Table 1. Data collection and refinement statistics.

Data collection	
PDB	3OZB
Space group	P4 ₁ 2 ₁ 2
Cell dimension	
a, b, c (Å)	99.5, 99.5, 334.9
α , β , γ (°)	90.0, 90.0, 90.0
Resolutions (Å)	20.0-2.8 (2.9-2.8)
R _{sym} (%)	17.2 (95.5)
I / σ I	9.6 (1.9)
Completeness (%)	100.0 (100.0)
Redundancy	7.0 (7.2)
Refinement	
Resolution (Å)	20.00-2.0
No. reflections	39915
R _{work} / R _{free} (%)	20.1 / 26.2
B-factors (Å ²)	
Protein	
(<i>main chain</i>)	39.9
(<i>side chain</i>)	40.9
Water	25.2
Ligand	46.0
No. of Atoms	
Protein	10800
Water	68
Ligand	60
R.m.s deviations	
Bond lengths (Å)	0.012
Bond angles (°)	1.46
Ramachran analysis	
allowed region	99.3%
disallowed region	0.7%

Numbers in parentheses are for the highest-resolution shell. One crystal was used for each data set.

Table 2. Substrate specificity of *Pa*MTIP.

Substrate	k_{cat} (s^{-1})	K_{m} (μM)	$K_{\text{cat}}/K_{\text{m}}$ ($\times 10^3 \text{ M}^{-1}\text{s}^{-1}$)
MTI	4.8 ± 0.2	2.6 ± 0.4	1800 ± 300
MTA ^a	N/A	70 ± 20	N/A
inosine	0.57 ± 0.04	90 ± 20	6 ± 1 _[300] ^b
adenosine	0.0549 ± 0.0005	23 ± 1	2.4 ± 0.1 _[750] ^b

^a MTA is not a substrate of *Pa*MTIP. K_{i} is used instead of K_{m} for MTA.

^b Numbers in [] are fold decreases of $K_{\text{cat}}/K_{\text{m}}$ in comparison with those of MTI.

^c Values are $\pm$ S.E.

Table 3. Summary of K_{i} values for *Pa*MTIP and *Pf*PNP.

Inhibitors	<i>Pa</i> MTIP		<i>Pf</i> PNP ^a	
	K_{i} (pM)	K_{i}^* (pM)	K_{i} (pM)	K_{i}^* (pM)
MT-ImmH	840 ± 50	76 ± 5	22000 ± 3000	2700 ± 400
PhT-ImmH	660 ± 90	35 ± 3	150000 ± 8000	ND
MT-DADMe-ImmH	800 ± 100	340 ± 20	11000 ± 4000	900 ± 100

^a Inhibition constants of *Pf*PNP were from Lewandowicz *et al.* (49).

^b Values are $\pm$ S.E.

Table 4. Microbial species searched for possible MTIP catalytic activity.

Feature ID	Locus_Tag	Descrip-tion	Species
31909191	VBIACiFer109666_1307	MTAP	Acidaminococcus fermentans DSM 20731
24310952	VBIACiSp85863_0249	MTAP	Acidaminococcus sp. D21
20652065	VBIACiFer29821_0353	MTAP	Acidithiobacillus ferrooxidans ATCC 23270
20658606	VBIACiFer6930_0524	MTAP	Acidithiobacillus ferrooxidans ATCC 53993
20839765	VBIAlcBor124741_1073	MTAP	Alcanivorax borkumensis SK2

24415594	VBIAlcSp134196_0029	MTAP	Alcanivorax sp. DG881
24433023	VBIAlcAci16525_1025	MTAP	Alicyclobacillus acidocaldarius LAA1
20846714	VBIAlcAci73240_1597	MTAP	Alicyclobacillus acidocaldarius subsp. acidocaldarius DSM 446
20861899	VBIAlcEhr114327_1328	MTAP	Alkalilimnicola ehrlichii MLHE-1
31920755	VBIAlcVin64954_0500	MTAP	Allochromatium vinosum DSM 180
20881494	VBIAlmDeg104956_0971	MTAP	Ammonifex degensii KC4
20958662	VBIAlcAeo85532_0465	MTAP	Aquifex aeolicus VF5
20973491	VBIAlcAro98752_3663	MTAP	Aromatoleum aromaticum EbN1
21012394	VBIAlcAzoSp26047_2034	MTAP	Azoarcus sp. BH72
21029193	VBIAlcAzoVin72790_1379	MTAP	Azotobacter vinelandii DJ
27182875	VBIAlcBerMar44794_0658	MTAP	Bermanella marisrubri
30885632	VBIAlcBetPro60814_0744	MTAP	beta proteobacterium KB13
21263520	VBIAlcCanAcc132554_3989	MTAP	Candidatus Accumulibacter phosphatis clade IIA str. UW-1
31968053	VBIAlcCanDes92303_0674	MTAP	Candidatus Desulforudis audaxviator MP104C
30383843	VBIAlcCarPac114674_0689	MTAP	Carboxydibrachium pacificum DSM 12653
21276013	VBIAlcCarHyd26463_1381	MTAP	Carboxydotherrmus hydrogenoformans Z-2901
21442613	VBIAlcChrVio67196_3821	MTAP	Chromobacterium violaceum ATCC 12472
27118095	VBIAlcCloBac45081_0978	MTAP	Clostridiales bacterium 1 7 47FAA
29068547	VBIAlcConLit32816_3743	MTAP	Congregibacter litoralis KT71
21474258	VBIAlcCopPro72829_0673	MTAP	Coprothermobacter proteolyticus DSM 5265
21602118	VBIAlcDecAro89105_1723	MTAP	Dechloromonas aromatica RCB
35334867	VBIAlcDefDes107323_0476	MTAP	Deferribacter desulfuricans SSM1
21665529	VBIAlcDesHaf15223_3920	MTAP	Desulfitobacterium hafniense DCB-2
21673960	VBIAlcDesHaf65307_2915	MTAP	Desulfitobacterium hafniense Y51
21719077	VBIAlcDesAce42372_1190	MTAP	Desulfotomaculum acetoxidans DSM 771
21730383	VBIAlcDesRed82656_2261	MTAP	Desulfotomaculum reducens MI-1
26640796	VBIAlcDetAlk125442_2011	MTAP	Dethiobacter alkaliphilus AHT 1
36975078	VBIAlcDialInv115189_0190	MTAP	Dialister invisus DSM 15470
21806196	VBIAlcDicThe33784_0766	MTAP	Dictyoglomus thermophilum H-6-12
21810560	VBIAlcDicTur93964_0965	MTAP	Dictyoglomus turgidum DSM 6724
25910268	VBIAlcGamPro11258_1251	MTAP	gamma proteobacterium HTCC5015
36991798	VBIAlcGamPro33483_0733	MTAP	gamma proteobacterium NOR5-3
22086164	VBIAlcHahChe29232_2456	MTAP	Hahella chejuensis KCTC 2396
22096829	VBIAlcHalHal112047_1253	MTAP	Halorhodospira halophila SL1
32206686	VBIAlcHalNea120669_1410	MTAP	Halothiobacillus neapolitanus c2
22108421	VBIAlcHelMod36755_1971	MTAP	Heliobacterium modesticaldum Ice1
22119229	VBIAlcHerAur93466_1152	MENA	Herpetosiphon aurantiacus ATCC 23779
38514308	VBIAlcHydSp35543_1620	MTAP	Hydrogenivirga sp. 128-5-R1-1
32212773	VBIAlcHydThe36152_1917	MTAP	Hydrogenobacter thermophilus TK-6
36565664	VBIAlcHydSp5595_0540	MTAP	Hydrogenobaculum sp. SN
22134890	VBIAlcHydSp64203_0132	MTAP	Hydrogenobaculum sp. Y04AAS1
27563531	VBIAlcJanSp77695_2689	MTAP	Janibacter sp. HTCC2649
27466181	VBIAlcLimSp111806_2854	MTAP	Limnobacter sp. MED105
30256927	VBIAlcLutNit9412_3599	MTAP	Lutiella nitroferrum 2002

22430377	VBIMagSp23654_1940	MTAP	Magnetococcus sp. MC-1
25938712	VBIMarGam125067_3745	MTAP	marine gamma proteobacterium HTCC2143
30952185	VBIMarGam120175_0624	MTAP	marine gamma proteobacterium HTCC2148
22459174	VBIMarAqu65105_2345	MTAP	Marinobacter aquaeolei VT8
30999028	VBIMarSp19762_0602	MTAP	Marinobacter sp. ELB17
36495797	VBIMegGen154667_0818	MTAP	Megasphaera genomosp. type 1 str. 28L
32268663	VBIMetFla97085_1558	MTAP	Methylobacillus flagellatus KT
22607820	VBIMetCap22254_2017	MTAP	Methylococcus capsulatus str. Bath
26225373	VBIMetThi34374_0895	MTAP	Methylophaga thiooxidans DMS010
26231633	VBIMetBac29390_0798	MTAP	Methylophilales bacterium HTCC2181
22612561	VBIMetMob89187_1188	MTAP	Methylostenobacter mobilis JLW8
38199151	VBIMetSp140979_1406	MTAP	Methylostenobacter sp. 301
22617878	VBIMetSp110381_1427	MTAP	Methylovorus sp. SIP3-4
22638531	VBIMooThe6753_0754	MTAP	Moorella thermoacetica ATCC 39073
25483803	VBINitMob112042_0928	MTAP	Nitrococcus mobilis Nb-231
35395780	VBINitHal115488_3010	MTAP	Nitrosococcus halophilus Nc4
25494267	VBINitOce87996_2021	MTAP	Nitrosococcus oceani AFC27
22704664	VBINitOce57959_0576	MTAP	Nitrosococcus oceani ATCC 19707
22714750	VBINitEur56163_1967	MTAP	Nitrosomonas europaea ATCC 19718
22719073	VBINitEut7577_1203	MTAP	Nitrosomonas eutropha C91
25500690	VBINitSp88817_2664	MTAP	Nitrosomonas sp. AL212
22724352	VBINitMul110821_0751	MTAP	Nitrospira multififormis ATCC 25196
23779746	VBISymThe116959_0804	MENA	Symbiobacterium thermophilum IAM 14863
22910709	VBIPelThe8413_1946	MTAP	Pelotomaculum thermopropionicum SI
22915331	VBIPerMar119911_0973	MTAP	Persephonella marina EX-H1
25555006	VBIPseAer126331_1953	MTAP	Pseudomonas aeruginosa 2192
25567207	VBIPseAer51129_2865	MTAP	Pseudomonas aeruginosa C3719
19813767	VBIPseAer113719_2130	MTAP	Pseudomonas aeruginosa LESB58
19826092	VBIPseAer80442_2070	MTAP	Pseudomonas aeruginosa PA7
37796667	VBIPseAer119691_1964	MTAP	Pseudomonas aeruginosa PAb1
25582692	VBIPseAer78937_3783	MTAP	Pseudomonas aeruginosa PACS2
19840565	VBIPseAer58763_3152	MTAP	Pseudomonas aeruginosa PAO1
19850174	VBIPseAer79785_2046	MTAP	Pseudomonas aeruginosa UCBPP-PA14
19889839	VBIPseFlu44242_3864	MTAP	Pseudomonas fluorescens Pf0-1
19909669	VBIPseMen131592_1613	MTAP	Pseudomonas mendocina ymp
			Pseudomonas savastanoi
37952625	VBIPseSav158853_3365	MTAP	pv. savastanoi NCPPB 3335
19963943	VBIPseStu31643_1746	MTAP	Pseudomonas stutzeri A1501
25590088	VBIPseSyr138828_1392	MTAP	Pseudomonas syringae pv. aesculi str. 2250
19975790	VBIPseSyr78478_3367	MTAP	Pseudomonas syringae pv. phaseolicola 1448A
19987196	VBIPseSyr42314_3354	MTAP	Pseudomonas syringae pv. syringae B728a
25607361	VBIPseSyr86336_4594	MTAP	Pseudomonas syringae pv. syringae FF5
25614561	VBIPseSyr56650_2715	MTAP	Pseudomonas syringae pv. tabaci ATCC 11528
19998439	VBIPseSyr93040_3591	MTAP	Pseudomonas syringae pv. tomato str. DC3000
25625381	VBIPseSyr88081_2476	MTAP	Pseudomonas syringae pv. tomato T1
28431114	VBIReiBla99298_1448	MTAP	Reinekea blandensis MED297

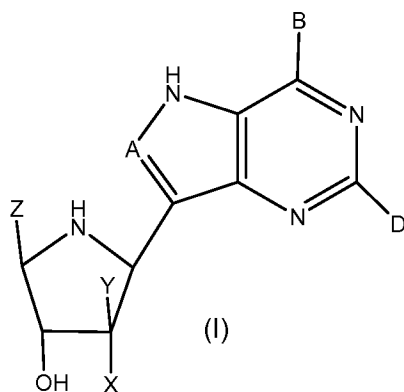
23402541	VBISacDeg56404_1951	MTAP	Saccharophagus degradans 2-40
35400680	VBISidLit69165_1281	MTAP	Sideroxydans lithotrophicus ES-1
23702535	VBISteMal45202_3320	MTAP	Stenotrophomonas maltophilia K279a
23710908	VBISteMal40512_2988	MTAP	Stenotrophomonas maltophilia R551-3
30945052	VBISteSp70125_0969	MTAP	Stenotrophomonas sp. SKA14
23764404	VBISulAzo123226_1591	MTAP	Sulfurihydrogenibium azorense Az-Fu1
23767117	VBISulSp94719_1189	MTAP	Sulfurihydrogenibium sp. YO3AOP1
28316110	VBISulYel28410_0490	MTAP	Sulfurihydrogenibium yellowstonense SS-5
23781549	VBISymThe116959_1695	MTAP	Symbiobacterium thermophilum IAM 14863
			Syntrophomonas wolfei
23856307	VBISynWol51738_0821	MTAP	subsp. wolfei str. Goettingen
38269016	VBISynLip21176_1328	MTAP	Syntrophothermus lipocalidus DSM 12680
23865162	VBISynAci70500_2363	MTAP	Syntrophus aciditrophicus SB
23880636	VBITHaSp83776_2179	MTAP	Thauera sp. MZ1T
38275831	VBITheSp141296_2211	MTAP	Thermincola sp. JR
29924698	VBITheBro57020_1537	MTAP	Thermoanaerobacter brockii subsp. finni Aka-1
30825907	VBITheEth125011_1168	MTAP	Thermoanaerobacter ethanolicus CCSD1
32497914	VBIThelta22270_1468	MTAP	Thermoanaerobacter italicus Ab9
			Thermoanaerobacter mathranii
38281044	VBITheMat18_1480	MTAP	subsp. mathranii str. A3
			Thermoanaerobacter pseudethanolicus
23886420	VBIThePse6203_0923	MTAP	ATCC 33223
31262795	VBITheSp37765_1746	MTAP	Thermoanaerobacter sp. X513
23893440	VBITheSp86957_1944	MTAP	Thermoanaerobacter sp. X514
30832536	VBITheSp21349_1908	MTAP	Thermoanaerobacter sp. X561
23897876	VBITheTen82880_1587	MTAP	Thermoanaerobacter tengcongensis MB4
32508736	VBITheAlb52854_1221	MTAP	Thermocrinis albus DSM 14484
23908977	VBITheYel104483_0962	MTAP	Thermodesulfovibrio yellowstonii DSM 11347
30839490	VBITheCar126673_2743	MTAP	Thermosinus carboxydivorans Nor1
23963030	VBITHiSp19295_1753	MTAP	Thioalkalivibrio sp. HL-EbGR7
32526054	VBITHiSp13812_1233	MTAP	Thioalkalivibrio sp. K90mix
23967725	VBITHiDen82923_0777	MTAP	Thiobacillus denitrificans ATCC 25259
29650470	VBIVeiDis1254_0157	MTAP	Veillonella dispar ATCC 17748
36458212	VBIVeiPar156829_1474	MTAP	Veillonella parvula ATCC 17745
32531194	VBIVeiPar80537_0927	MTAP	Veillonella parvula DSM 2008
36275462	VBIVeiSp18830_0337	MTAP	Veillonella sp. 3 1 44
36279337	VBIVeiSp6271_0334	MTAP	Veillonella sp. 6 1 27
32547531	VBIXanAlb89132_2491	MTAP	Xanthomonas albilineans
24054663	VBIXanAxo33670_1412	MTAP	Xanthomonas axonopodis pv. citri str. 306
			Xanthomonas campestris
24067511	VBIXanCam24967_3134	MTAP	pv. campestris str. 8004
			Xanthomonas campestris pv.
24073319	VBIXanCam115730_1376	MTAP	campestris str. ATCC 33913
			Xanthomonas campestris
24085960	VBIXanCam108527_3058	MTAP	pv. campestris str. B100
			Xanthomonas campestris
25699756	VBIXanCam81519_3238	MTAP	pv. musacearum NCPPB4381

26313008	VBIXanCam29727_1092	MTAP	Xanthomonas campestris pv. vasculorum NCPPB702
24092240	VBIXanCam71633_1615	MTAP	Xanthomonas campestris pv. vesicatoria str. 85-10
35684919	VBIXanFus21887_3741	MTAP	Xanthomonas fuscans subsp. aurantifolii str. ICPB 10535
35674697	VBIXanFus92710_3232	MTAP	Xanthomonas fuscans subsp. aurantifolii str. ICPB 11122
24102870	VBIXanOry111333_2072	MTAP	Xanthomonas oryzae pv. oryzae KACC10331
24113099	VBIXanOry49434_1979	MTAP	Xanthomonas oryzae pv. oryzae MAFF 311018
24125601	VBIXanOry73153_3157	MTAP	Xanthomonas oryzae pv. oryzae PXO99A
26326018	VBIXanOry69950_3062	MTAP	Xanthomonas oryzae pv. oryzicola BLS256
24134970	VBIXylFas578_2504	MTAP	Xylella fastidiosa 9a5c
26340625	VBIXylFas50652_2177	MTAP	Xylella fastidiosa Dixon
24139674	VBIXylFas124301_1785	MTAP	Xylella fastidiosa M12
24145269	VBIXylFas85937_1803	MTAP	Xylella fastidiosa M23
26353973	VBIXylFas52623_5708	MTAP	Xylella fastidiosa subsp. sandyi Ann-1
26344082	VBIXylFas52623_0929	MTAP	Xylella fastidiosa subsp. sandyi Ann-1
24150927	VBIXylFas71109_1769	MTAP	Xylella fastidiosa Temecula1

MTAP = 5'-methylthioadenosine phosphorylase; MENA = Menaquinone via futasoline step 4, possible alternative.

Description of Chemical Compounds

[00130] In one embodiment, as described in part in U.S. Patent No. 5,985,848 and in PCT International Patent Application Publication No. WO 99/19338, the contents of which are herein incorporated by reference, the MTIP inhibitor comprises a compound having formula (I):



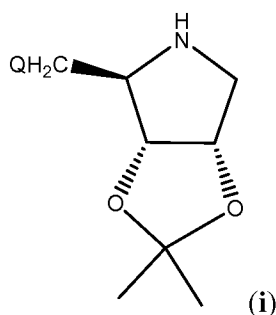
wherein A is CH or N; B is OH; D is chosen from H, OH, NH₂, or SCH₃; and X and Y are independently selected from H, OH or halogen, except that when one of X and Y is hydroxy or halogen, the other is hydrogen; Z is selected from CH₂SQ, CH₂OQ or Q, where Q is an optionally substituted alkyl, aralkyl, aralkenyl or aryl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino,

carboxy, or straight- or branched-chain C₁-C₆ alkyl; provided that Z is not CH₂OH; or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

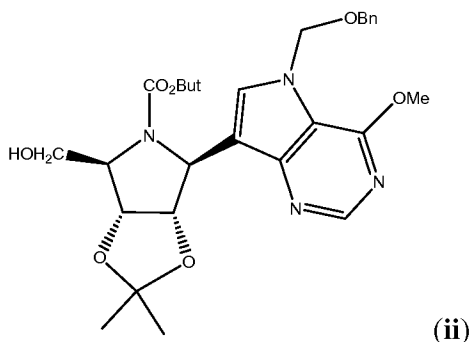
[00131] Methods to make compounds of formula (I) can be found in WO 99/19338 and in U.S. Patent No. 5,985,848.

[00132] Methods to make compounds of formula (I) in which X = OH and Y = H can be found in PCT International Patent Application Publication No. WO 00/061783 and in U.S. Patent No. 6,693,193, the contents of which are herein incorporated by reference.

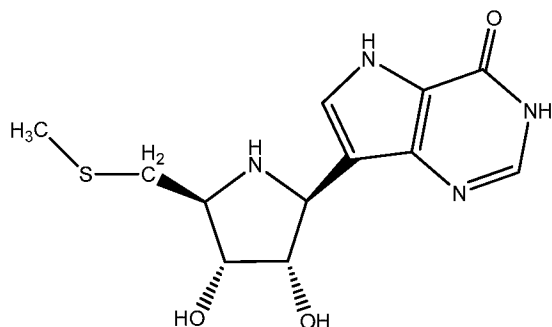
[00133] 1,4-Dideoxy-1,4-imino-2,3-O-isopropylidene-D-ribose can be converted into compounds of formula (i) below. Compounds of formula (i) can be converted into compounds of formula (I) by the methods provided in WO 00/061783 and in U.S. Patent No. 6,693,193.



[00134] The compound of Formula (ii) below can be converted into compounds of formula (I) where Z = Q by oxidation at C-5', Wittig reaction with the resulting aldehyde, optionally hydrogenating the double bond so formed, then removing the protecting groups by acid hydrolysis or a combination of acid hydrogenolysis and acid hydrolysis. Compound (ii) can be prepared by selectively removing the 5'-O-*tert*-butyldimethylsilyl protecting group of compound **40** in Evans et al, *J. Org. Chem.*, 66 (2001) 5723, e.g. by the use of tetrabutylammonium fluoride.



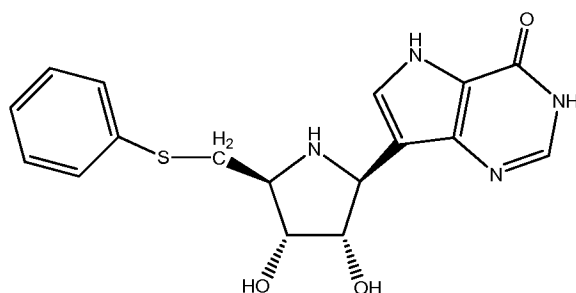
[00135] A method to make the compound



can be found in WO 99/19338 and in U.S.

Patent No. 5,985,848, where it was referred to as (1S)-1,4-dideoxy-1-C-(4-hydroxypyrrolo[3,2-d]pyrimidin-7-yl)-1,4-imino-5-methylthio-D-ribose. Subsequently, the compound was reported as an inhibitor of the PNP of *Plasmodium falciparum* and its method of synthesis was described (W. Shi et al, *J. Biol. Chem.*, 279 (2004) 18103) (33).

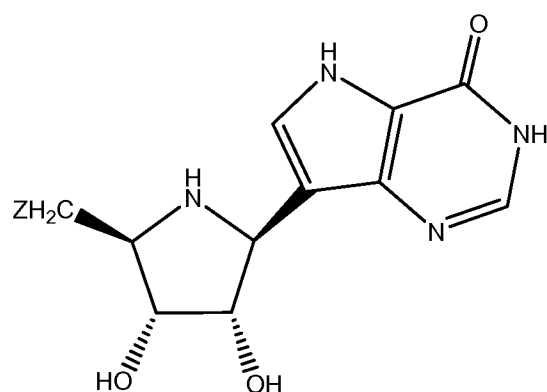
[00136] The compound



is reported in connection with its inhibition

of the human and *Plasmodium falciparum* PNP and a method of synthesis was described (A. Lewandowicz et al, *J. Biol. Chem.*, 280 (2005) 30320) (49).

[00137] The compound

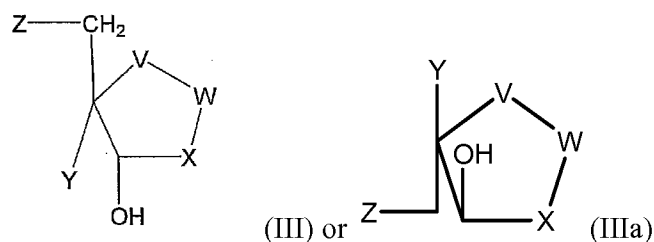


where Z = H is disclosed in A. Lewandowicz et

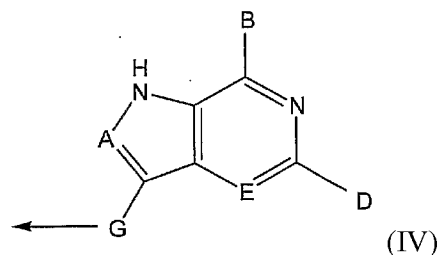
al, *J. Biol. Chem.*, 280 (2005) 30320 (49) as an inhibitor of PNP.

[00138] In another embodiment, as described in U.S. Patent No. 7,553,839 and in PCT International Patent Application Publication No. WO 04/018496, and in WO 2007/069924

the contents of which are herein incorporated by reference, the MTIP inhibitor comprises a compound having formula (III) or (IIIa):



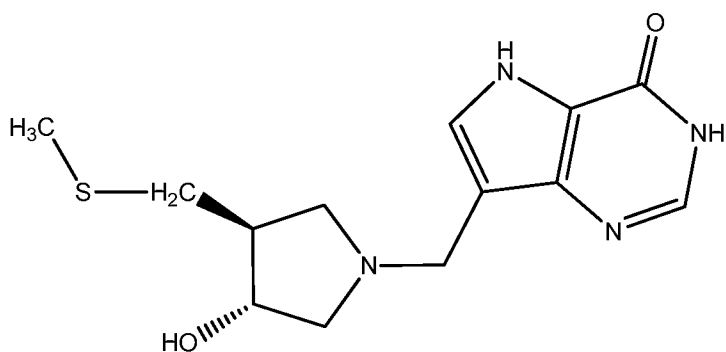
wherein V is CH₂ and W is NR¹; X is CH₂; Y is selected from hydrogen, halogen and hydroxyl; Z is selected from hydrogen, halogen, SQ, OQ and Q, where Q is an optionally substituted alkyl, aralkyl or aryl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; R¹ is a radical of the formula (IV)



A is selected from N, CH and CR, where R is selected from halogen or optionally substituted alkyl, aralkyl or aryl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; B is OH; D is selected from OH, NH₂, SCH₃ and hydrogen; E is N; G is CH₂; or a tautomer thereof or a pharmaceutically acceptable salt thereof; or an ester thereof.

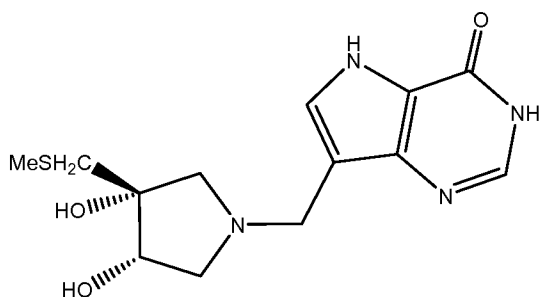
[00139] Methods to make compounds of formula (III) and (IIIa) are provided in U.S. Patent No. 7,553,839 and in WO 04/018496, and in WO 2007/069924, and also in US 7,655,795 and WO 2004/069856, the contents of all of which are herein incorporated by reference.

[00140] The compound



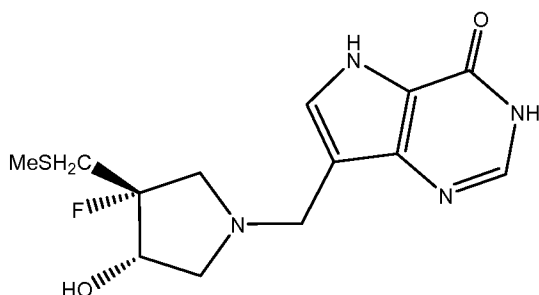
is disclosed in WO 2004/018496 and in U.S. Patent No. 7,553,839 as “(3R,4S)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(methylthiomethyl)pyrrolidine”, exemplified as compound 37 (Example 29) and shown to have PNP inhibitory properties. It is disclosed as (3R,4S)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(methylthiomethyl)-pyrrolidine in WO 2004/069856 and in US 7,655,795 as a compound that can be prepared by the process disclosed and claimed therein. The compound is also reported as an inhibitor of human and *Plasmodium falciparum* PNP and a method of synthesis is described (A. Lewandowicz et al, *J. Biol. Chem.*, 280 (2005) 30320) (49).

[00141] The compound



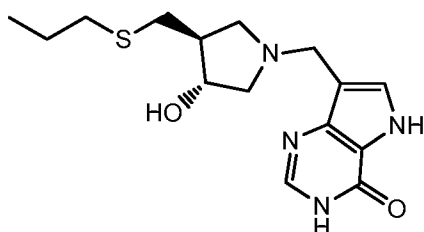
is disclosed in in WO 2004/018496 and in U.S. Patent No. 7,553,839, as “(3S,4R)-1-[(9-deazahypoxanthin-9-yl)methyl]-3,4-dihydroxy-4-methylthiomethylpyrrolidine”, exemplified as compound 33 (Example 28) and shown to have PNP inhibitory properties. This is an example where Y = OH. It is also reported in connection with its inhibition of the human and *Plasmodium falciparum* PNP and a method of synthesis is described (A. Lewandowicz et al, *J. Biol. Chem.*, 280 (2005) 30320) (49).

[00142] The compound



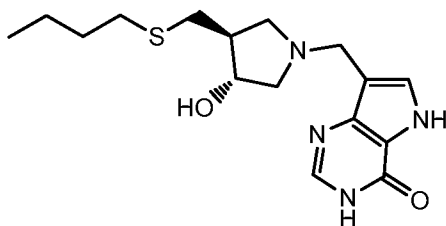
as a racemic mixture with its enantiomer is reported in connection with its inhibition of the human and mutated human PNP and a method of synthesis is described (A. Murkin et al, *Bioorg. Med. Chem. Lett.*, 18 (2008) 5900.

[00143] The compound



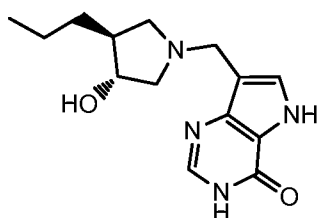
is disclosed in US 7,655,795 and WO 2004/069856 as “(3R,4S)-1-[(9-deazahypoxanthin-9-yl)-3-hydroxy-4-(propylthiomethyl)-pyrrolidine” and exemplified as compound 24 in Example 1.20 as a compound that can be prepared by the process disclosed and claimed therein. It is reported in connection with its inhibition of human and *Plasmodium falciparum* PNP and a method of synthesis was described (A. Lewandowicz et al, *J. Biol. Chem.*, 280 (2005) 30320) (49).

[00144] The compound:



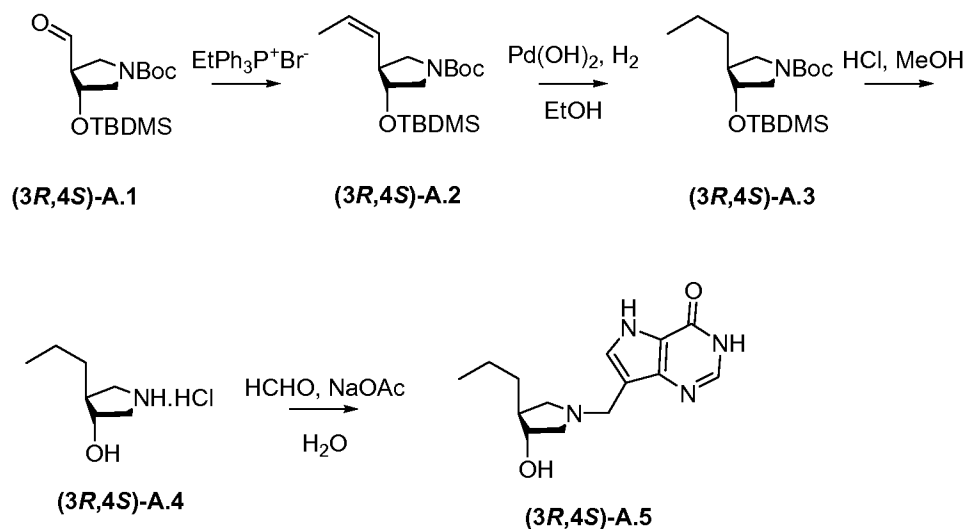
is disclosed in US 7,655,795 and WO 2004/069856 as “(3R,4S)-4-(butylthiomethyl)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxy-pyrrolidine”, and exemplified as compound 25 in Example 1.21.

[00145] The compound:



is disclosed as “(3R,4S)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-propylpyrrolidine”, compound 28 in Example 1.24 of in US 7,655,795 and WO 2004/069856.

[00146] This compound can also be prepared according to the following procedure:



[00147] *(3R,4S)-tert-Butyl 3-(tert-butyldimethylsilyloxy)-4-formylpyrrolidine-1-carboxylate (A.1)*. This compound is prepared as described in Longshaw, Alistair I.; Adanitsch, Florian; Gutierrez, Jemy A.; Evans, Gary B.; Tyler, Peter C.; Schramm, Vern L. Design and Synthesis of Potent "Sulfur-Free" Transition State Analogue Inhibitors of 5'-Methylthioadenosine Nucleosidase and 5'-Methylthioadenosine Phosphorylase. *Journal of Medicinal Chemistry* (2010), 53(18), 6730-6746.

[00148] *(3R,4S)-tert-Butyl 3-(tert-butyldimethylsilyloxy)-4-(propen-2-yl)pyrrolidine-1-carboxylate (A.2)*. *n*-Butyllithium (1.95 mL, 2.7 mmol, 1.4 M solution in hexanes) was added drop-wise to a stirred suspension of ethyltriphenylphosphonium bromide (2.03 g, 5.5 mmol) in THF (10 mL) at 0 °C. After 30 min a solution of **A.1** (977 mg, 3.0 mmol) in THF (10 mL) was added. The reaction mixture was stirred overnight and allowed to warm to room temperature. The reaction was quenched with water (30 mL) and extracted with ethyl acetate (100 mL). The combined organic phases were washed with saturated aqueous sodium hydrogen carbonate solution (50 mL) and brine (50 mL) and then dried (MgSO₄), filtered and concentrated in vacuo. Flash chromatography of the residue (1 : 9, EtOAc : Petrol) afforded

A.2 as a colorless oil (250 mg, 40%). ^1H NMR (500 MHz, CDCl_3): δ = 5.59 (m, 1H), 5.17 (m, 2H), 3.95 (m, 1H), 3.66 - 3.48 (m, 2H), 3.17 - 2.32 (m, 2H), 1.67 (d, J = 6.6 Hz, 2H), 1.46 (s, 9H), 0.87 (s, 9H) and 0.04 ppm (s, 6H).

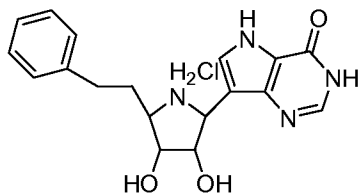
[00149] *3-(tert-butyltrimethylsilyloxy)-4-propylpyrrolidine-1-carboxylate (A.3).*

Perlman's catalyst (75 mg, 0.9 mmol, 20 wt% on carbon) was added to a solution of **A.2** (221 mg, .65 mmol) in ethanol (5 mL) under an argon atmosphere. The reaction mixture was placed under a hydrogen atmosphere and stirred overnight and then filtered through Celite[®] and concentrated under reduced pressure to afford the title compound **A.3** (.220 mg, 99%). NMR confirmed that no olefinic protons remained and that no further purification was necessary.

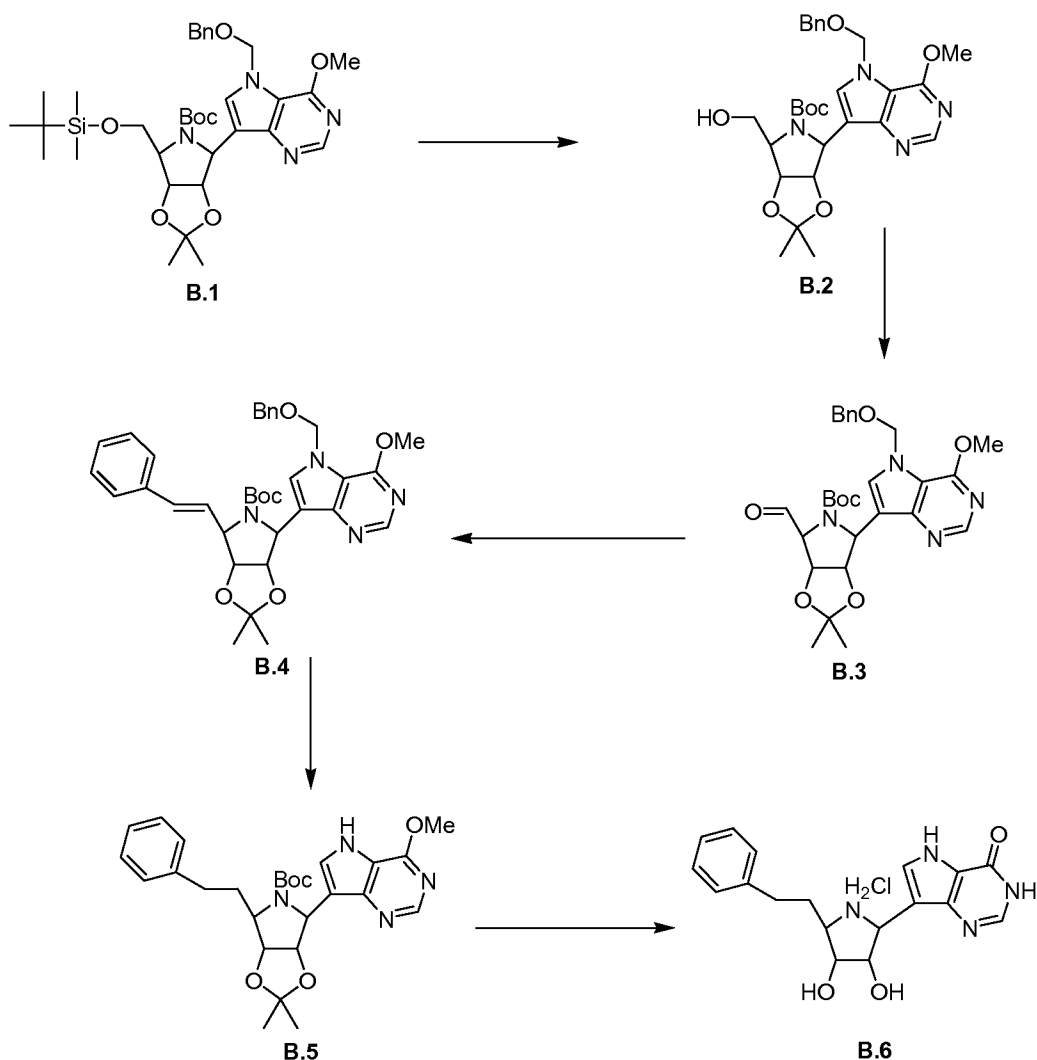
[00150] *(3R,4S)-4-Propyl-3-hydroxypyrrolidine (A.4).* Concentrated HCl (1 ml, 12 mmol) was added to a solution of **A.3** (.22 g, 0.62 mmol) in methanol (5 ml) and concentrated in vacuo. The resulting residue was dissolved in additional concentrated HCl (1 ml, 12 mmol) and concentrated in vacuo. The residue was dissolved in water (10 ml) and washed with CHCl_3 (2 x 20 ml) and the water layer concentrated in vacuo to afford **A.4**. ^{13}C NMR (125 MHz, D_2O): δ = 73.4, 51.1, 48.9, 45.0, 32.6, 20.2 and 13.3 ppm.

[00151] *(3R,4S)-1-[(9-Deazahypoxanthin-9-yl)methyl]-4-propyl-3-hydroxypyrrolidine (A.5).* 9-Deazahypoxanthine (0.147 g, 1.09 mmol) was added to a solution of **A.4** (.090 g, 0.54 mmol) and sodium acetate (0.089 g, 1.09 mmol) in water (2 ml). After 5 min aqueous formaldehyde (0.084 ml, 1.087 mmol) was added and the resulting suspension warmed to 95 °C bath temperature and the resulting mixture stirred overnight. The reaction mixture was then cooled to room temperature and absorbed onto silica gel. The resulting residue was purified by chromatography eluting with 25% 7N NH_3 in $\text{MeOH}:\text{CHCl}_3$ to afford **A.5** (90 mg, 60%) as a white solid. ^{13}C NMR (125 MHz, D_2O): δ = 163.5, 151.7, 145.4, 127.7, 118.8, 109.7, 76.2, 59.9, 57.1, 47.3, 46.1, 34.6, 20.6, and 13.4 ppm.

[00152] The compound:



is prepared according to the following procedure:



[00153] To a solution of **B.1** (1.3 g, 1.985 mmol) in THF (30 mL) was added acetic acid (0.114 mL, 1.985 mmol) and then TBAF (tetrabutyl ammonium fluoride 1M THF) (1.730 mL, 5.96 mmol). The solution was stirred at ~ 50-60 °C for 16 h. After evaporation, toluene was added and the solution was washed water, dried (MgSO₄) and concentrated. Chromatography (25-50% EtOAc/Hex) gave **B.2** as a foam (1.0 g, 1.85 mmol, 93%).

[00154] To a stirred solution of **B.2** (100 mg, 185 μmol) in dry DCM (3 mL) was added Dess-Martin periodinane (157 mg, 370 μmol). After 16 h the mixture was concentrated to dryness. Chromatography (20-60% EtOAc/Hex) gave **B.3** as a syrupy product (58 mg, 108 μmol, 58%).

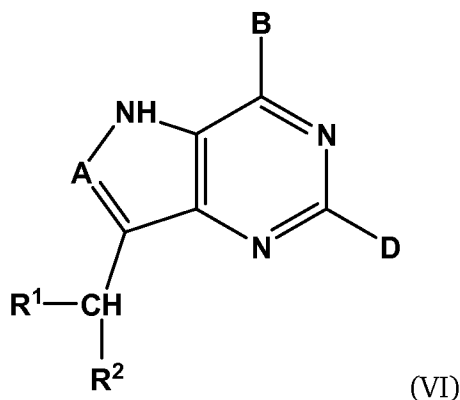
[00155] A 1M solution of lithium bis(trimethylsilyl)amide in THF (204 μL, 204 μmol) was added to a suspension of benzyl triphenylphosphonium bromide (97 mg, 225 μmol) in dry THF (3 mL) at -75 °C and then the mixture was allowed to warm to 0 °C. After 30 mins at 0 °C a solution of **B.3** (55 mg, 102 μmol) in THF (1 mL) was added and then the mixture

was stirred at RT. After 30 mins chloroform was added and the mixture was washed with water, dried (MgSO₄) and concentrated. Chromatography of the residue (15-40% EtOAc/Hex) gave **B.4** as a syrup (59 mg 96 μ mol, 94%).

[00156] To a solution of **B.4** (55 mg, 90 μ mol) in EtOH and some NH₃/MeOH was added 10% Pd/C and the mixture was stirred under H₂. After 2 days the mixture was filtered and the filtrate concentrated. Chromatography of the residue (20-60% EtOAc/Hex) gave **B.5** as a syrup (25 mg, 50 μ mol, 56%).

[00157] A solution of **B.5** (25 mg, 50.5 μ mol) in MeOH (4 mL) and conc aq HCl (4 mL) was heated under reflux for 3 h and then concentrated to dryness. The residue in water was lyophilized to a white powder of **B.6** (18 mg, 48 μ mol, 94%). ¹H nmr (d⁴MeOH with a little DCl/D₂O) δ 9.1 (1H, s), 8.1 (1H, s), 7.31 (4H, m), 7.20 (1H, m), 4.96 (1H, d), 4.73 (1H, dd), 4.30 (1H, t), 3.68 (1H, m), 2.84 (2H, m), 2.27 (2H, m). ¹³C nmr δ 152.5, 146.7, 141.7, 132.5, 131.9, 129.7, 127.4, 119.8, 106.5, 74.1, 73.8, 66.6, 57.3, 34.0, 33.5. HRMS calc for MH⁺ C₁₈H₂₁N₄O₃ 341.1614; found 341.1618.

[00158] In another embodiment, as described in PCT International Patent Application Publication No. WO 2008/030119, the contents of which are herein incorporated by reference, the MTIP inhibitor comprises a compound having formula (VI):

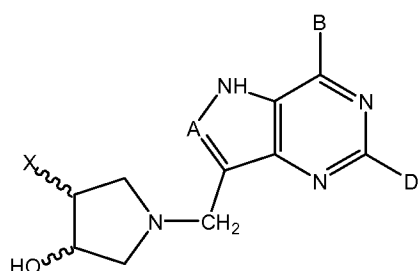


wherein R¹ is H or NR³R⁴; R² is H or an alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group, each of which is optionally substituted with one or more hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, or NR³R⁴ groups, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; provided that when R¹ is H, R² is an alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group which is substituted with at least one NR³R⁴ group, and optionally substituted with one or more hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid,

carboxylate alkyl ester or nitro group; R^3 and R^4 , independently of each other, are H or an alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group, each of which is optionally substituted with one or more hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, or NR^3R^4 groups, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; A is N or CH; B is OH; and D is H, OH, NH_2 , or SCH_3 ; or a tautomer thereof, or a pharmaceutically acceptable salt thereof, or an ester prodrug form thereof.

[00159] Methods to make compounds of formula (VI) are provided in WO 08 030119. Certain of the amino-alcohols required for use in the synthesis of the compounds of formula (VI) can be obtained through reduction of amino-acids to the corresponding amino-alcohols.

[00160] In another embodiment, the MTIP inhibitor comprises a compound having formula (VII):



(VII)

wherein X is an alkyl, cycloalkyl, aralkyl, aralkenyl, alkenyl, alkynyl or aryl group, each of which is optionally substituted with one or more substituents selected from the group consisting of hydroxy, alkoxy, cycloalkyl, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, cyano, thiazole and NR^2R^3 group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; or X is SR^1 ; or X is NR^2R^3 ; R^1 , R^2 and R^3 are independently selected from the group consisting of alkyl, alkenyl, alkynyl, aralkyl or aryl, each of which is optionally substituted with one or more substituents selected from the group consisting of hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, cyano and NR^2R^3 group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; A is N or CH; B is OH; D is H, OH, NH_2 , or SCH_3 ; provided that X is not CH_2Z , where Z is selected from OH, hydrogen, halogen, SQ^1 , OQ^2 and Q^3 , where Q^1 is an optionally substituted alkyl, aralkyl or aryl group, Q^2 is an optionally substituted alkyl group

and Q³ is an optionally substituted alkyl group; or a tautomer thereof, or a pharmaceutically acceptable salt thereof, or an ester prodrug form thereof.

[00161] Methods to make compounds of formula (VII) are provided in U.S. Patent No. 7,553,839 and in WO 04/018496, the contents of all of which are herein incorporated by reference, where a 3-hydroxypyrrolidine is coupled to 9-deaza9-formylpurine derivative by reductive amination, and in US Patent No. 7,655,795 and WO 2004/069856, the contents of all of which are herein incorporated by reference, where a 3-hydroxypyrrolidine is coupled to 9-deazapurine and formaldehyde in a Mannich reaction. Suitable 3-hydroxypyrrolidines may be prepared by methods detailed in PCT/NZ2010/000148 and in A. Longshaw et al., J. Med. Chem., 53 (2010) 6730.

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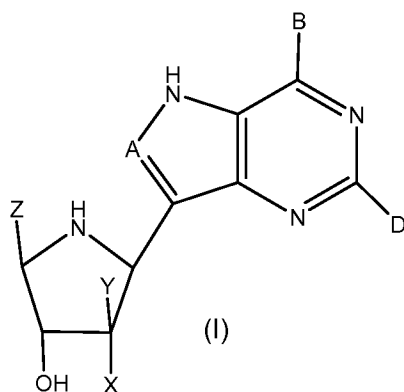
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What is claimed is:

1. A method for treating an infection caused by bacteria that use 5'-methylthioinosine phosphorylase (MTIP) in a quorum sensing pathway comprising administering to a subject having the infection a sub-growth inhibiting amount of a 5'-methylthioinosine phosphorylase (MTIP) inhibitor.
2. The method of Claim 1, wherein the bacteria is *Pseudomonas aeruginosa*, *Pseudomonas syringae* or *Xanthomonas campestris*.
3. The method of Claim 1 or 2, wherein the subject is an animal
4. The method of Claim 3, wherein the subject is a human.
5. The method of Claim 1 or 2, wherein the subject is a plant.
6. The method of Claim 1, wherein the bacteria is *Pseudomonas aeruginosa* and the subject is a human.
7. The method of Claim 1, wherein the bacteria is *Pseudomonas syringae* or *Xanthomonas campestris* and the subject is a plant.
8. The method of any of Claims 1-7, wherein the MTIP inhibitor comprises a compound having formula (I):



wherein

A is CH or N;

B is OH;

D is chosen from H, OH, NH₂, or SCH₃;

X and Y are independently selected from H, OH or halogen, except that when one of X and Y is hydroxy or halogen, the other is hydrogen;

Z is selected from CH₂SQ, CH₂OQ or Q, where Q is an optionally substituted alkyl, aralkyl, aralkenyl or aryl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; provided that Q is not CH₂OH;

or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof; or a prodrug thereof.

9. The method of Claim 8, wherein A is CH.

10. The method of Claims 8 or 9, wherein D is H.

11. The method of any of Claims 8-10, wherein X is OH and Y is H.

12. The method of any of Claims 8-11, wherein Z is CH₂SQ or Q.

13. The method of any of Claims 8-12, where Z is CH₂SQ, and Q is an optionally substituted C₁-C₆ alkyl group, aryl group or aralkyl group.

14. The method of any of Claims 8-12, where Z is Q, and Q is an optionally substituted C₁-C₆ alkyl group, aralkyl group, or aralkenyl group.

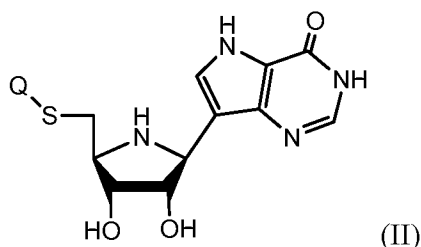
15. The method of any of Claims 8-11, wherein Z is phenylthiomethyl, p-chlorophenylthiomethyl, methylthiomethyl, ethylthiomethyl, propylthiomethyl, n-propylthiomethyl or isopropylthiomethyl, butylthiomethyl, -CH₂S(CH₂)₂CHNH₂COOH, methyl, ethyl, propyl, butyl, benzyl, phenylethenyl or -CH₂(CH₂)₃CH(NH₂)COOH.

.

16. The method of Claim 8, wherein A is CH or N; B is OH; D is chosen from H, OH, NH₂, or SCH₃; and X is OH; Y is H; Z is Q, where Q is an optionally substituted alkyl,

alkenyl, aralkyl or aryl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

17. The method of Claim 8, wherein the MTIP inhibitor comprises a compound having formula (II):



wherein Q is an optionally substituted alkyl, aralkyl or aryl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl;

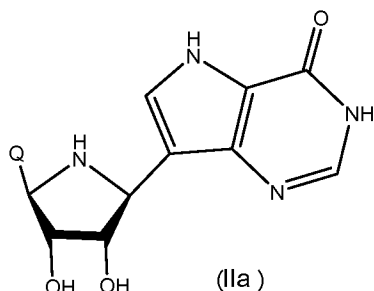
or a tautomer thereof; or a pharmaceutically acceptable salt thereof, or an ester thereof; or a prodrug thereof.

18. The method of Claim 17, wherein Q is a C₁-C₆ alkyl group or an aryl group.

19. The method of Claim 17, wherein Q is methyl.

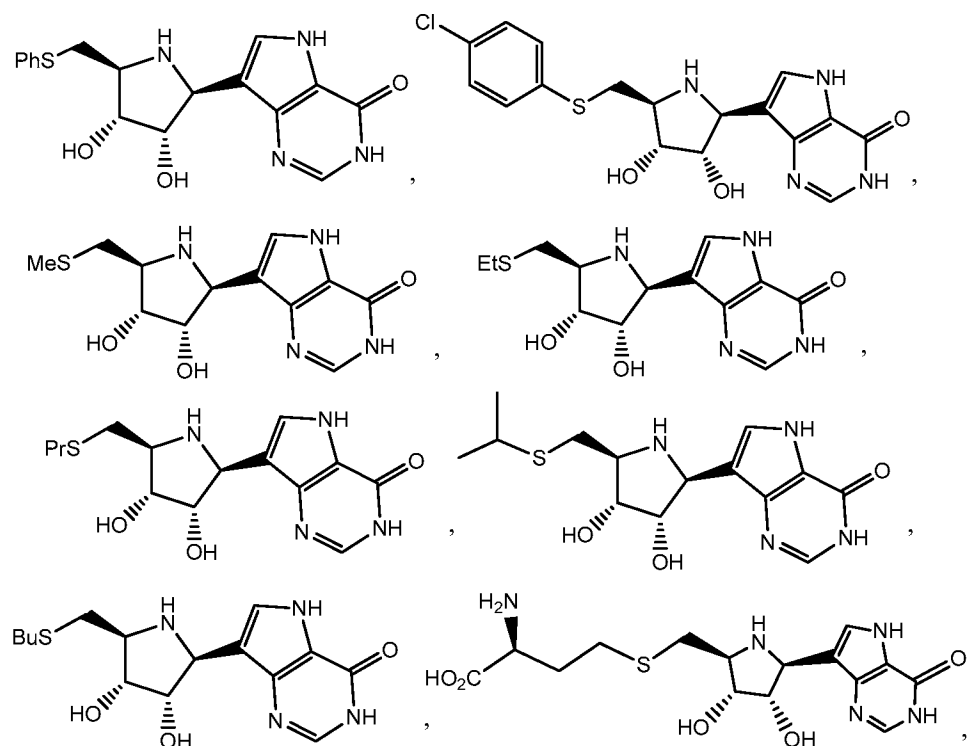
20. The method of Claim 17, wherein Q is phenyl.

21. The method of Claim 8, wherein wherein the MTIP inhibitor comprises a compound having formula (IIa):

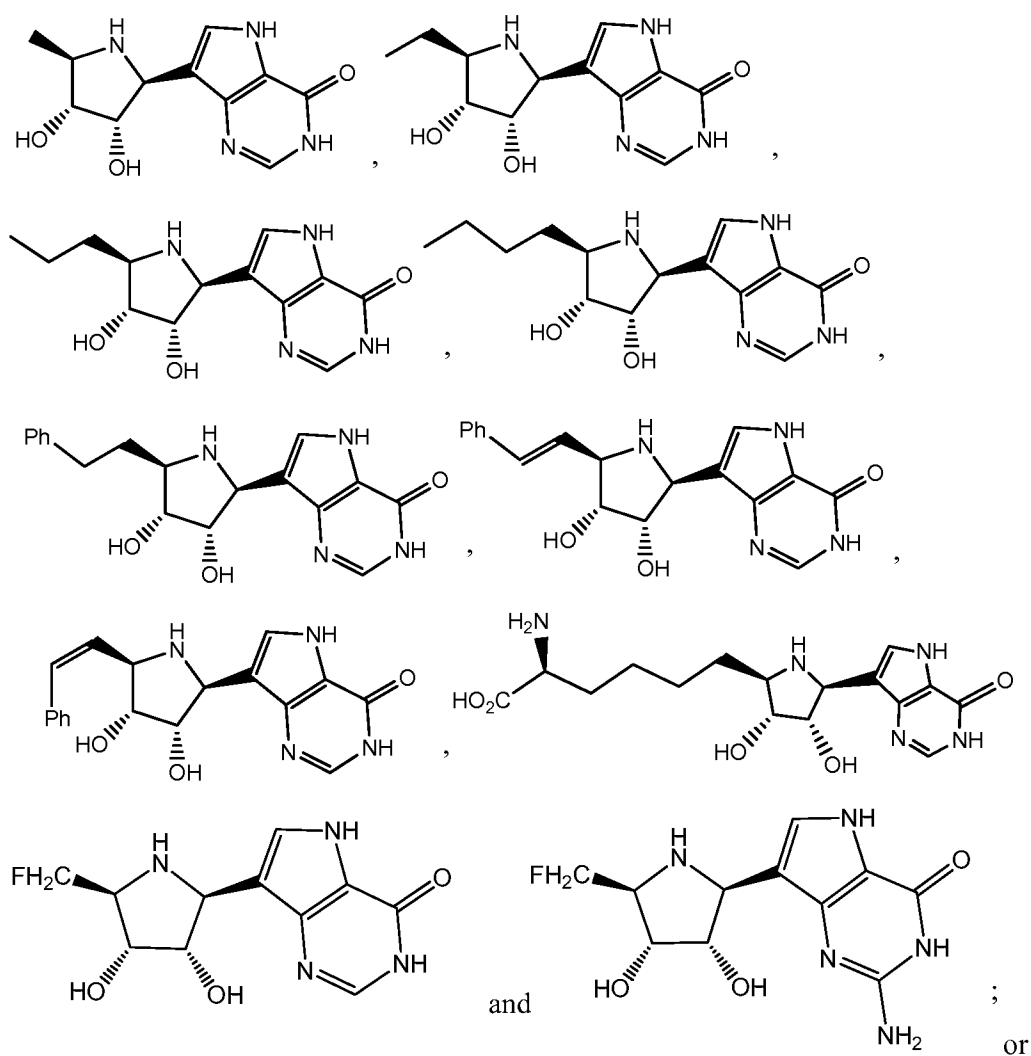


wherein Q is alkyl, alkenyl, aralkyl, aralkenyl or aryl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; or a tautomer thereof; or a pharmaceutically acceptable salt thereof, or an ester thereof.

22. The method of Claim 8, wherein the MTIP inhibitor comprises a compound having the structure selected from the group consisting of:

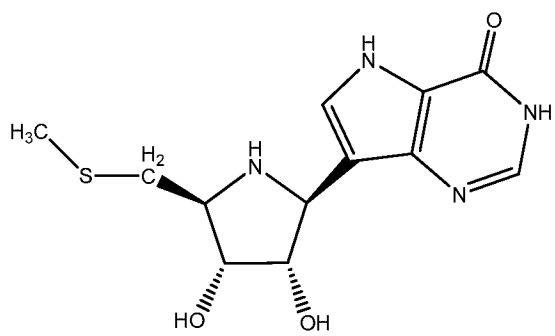


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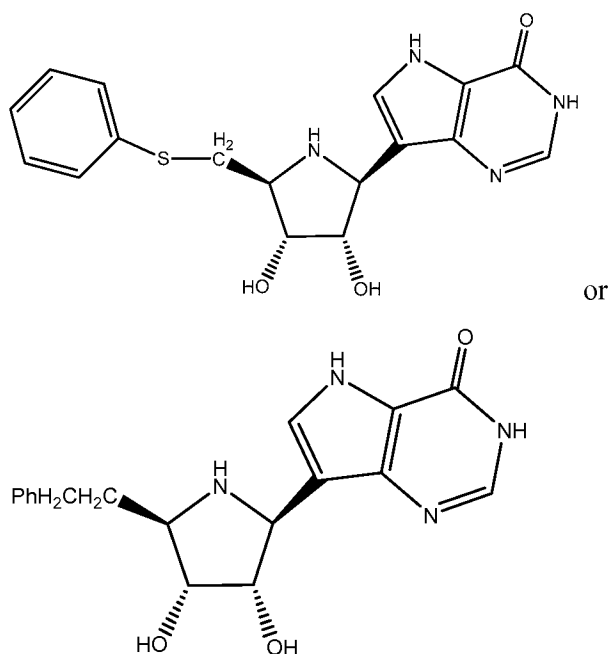


a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

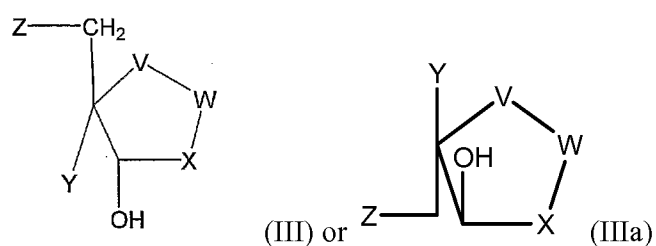
23. The method of Claim 8, wherein the MTIP inhibitor comprises a compound having the structure:



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24. The method of any of Claims 1-7, wherein the MTIP inhibitor comprises a compound having formula (III) or formula (IIIa):



wherein

V is CH₂;

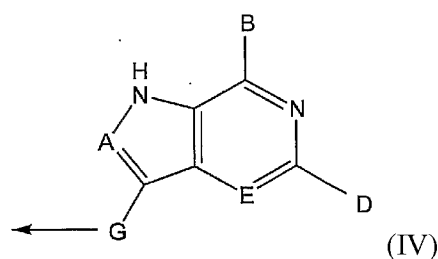
W is NR¹;

X is CH₂;

Y is selected from hydrogen, halogen and hydroxyl;

Z is selected from hydrogen, halogen, SQ, OQ and Q, where Q is an optionally substituted alkyl, aralkyl or aryl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl;

R¹ is a radical of the formula (IV)



A is selected from N, CH and CR, where R is selected from halogen or optionally substituted alkyl, aralkyl or aryl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl;

B is OH;

D is selected from OH, NH₂, SCH₃ and hydrogen;

E is N;

G is CH₂;

or a tautomer thereof or a pharmaceutically acceptable salt thereof; or an ester thereof.

25. The method of Claim 24, wherein Y is H.

26. The method of Claim 24 or 25, wherein A is CH.

27. The method of any of Claims 24-26, wherein D is H.

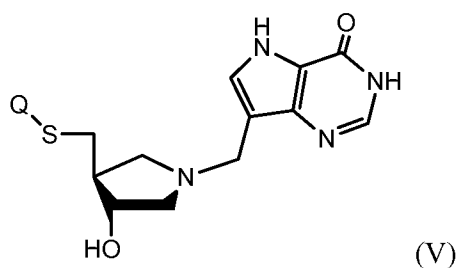
28. The method of any of Claims 24-27, wherein Z is SQ.

29. The method of any of Claims 24-28, wherein Q is an optionally substituted aryl group or aralkyl group, or Q is an optionally substituted C₁-C₆ alkyl group.

30. The method of any of Claims 24-29, wherein Q is methyl.

31. The method of any of Claims 24-29, wherein Q is phenyl.

32. The method of Claim 24, wherein the MTIP inhibitor comprises a compound having formula (V):



where Q is aryl, aralkyl or alkyl, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, and straight- or branched-chain C₁-C₆ alkyl;

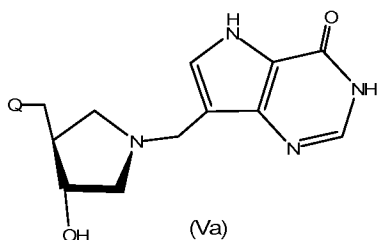
or a tautomer thereof; or an ester thereof; or a pharmaceutically acceptable salt thereof.

33. The method of Claim 32, wherein Q is an optionally substituted aryl group or aralkyl group, or Q is an optionally substituted C₁-C₆ alkyl group.

34. The method of Claim 33, wherein Q is methyl.

35. The method of Claim 33, wherein Q is phenyl.

36. The method of Claim 24, wherein the MTIP inhibitor comprises a compound having formula (Va):

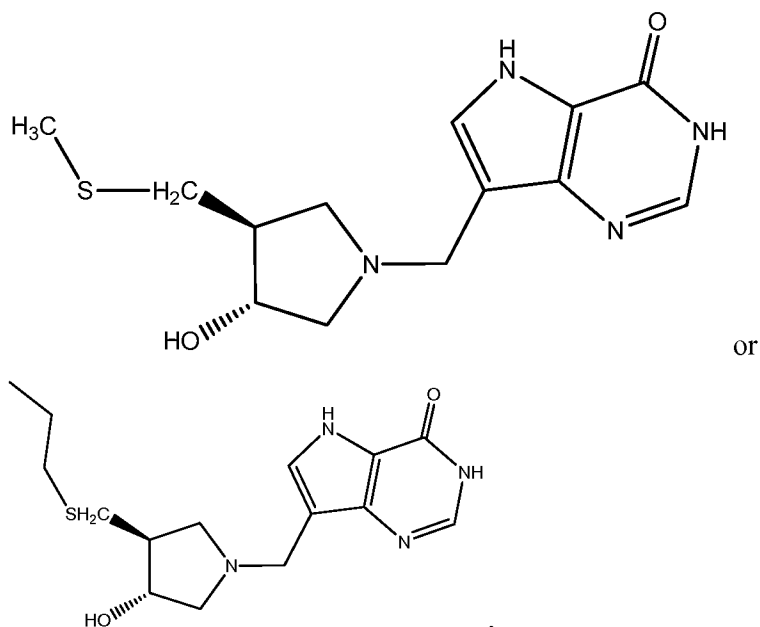


where Q is aryl, aralkyl or alkyl, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, and straight- or branched-chain C₁-C₆ alkyl; or a tautomer thereof; or an ester thereof; or a pharmaceutically acceptable salt thereof, or a prodrug thereof.

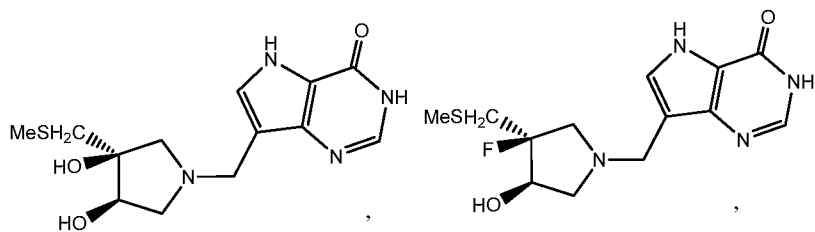
37. The method of Claim 24, wherein the MTIP inhibitor comprises a compound having the structure selected from the group consisting of:

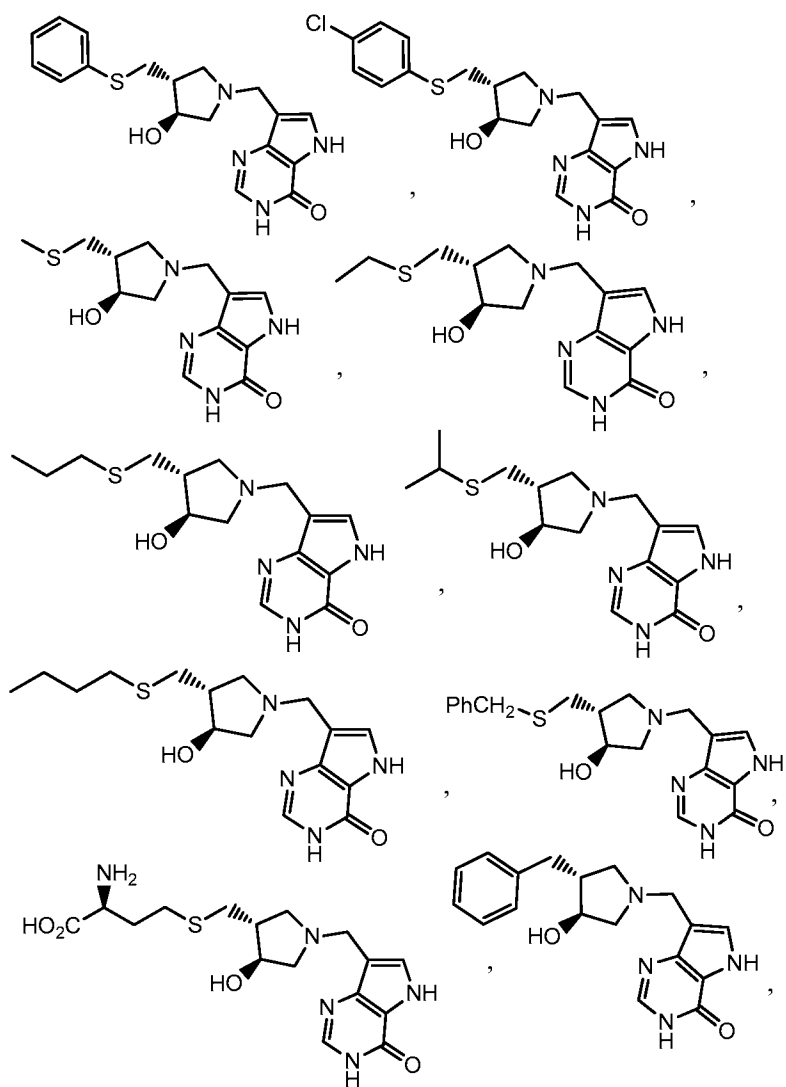
or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

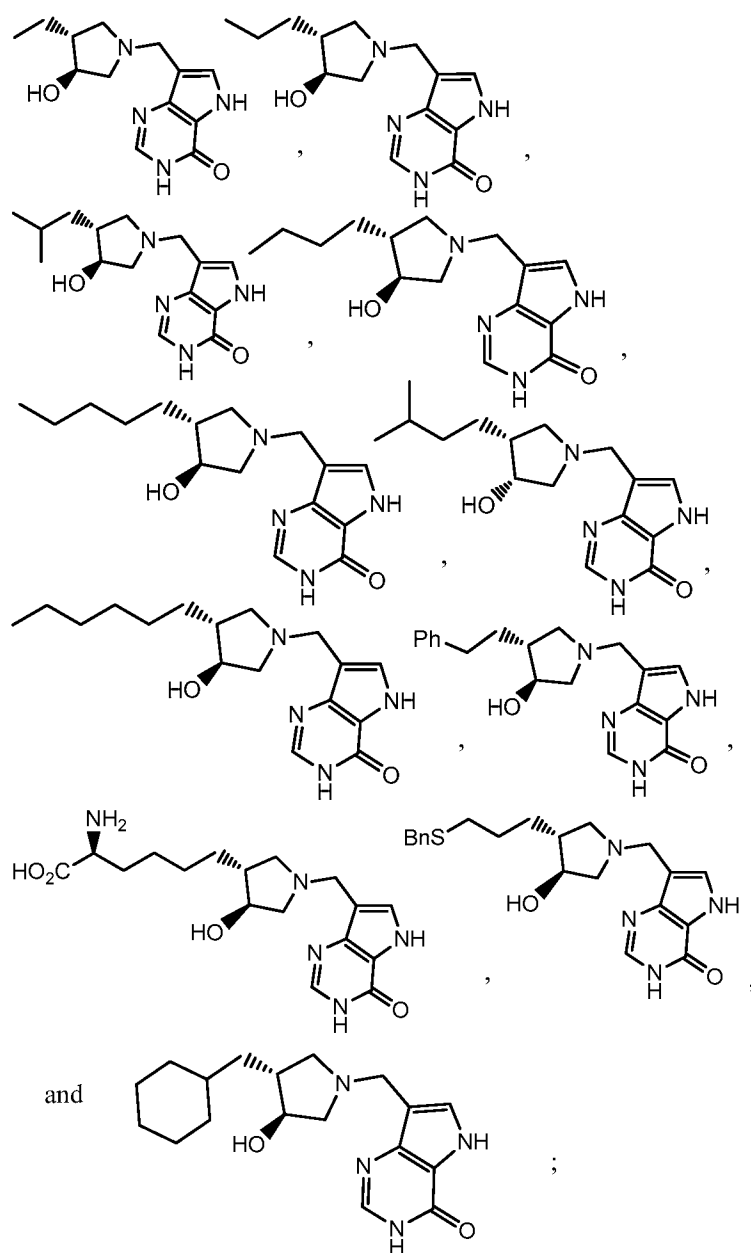
38. The method of Claim 24, wherein the the MTIP inhibitor comprises a compound having the structure:



39. The method of Claim 24, wherein the MTIP inhibitor comprises a compound having the structure selected from the group consisting of:

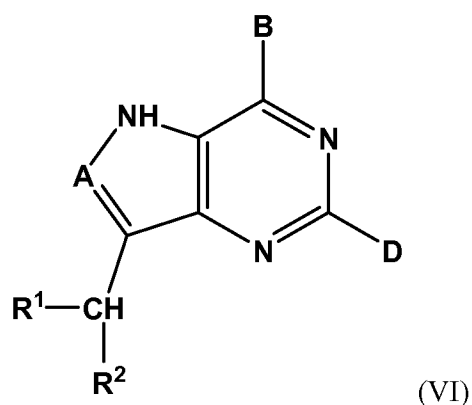






or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

40. The method of any of Claims 1-7, wherein the MTIP inhibitor comprises a compound having formula (VI):



wherein

R^1 is H or NR^3R^4 ;

R^2 is H or an alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group, each of which is optionally substituted with one or more hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, or NR^3R^4 groups, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups;

provided that when R^1 is H, R^2 is an alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group which is substituted with at least one NR^3R^4 group, and optionally substituted with one or more hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester or nitro group;

R^3 and R^4 , independently of each other, are H or an alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group, each of which is optionally substituted with one or more hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, or NR^3R^4 groups, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups;

A is N or CH;

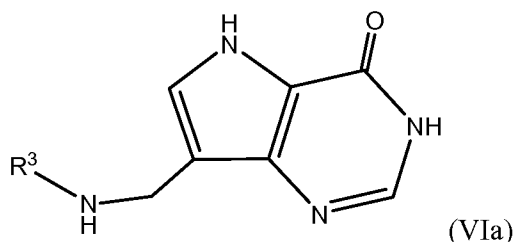
B is OH; and

D is H, OH, NH_2 , or SCH_3 ;

or a tautomer thereof, or a pharmaceutically acceptable salt thereof, or an ester prodrug form thereof.

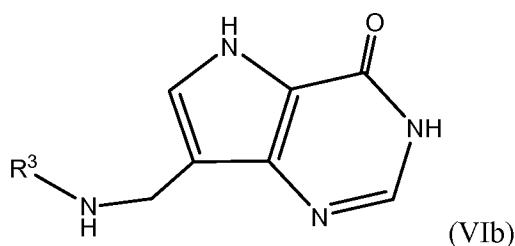
41. The method of Claim 40, wherein A is CH.

42. The method of Claim 40 or 41, wherein D is H.
43. The method of any of Claims 40-4, wherein R^1 is NR^3R^4 .
44. The method of any of Claims 40-43, wherein R^2 is H.
45. The method of Claim 40, wherein the MTIP inhibitor comprises a compound having formula (VIa):



wherein R^3 is an alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group, each of which is optionally substituted with one or more hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, or nitro group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; or a tautomer thereof, or a pharmaceutically acceptable salt thereof, or an ester prodrug form thereof.

46. The method of Claim 40, wherein the MTIP inhibitor comprises a compound having formula (VIb):



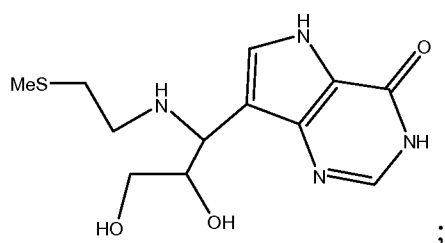
wherein R^3 is an alkyl, alkenyl, alkynyl, aralkyl, aralkenyl, aralkynyl, or aryl group, each of which is substituted with at least one alkoxy, thiol, alkylthio, arylthio, or aralkylthio group and optionally substituted with one or more hydroxy, halogen, carboxylic acid, carboxylate alkyl ester, or nitro group, where each alkylthio, arylthio and aralkylthio group is optionally

substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; or a tautomer thereof, or a pharmaceutically acceptable salt thereof, or an ester prodrug form thereof.

47. The method of Claim 40, wherein the MTIP inhibitor comprises a compound selected from the group consisting of:

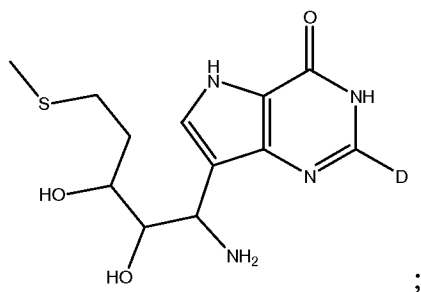
2-amino-7-(2,3-dihydroxy-1-(2-(methylthio)ethylamino)propyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(2,3-dihydroxy-1-(2-(methylthio)ethylamino)propyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



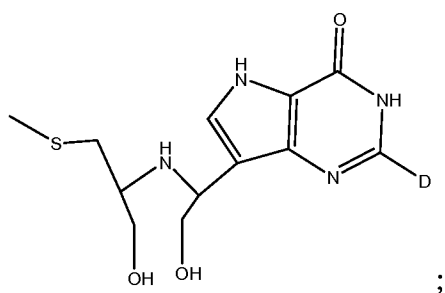
2-amino-7-(1-amino-2,3-dihydroxy-5-(methylthio)pentyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(1-amino-2,3-dihydroxy-5-(methylthio)pentyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



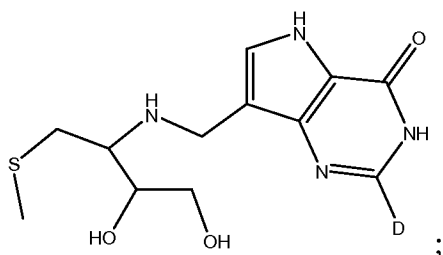
2-amino-7-(2-hydroxy-1-(1-hydroxy-3-(methylthio)propan-2-ylamino)ethyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(2-hydroxy-1-(1-hydroxy-3-(methylthio)propan-2-ylamino)ethyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



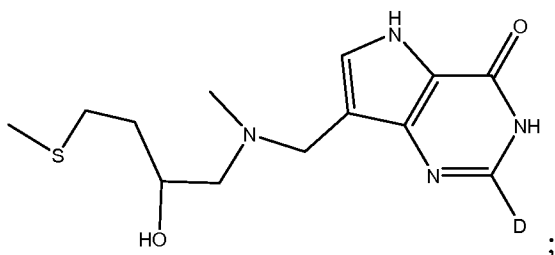
2-amino-7-((3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-((3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



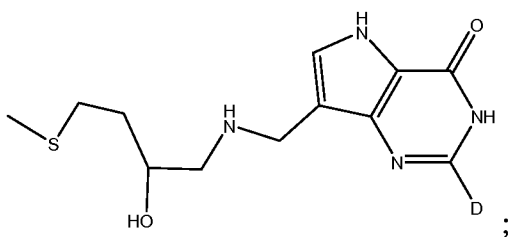
2-amino-7-(((2-hydroxy-4-(methylthio)butyl)(methyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(((2-hydroxy-4-(methylthio)butyl)(methyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



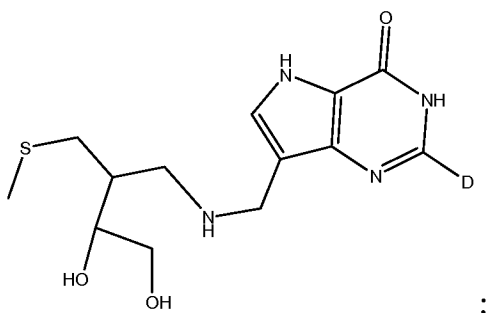
2-amino-7-((2-hydroxy-4-(methylthio)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-((2-hydroxy-4-(methylthio)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



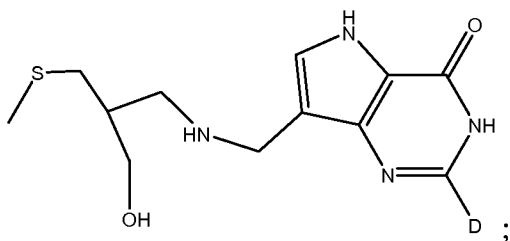
2-amino-7-((3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-((3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



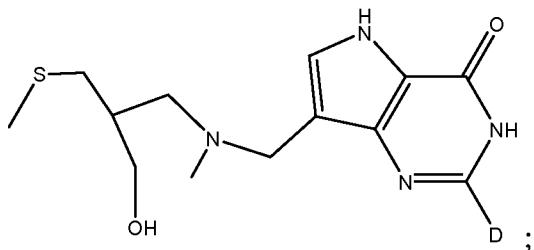
2-amino-7-((3-hydroxy-2-(methylthiomethyl)propylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-((3-hydroxy-2-(methylthiomethyl)propylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



7-(((3-hydroxy-2-(methylthiomethyl)propyl)(methyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

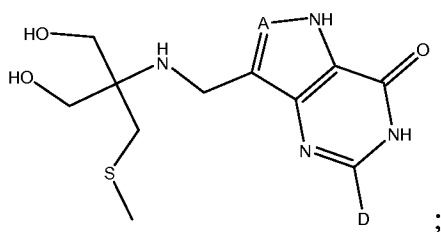
2-amino-7-(((3-hydroxy-2-(methylthiomethyl)propyl)(methyl)amino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



7-((1,3-dihydroxy-2-(methylthiomethyl)propan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-((1,3-dihydroxy-2-(methylthiomethyl)propan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

5-amino-3-((1,3-dihydroxy-2-(methylthiomethyl)propan-2-ylamino)methyl)-1H-pyrazolo[4,3-d]pyrimidin-7(6H)-one;



7-((benzyl((2*R*,3*R*)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

7-((benzyl((2*S*,3*S*)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

7-((benzyl((2*R*,3*S*)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

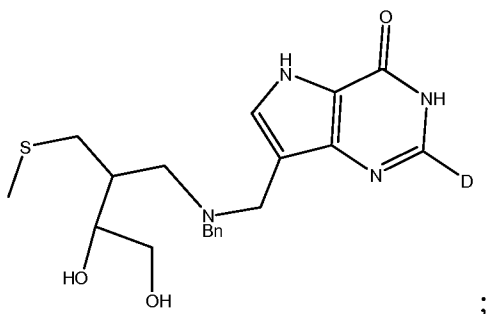
7-((benzyl((2*S*,3*R*)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-((benzyl((2*R*,3*R*)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-((benzyl((2*S*,3*S*)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-((benzyl((2*R*,3*S*)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-((benzyl((2*S*,3*R*)-3,4-dihydroxy-2-(methylthiomethyl)butyl)amino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;



7-(((2*R*,3*R*)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

7-(((2*S*,3*S*)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

7-(((2*R*,3*S*)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

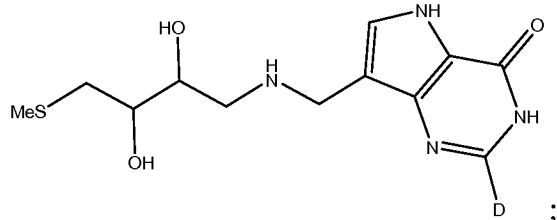
7-(((2S,3R)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-(((2R,3R)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-(((2S,3S)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-(((2R,3S)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-(((2S,3R)-2,3-dihydroxy-4-(methylthio)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;



7-(((2R,3R)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(((2S,3S)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(((2R,3S)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

7-(((2S,3R)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-(((2R,3R)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-(((2S,3S)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

2-amino-7-(((2R,3S)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

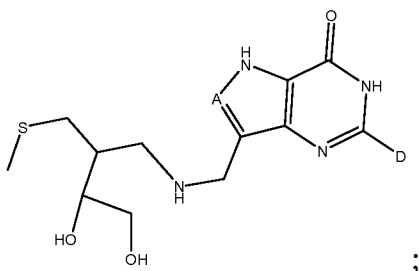
2-amino-7-(((2S,3R)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

3-(((2R,3R)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-1H-pyrazolo[4,3-d]pyrimidin-7(6H)-one;

3-(((2*S*,3*S*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;

3-(((2*R*,3*S*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;

3-(((2*S*,3*R*)-3,4-dihydroxy-1-(methylthio)butan-2-ylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;



3-(((2*R*,3*R*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;

3-(((2*S*,3*S*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;

3-(((2*R*,3*S*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;

3-(((2*S*,3*R*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-1*H*-pyrazolo[4,3-*d*]pyrimidin-7(6*H*)-one;

2-amino-7-(((2*R*,3*R*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-(((2*S*,3*S*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

2-amino-7-(((2*R*,3*S*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

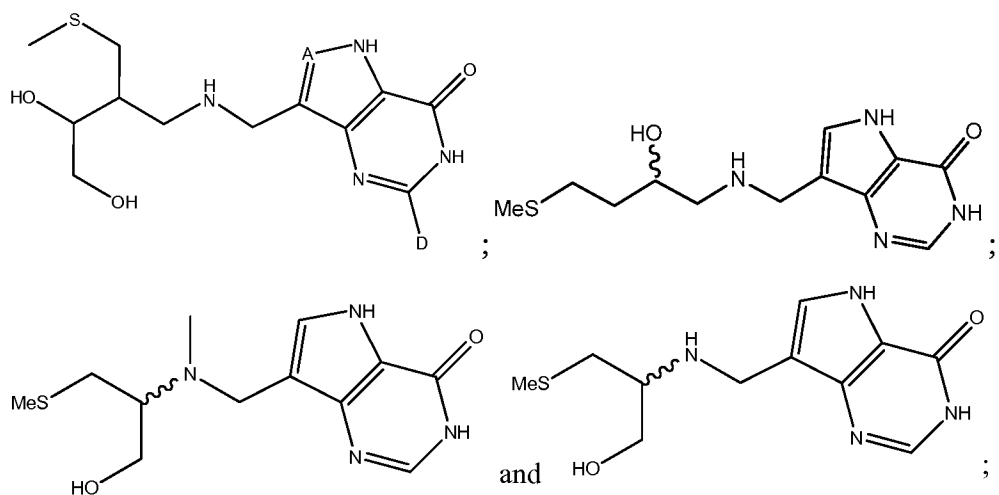
2-amino-7-(((2*S*,3*R*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

7-(((2*R*,3*R*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

7-(((2*S*,3*S*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

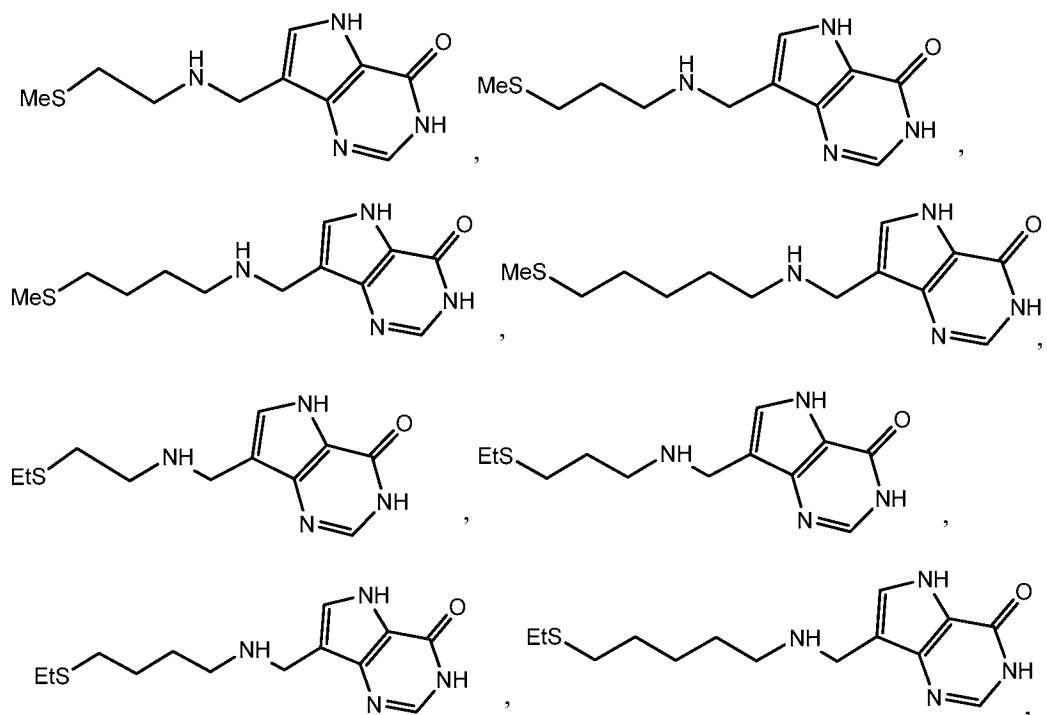
7-(((2*R*,3*S*)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3*H*-pyrrolo[3,2-*d*]pyrimidin-4(5*H*)-one;

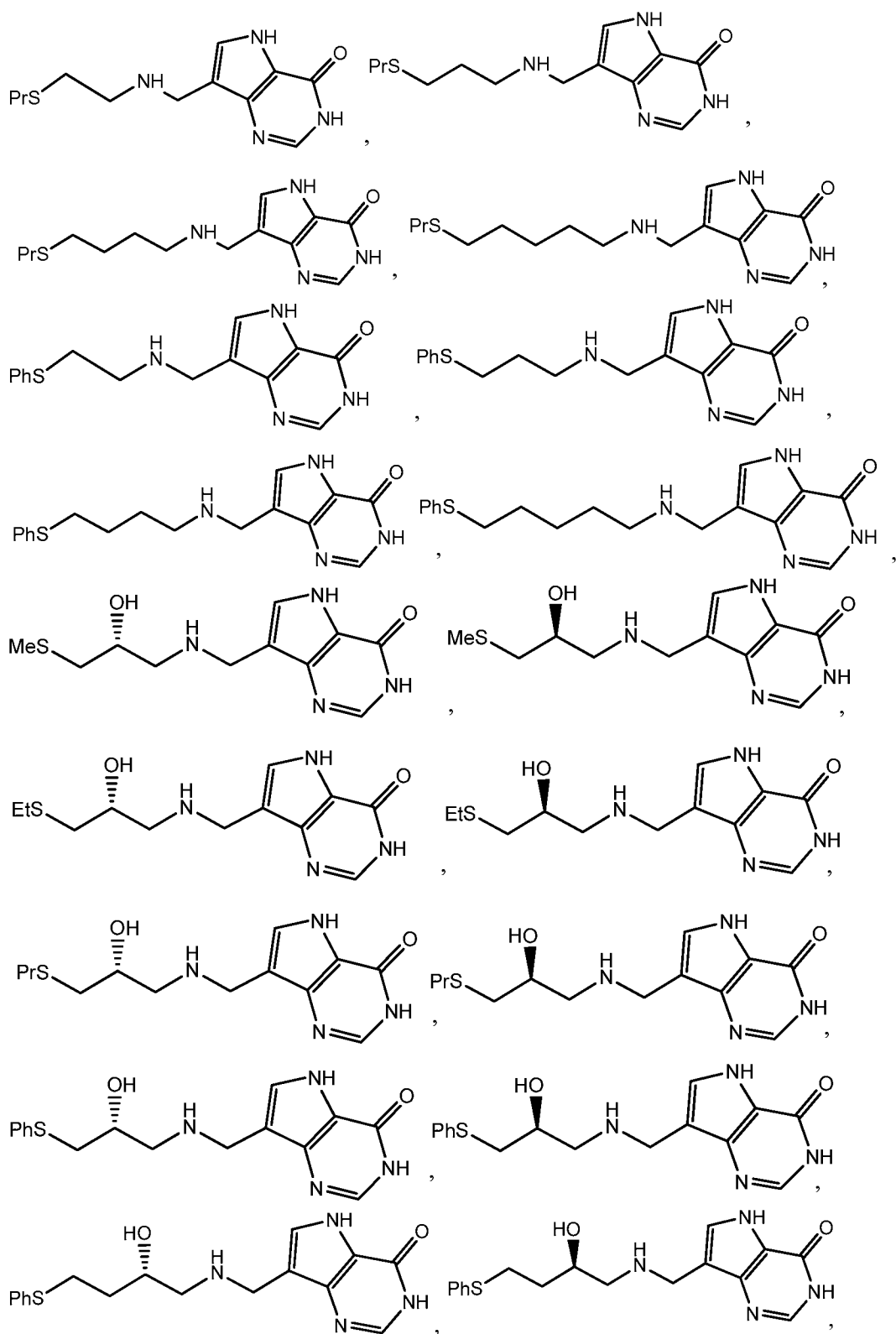
7-(((2S,3R)-3,4-dihydroxy-2-(methylthiomethyl)butylamino)methyl)-3H-pyrrolo[3,2-d]pyrimidin-4(5H)-one;

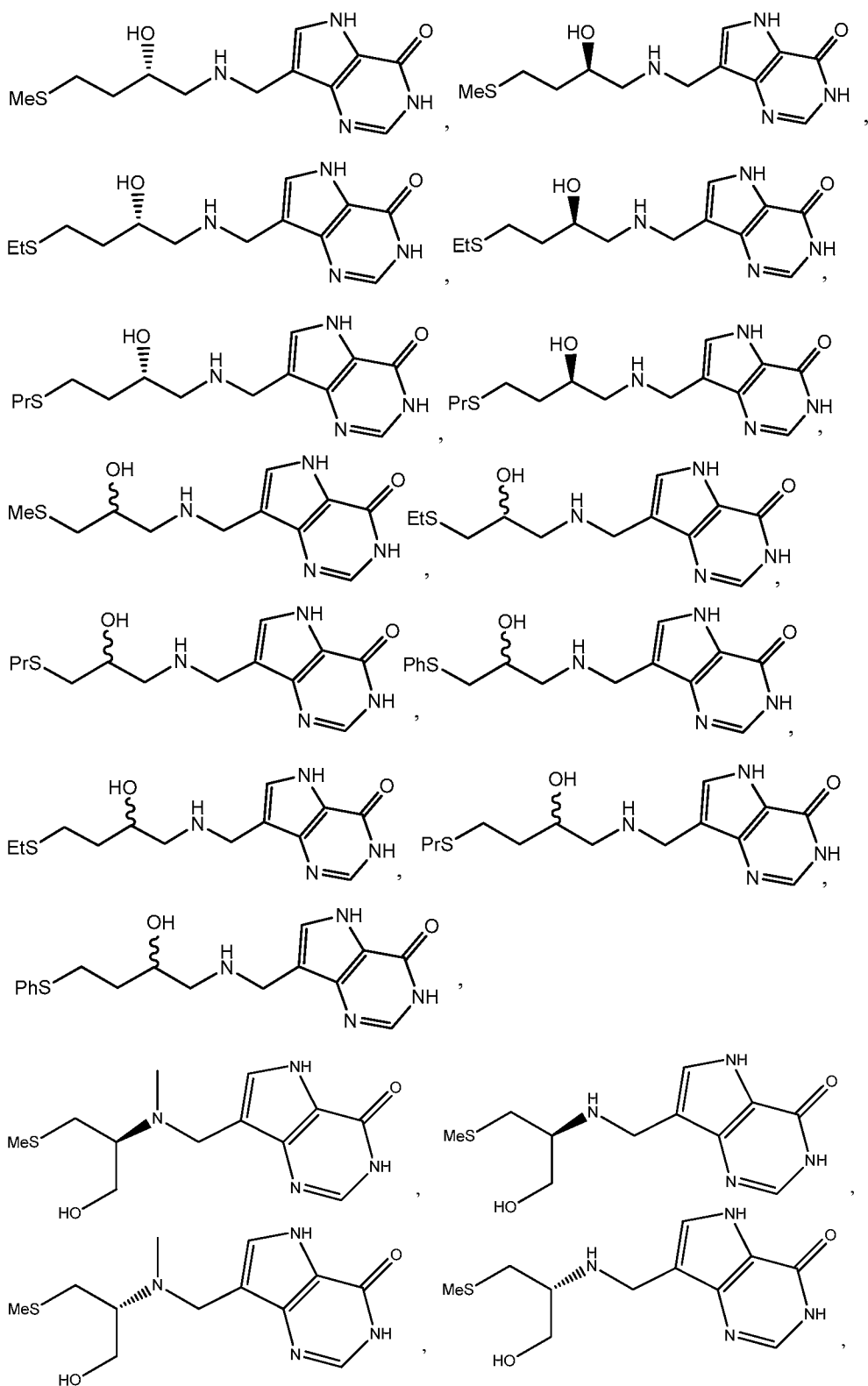


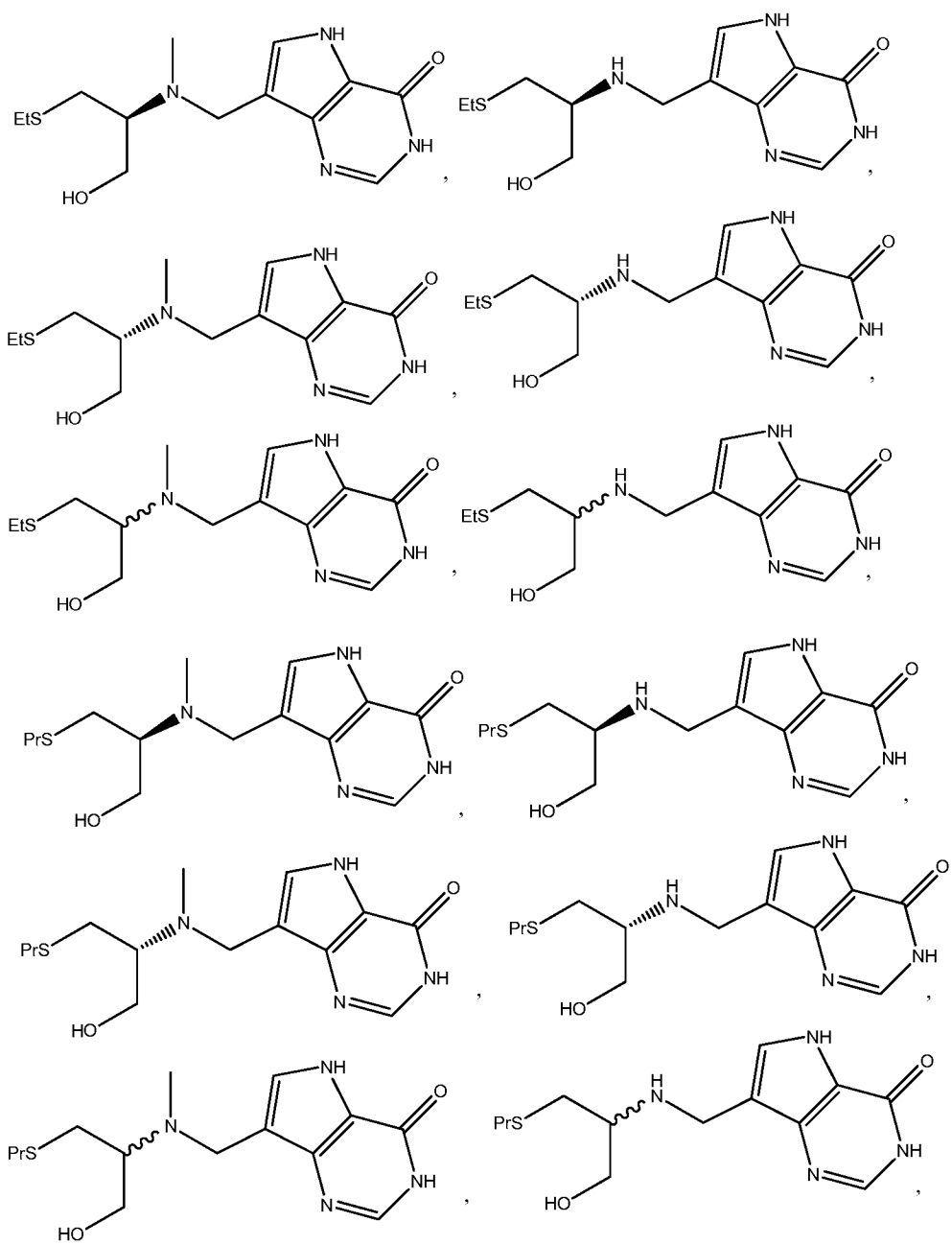
or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

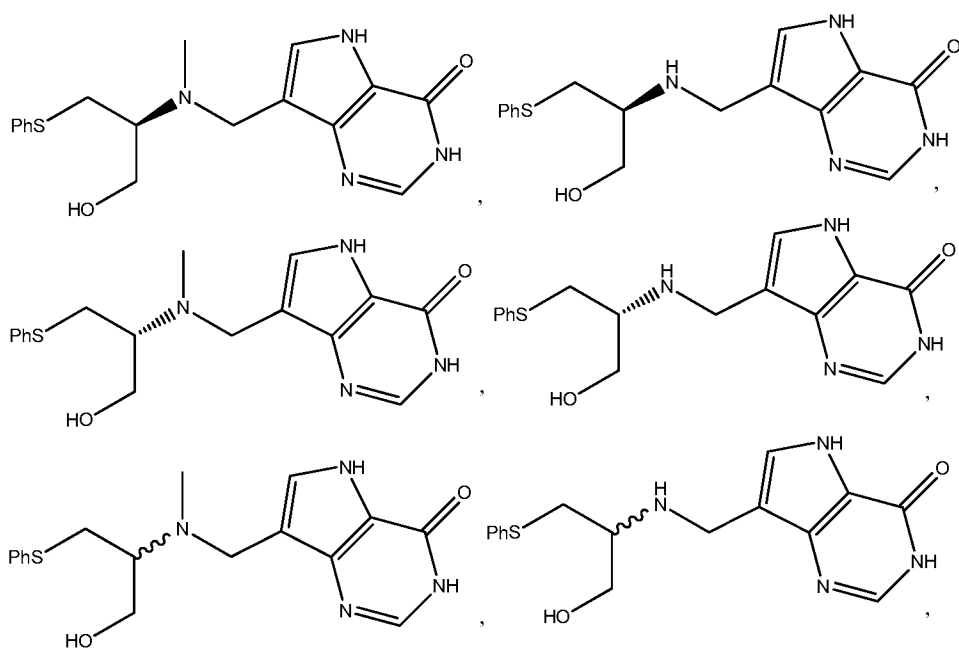
48. The method of Claim 40, wherein the MTIP inhibitor comprises a compound selected from the group consisting of:

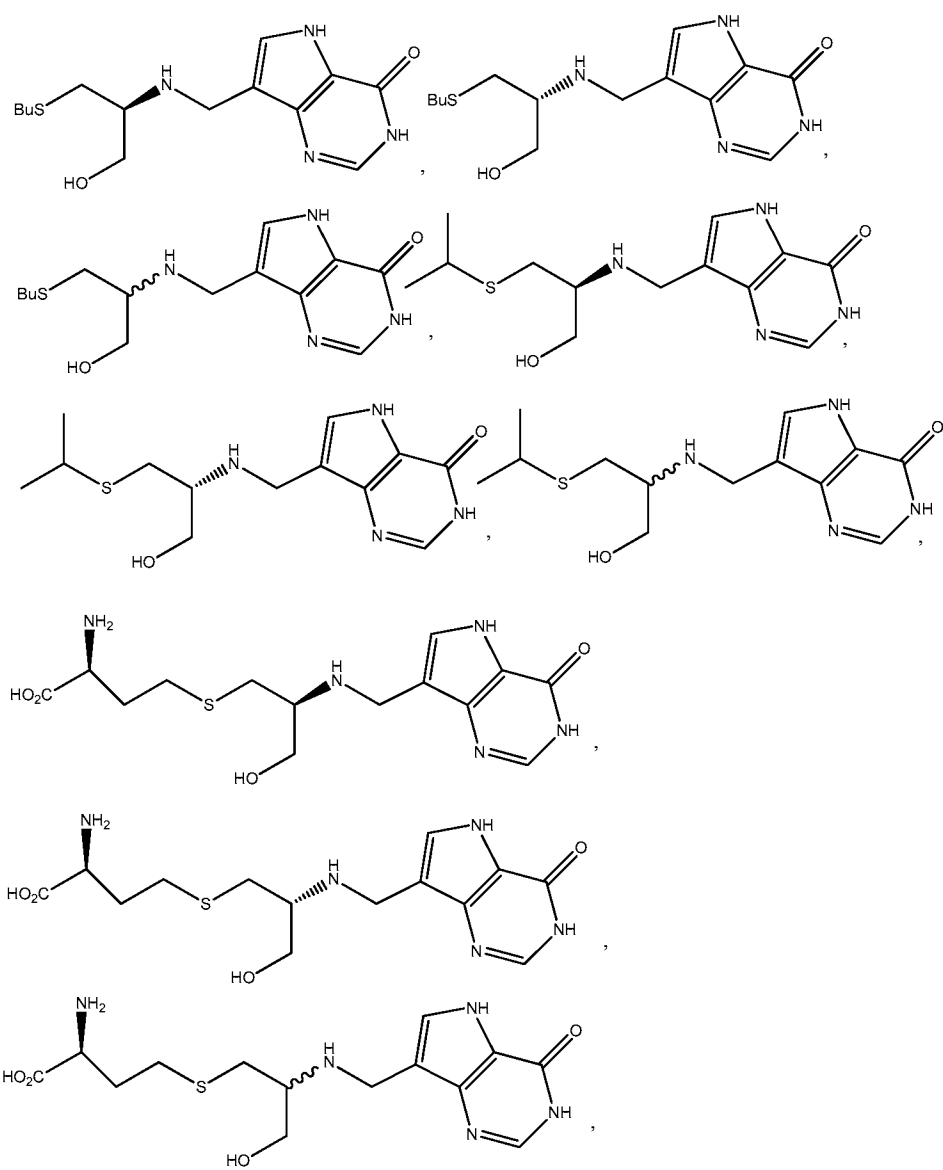


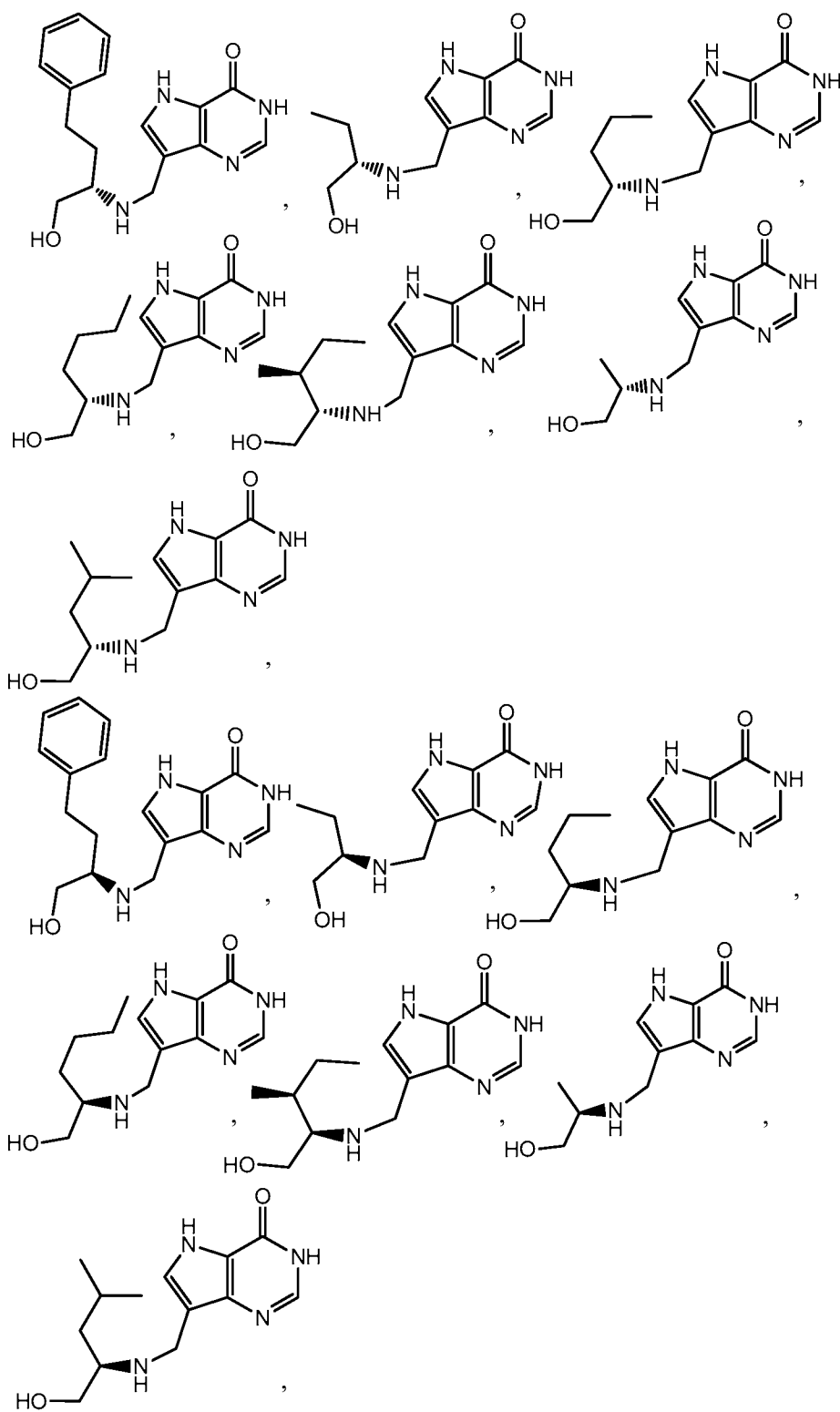






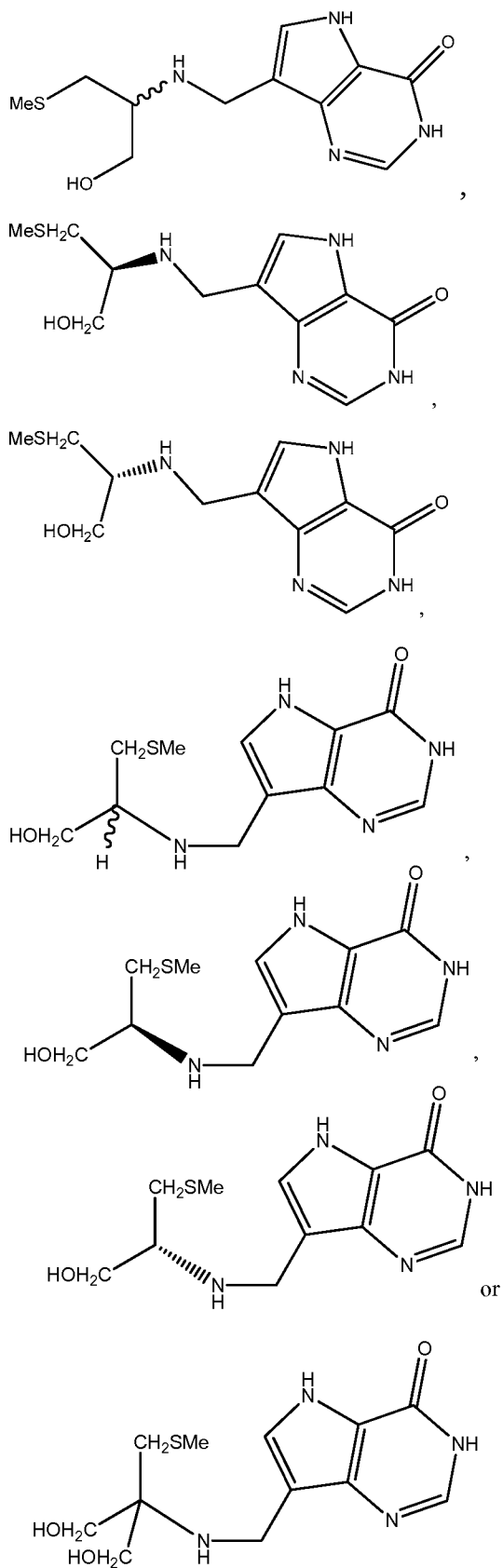




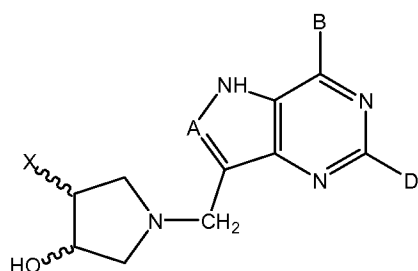


or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

49. The method of Claim 40, wherein the compound has the structure



50. The method of any of Claims 1-7, wherein the MTIP inhibitor comprises a compound having formula (VII):



(VII)

wherein

X is an alkyl, cycloalkyl, aralkyl, aralkenyl, alkenyl, alkynyl or aryl group, each of which is optionally substituted with one or more substituents selected from the group consisting of hydroxy, alkoxy, cycloalkyl, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, cyano, thiazole and NR^2R^3 group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups; or X is SR^1 ; or X is NR^2R^3 ;

R^1 , R^2 and R^3 are independently selected from the group consisting of alkyl, alkenyl, alkynyl, aralkyl or aryl, each of which is optionally substituted with one or more substituents selected from the group consisting of hydroxy, alkoxy, thiol, alkylthio, arylthio, aralkylthio, halogen, carboxylic acid, carboxylate alkyl ester, nitro, cyano and NR^2R^3 group, where each alkylthio, arylthio and aralkylthio group is optionally substituted with one or more alkyl, halogen, amino, hydroxy, or alkoxy groups;

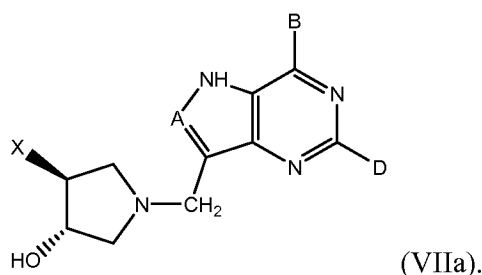
A is N or CH;

B is OH;

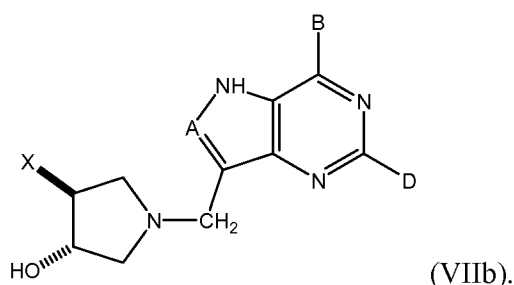
D is H, OH, NH_2 , or SCH_3 ;

provided that X is not CH_2Z , where Z is selected from OH, hydrogen, halogen, SQ^1 , OQ^2 and Q^3 , where Q^1 is an optionally substituted alkyl, aralkyl or aryl group, Q^2 is an optionally substituted alkyl group and Q^3 is an optionally substituted alkyl group; or a tautomer thereof, or a pharmaceutically acceptable salt thereof, or an ester prodrug form thereof.

51. The method of Claim 50, wherein the MTIP inhibitor comprises a compound having formula (VIIa):



52. The method of Claim 50, wherein the MTIP inhibitor comprises a compound having formula (VIIb):



53. The method of any of Claims 50-52, wherein Q¹ is an optionally substituted alkyl, aralkyl or aryl group, Q² is an optionally substituted alkyl group and Q³ is an optionally substituted alkyl group.

54. The method of any of Claims 50-53, wherein A is CH.

55. The method of any of Claims 50-54, wherein D is H.

56. The method of Claim 50, wherein the compound is selected from the group consisting of:

- (±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-phenylpyrrolidine;
- (±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-vinylpyrrolidine;
- (±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-4-ethynyl-3-hydroxypyrrrolidine;
- (±)-*trans*-4-Allyl-1-[(9-deazahypoxanthin-yl)methyl]-3-hydroxypyrrrolidine;
- (±)-*trans*-4-Cyclopropyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrrolidine;
- (±)-*trans*-4-Cyclohexyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrrolidine;
- (±)-*trans*-4-Cyclohexylmethyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrrolidine;

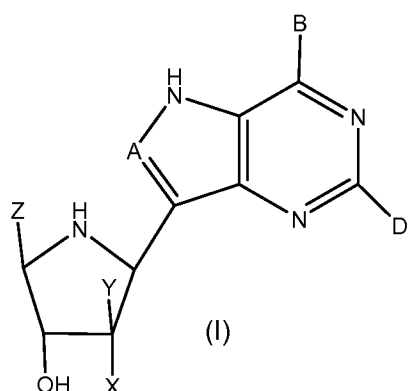
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(prop-1-en-2-yl)-pyrrolidine;
(±)-*trans*-4-Butyl-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(pent-3-yl)-pyrrolidine;
(±)-*trans*-4-Cyclopentyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(1*H*-1,2,3-triazol-1-yl)pyrrolidine;
(±)-*trans*-4-(1-Benzyl-1*H*-1,2,3-triazol-4-yl)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-4-(3-Benzylthiopropyl)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*cis*-1-[(9-Deazahypoxanthin-9-yl)methyl]-4-ethyl-3-hydroxypyrrolidine;
(3*R*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-phenylpyrrolidine;
(3*R*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-vinylpyrrolidine;
(3*R*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-4-ethynyl-3-hydroxy-pyrrolidine;
(3*R*,4*S*)-4-Allyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(3*R*,4*S*)-4-Cyclopropyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(3*R*,4*S*)-4-Cyclohexyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(3*R*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(prop-1-en-2-yl)-pyrrolidine;
(3*R*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(pent-3-yl)-pyrrolidine;
(3*R*,4*S*)-4-Cyclopentyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(3*S*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(1*H*-1,2,3-triazol-1-yl)pyrrolidine;
(3*R*,4*R*)-4-(1-Benzyl-1*H*-1,2,3-triazol-4-yl)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(2-methylpropyl)pyrrolidine;
(±)-*trans*-4-Butyl-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxypyrrolidine;
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(thiazol-2-yl)-pyrrolidine;
(3*R*,4*S*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(thiazol-2-yl)-pyrrolidine;
(±)-*trans*-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(tetrazol-5-yl)-pyrrolidine;
(3*R*,4*R*)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(tetrazol-5-yl)-pyrrolidine; and
(3*S*,4*R*)-1-[(9-Deaza-adenin-9-yl)methyl]-4-ethyl-3-hydroxypyrrolidine;
or a tautomer thereof, or a pharmaceutically acceptable salt thereof, or an ester thereof.

57. The method of any of Claims 1-56, wherein the sub-growth inhibiting amount of the MTIP inhibitor inhibits quorum sensing in the bacteria.
58. A pharmaceutical composition or an agrochemical composition comprising a sub-bacterial-growth inhibiting amount of a 5'-methylthioinosine phosphorylase (MTIP) inhibitor and a pharmaceutically acceptable carrier or an agrochemically acceptable carrier.
59. The pharmaceutical composition of Claim 58, wherein the sub-bacterial-growth inhibiting amount of the MTAN inhibitor inhibits quorum sensing in bacteria.
60. The pharmaceutical composition of Claim 58 or 59, wherein the bacteria is *Pseudomonas aeruginosa*, *Pseudomonas syringae* or *Xanthomonas campestris*.
61. The pharmaceutical composition of any one of Claims 58-60, wherein the pharmaceutical composition is formulated in dosage form.
62. A method for determining whether or not a compound is a candidate for treating an infection caused by bacteria that use 5'-methylthioinosine phosphorylase (MTIP) in a quorum sensing pathway, the method comprising determining whether or not the compound inhibits MTIP, wherein a compound that inhibits MTIP is a candidate for treating an infection caused by bacteria that use MTIP in a quorum sensing pathway and wherein a compound that does not inhibit MTIP is not a candidate for treating an infection caused by bacteria that use MTIP in a quorum sensing pathway.
63. The method of Claim 62, wherein the bacteria is *Pseudomonas aeruginosa*, *Pseudomonas syringae* or *Xanthomonas campestris*.
64. The method of any of Claims 1-57 or 62-63, or the composition of any of Claims 58-61, wherein the MTIP inhibitor has an IC₅₀ value less than 50 nanomolar for MTIP.
65. The method of any of Claims 1-57 or 62-64, or the composition of any of Claims 58-61 or 64, wherein the MTIP inhibitor has an IC₅₀ value greater than or equal to 50 nanomolar for human purine nucleoside phosphorylase (PNP).

66. The method of any of Claims 1-57 or 62-65, or the composition of any of Claims 58-61 or 64-65, wherein the MTIP inhibitor has an IC₅₀ value greater than or equal to 50 nanomolar for 5'-methylthioadenosine nucleosidase (MTAN).

67. The method of any of Claims 1-57 or 62-66, or the composition of any of Claims 58-61 or 64-66, wherein the MTIP inhibitor has an IC₅₀ value greater than or equal to 50 nanomolar for 5'-methylthioadenosine phosphorylase (MTAP).

68. A compound having the formula:



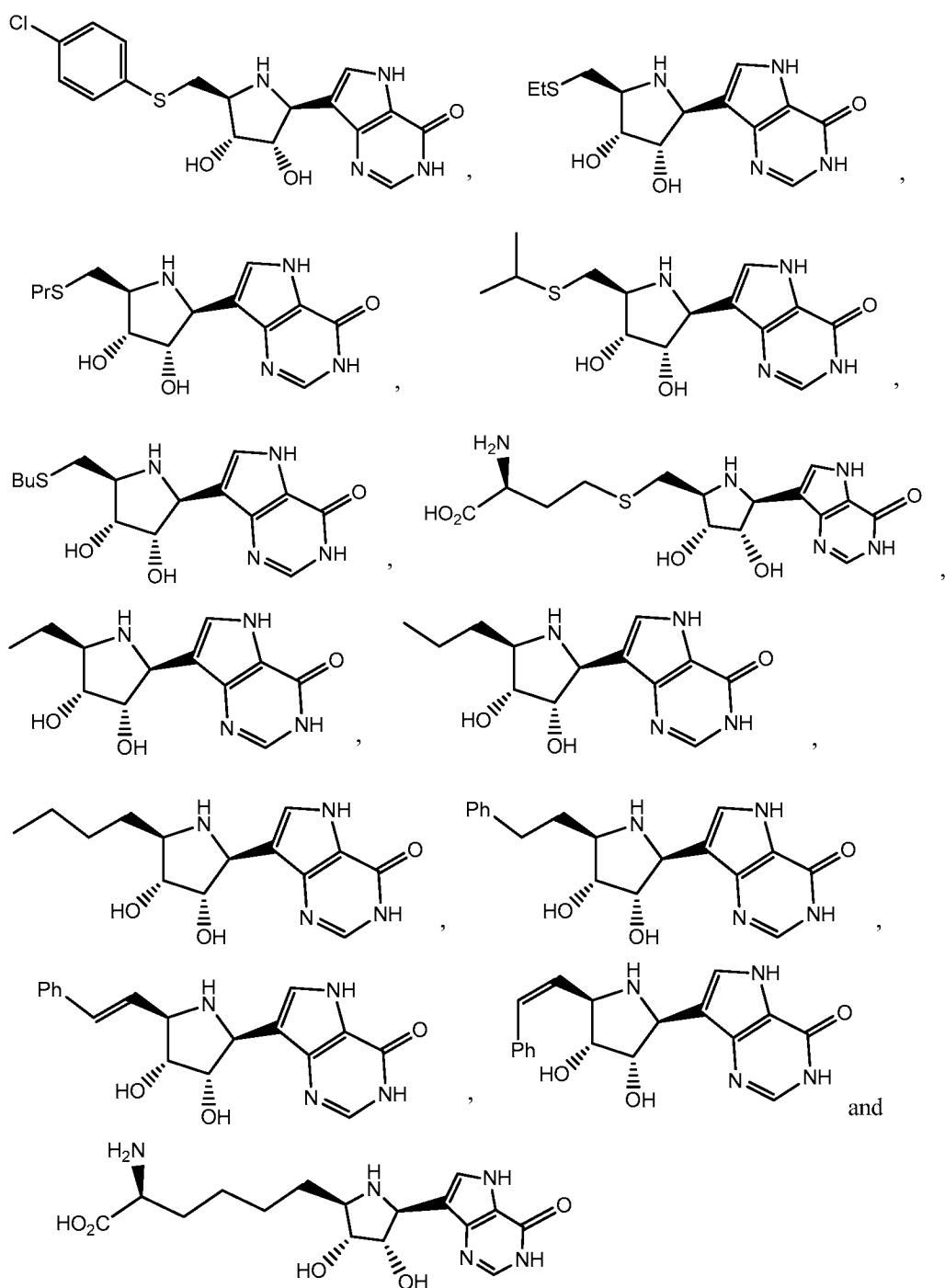
wherein A is CH or N; B is OH; D is chosen from H, OH, NH₂, or SCH₃; X is OH; Y is H; and Z is Q, where Q is a methyl group which is substituted with one or more substituents selected from the group consisting of methoxy, amino and carboxy, or Q is an optionally substituted, alkenyl, aralkyl, aralkenyl, aryl group or C₂-C₁₀ alkyl group, each of which is optionally substituted with one or more substituents selected from hydroxy, halogen, methoxy, amino, carboxy, or straight- or branched-chain C₁-C₆ alkyl; or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

69. The compound of Claim 68, wherein A is CH.

70. The compound of Claim 69 or 70, wherein D is H.

71. A compound selected from the group consisting of :

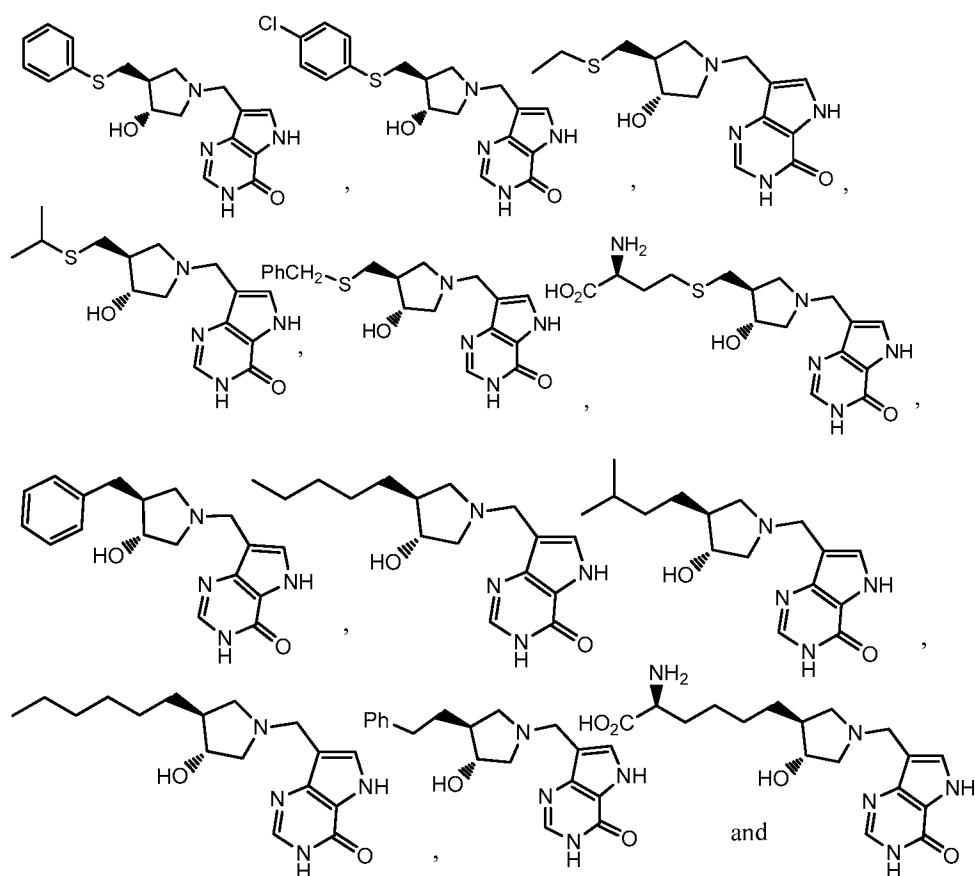
-132-



or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

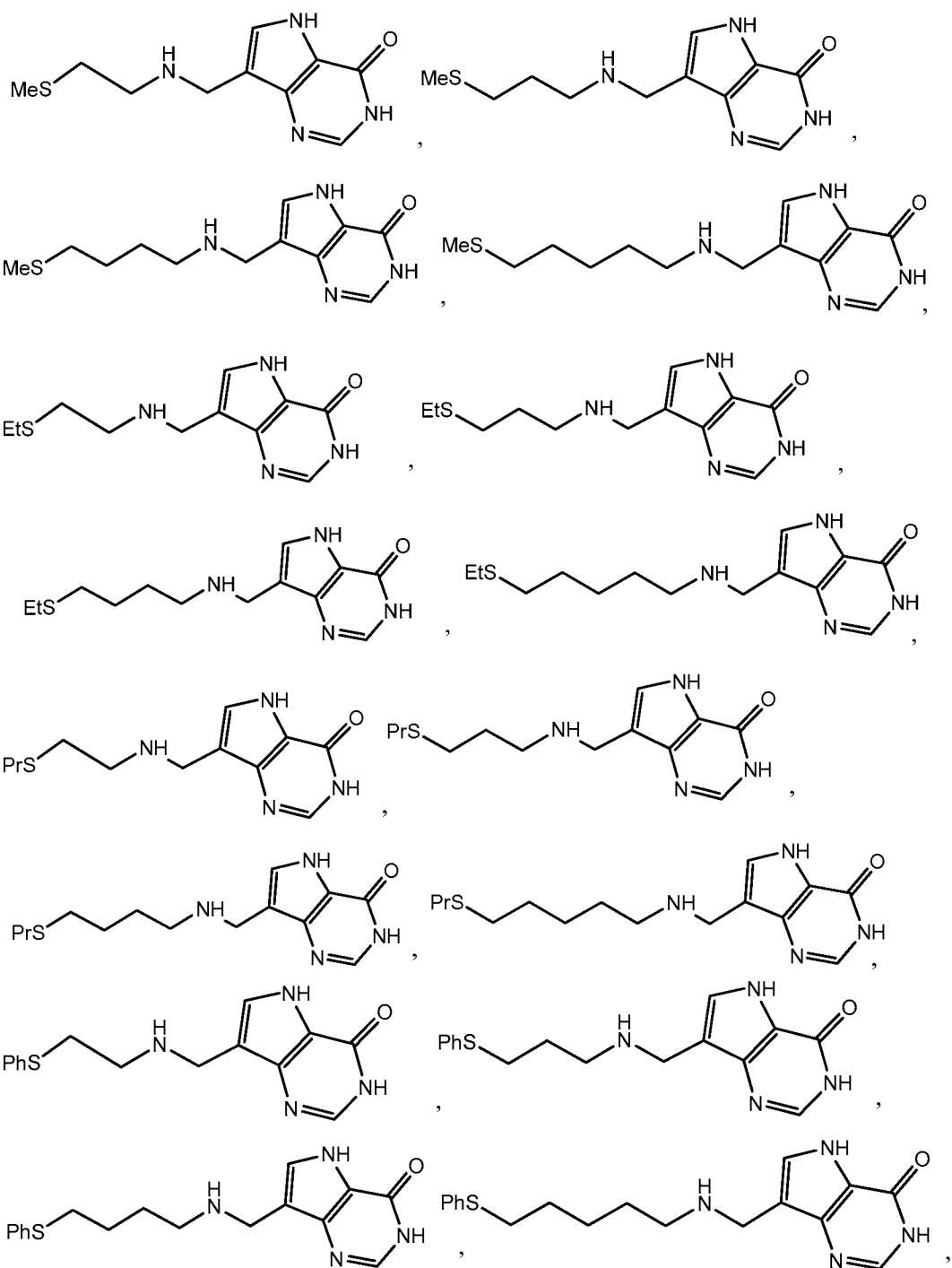
72. A compound selected from the group consisting of:

-133-

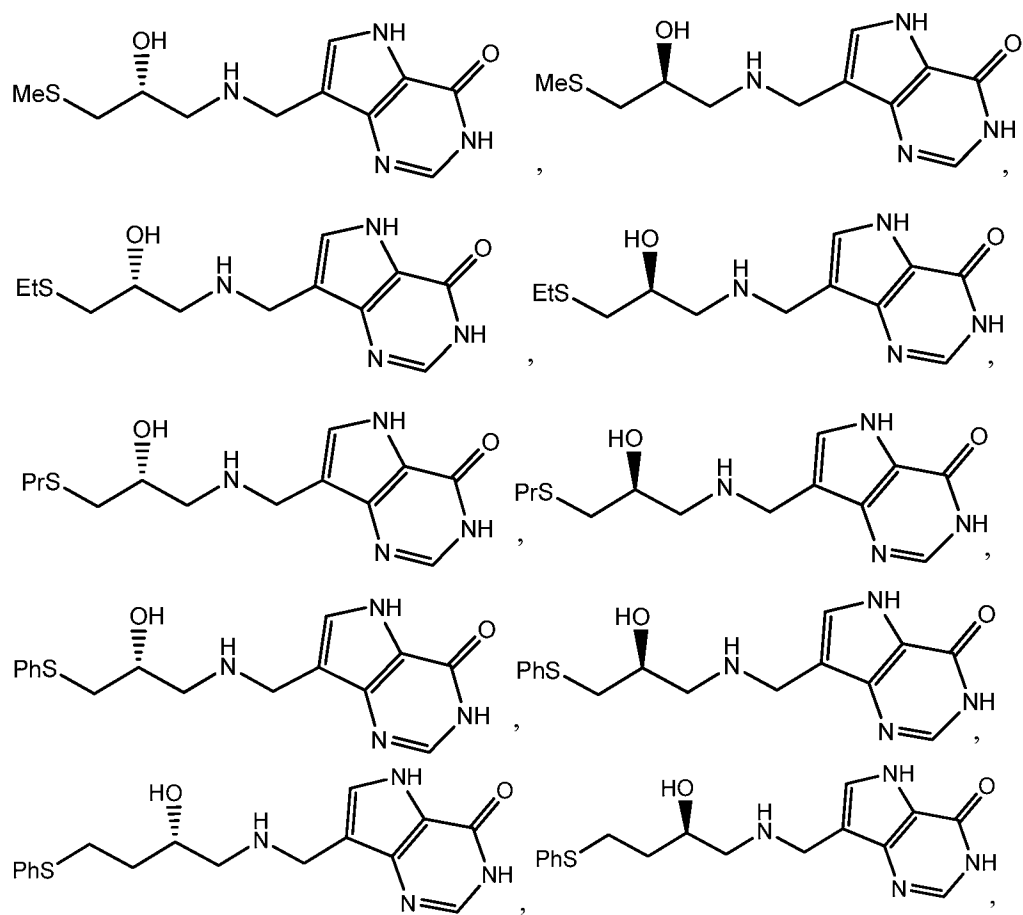


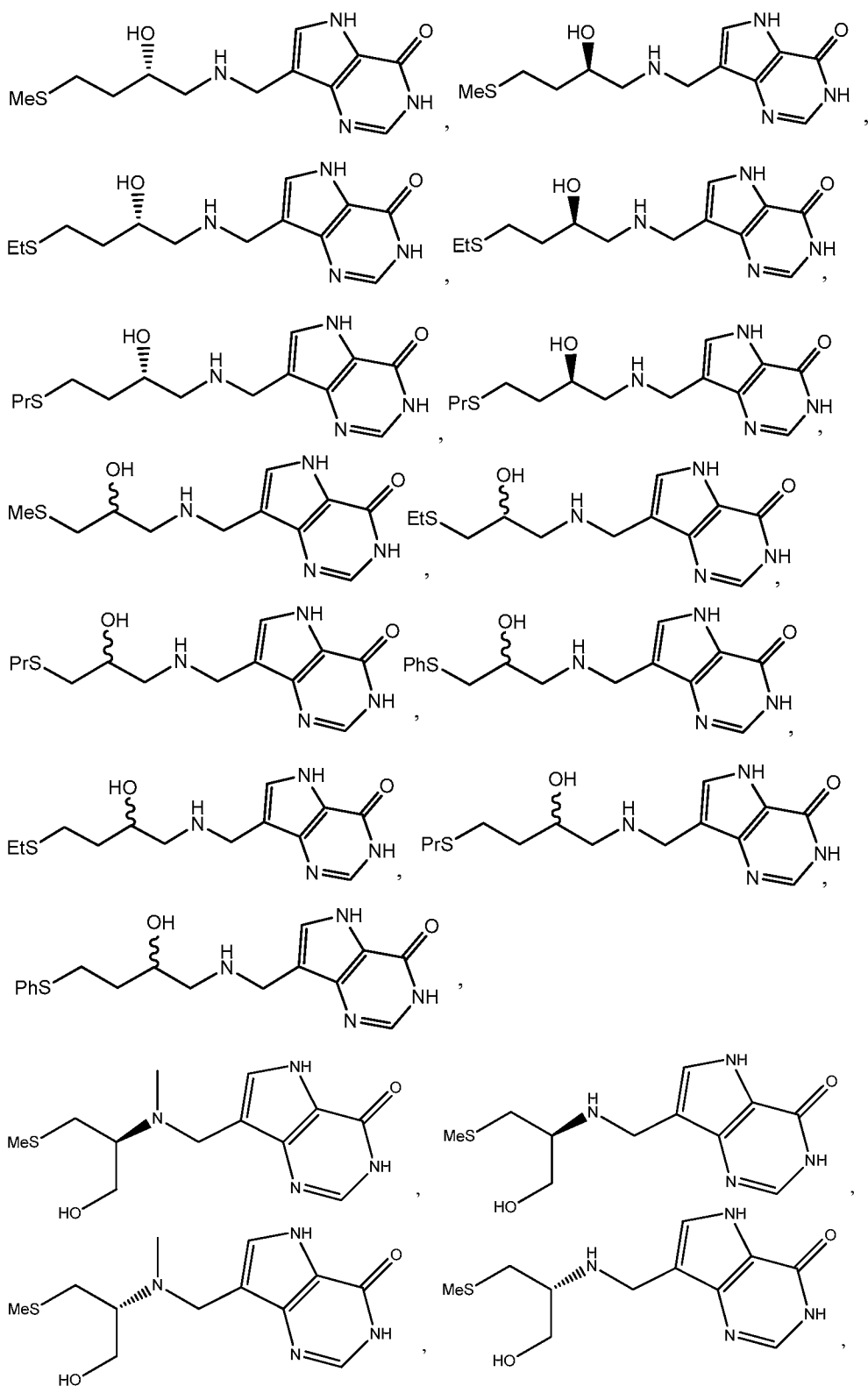
or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

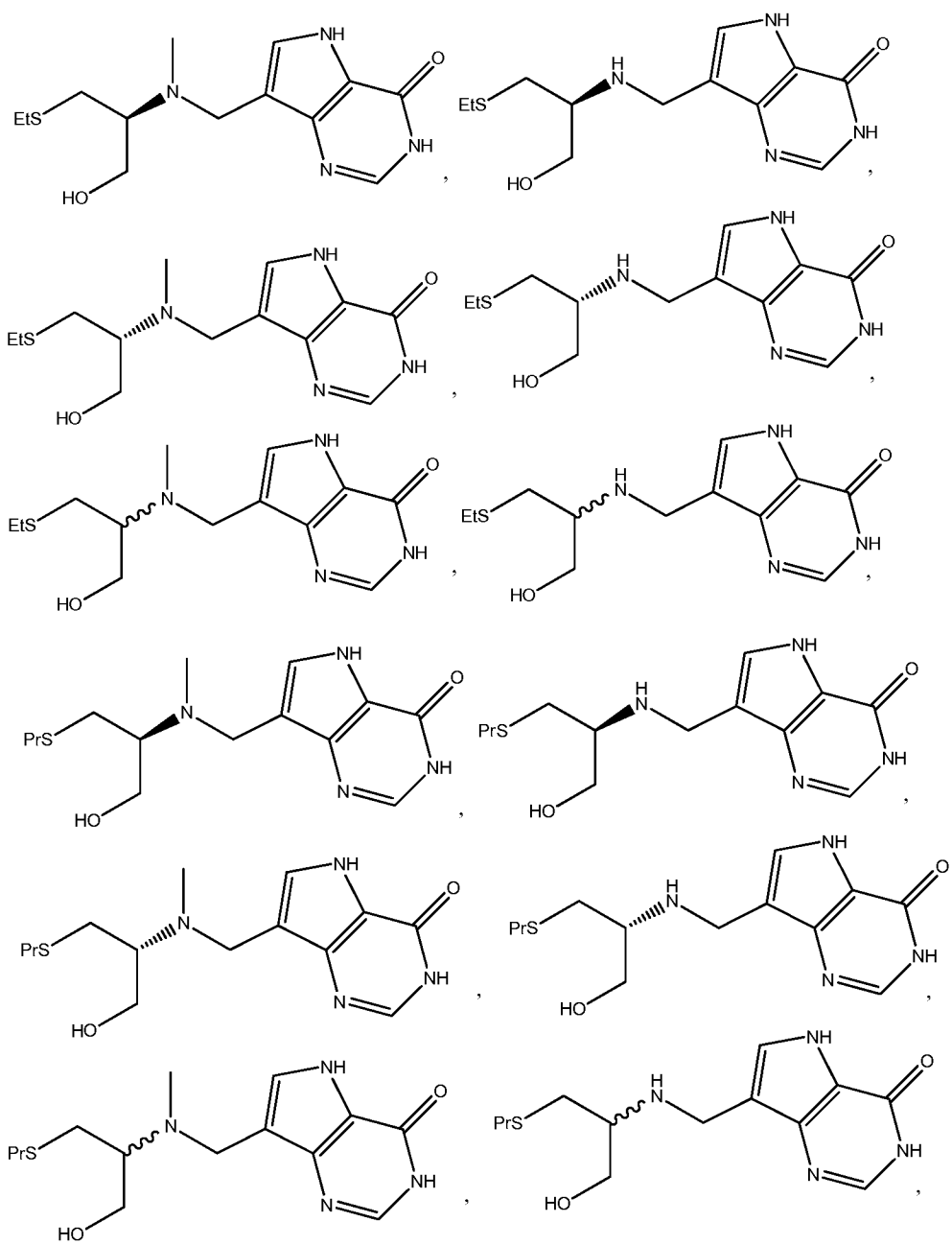
73. A compound selected from the group consisting of:

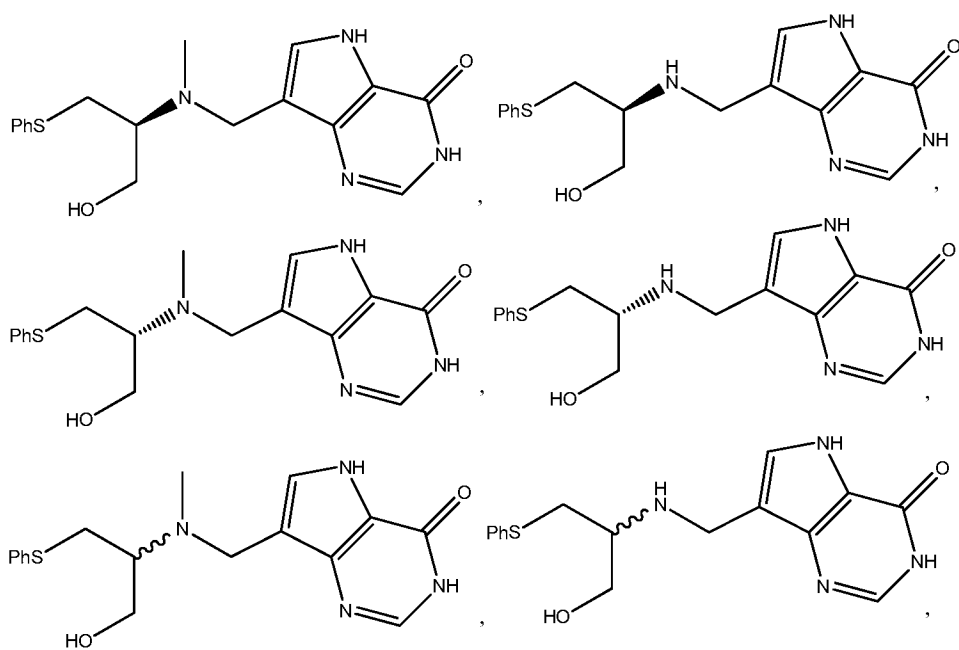


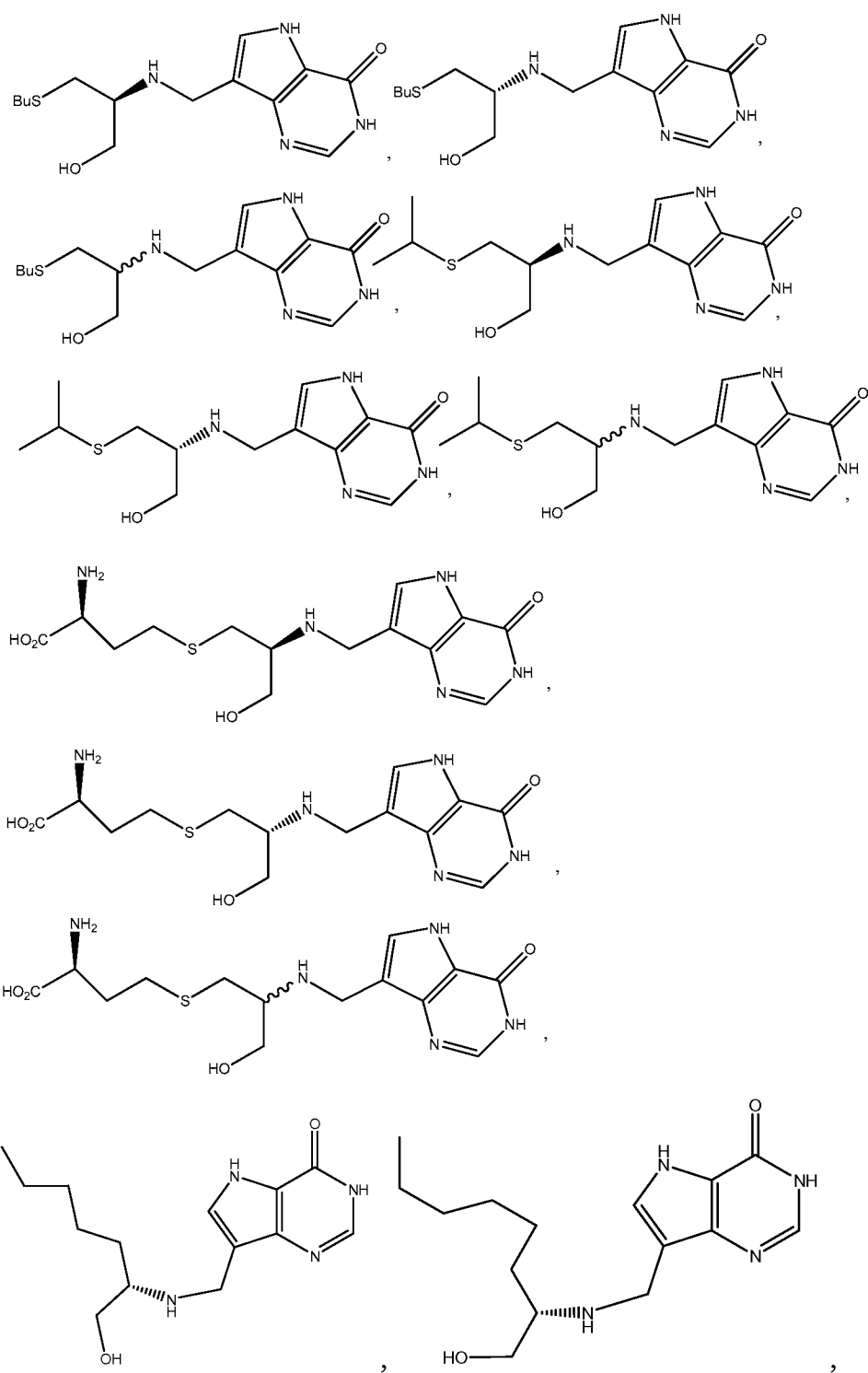
-135-

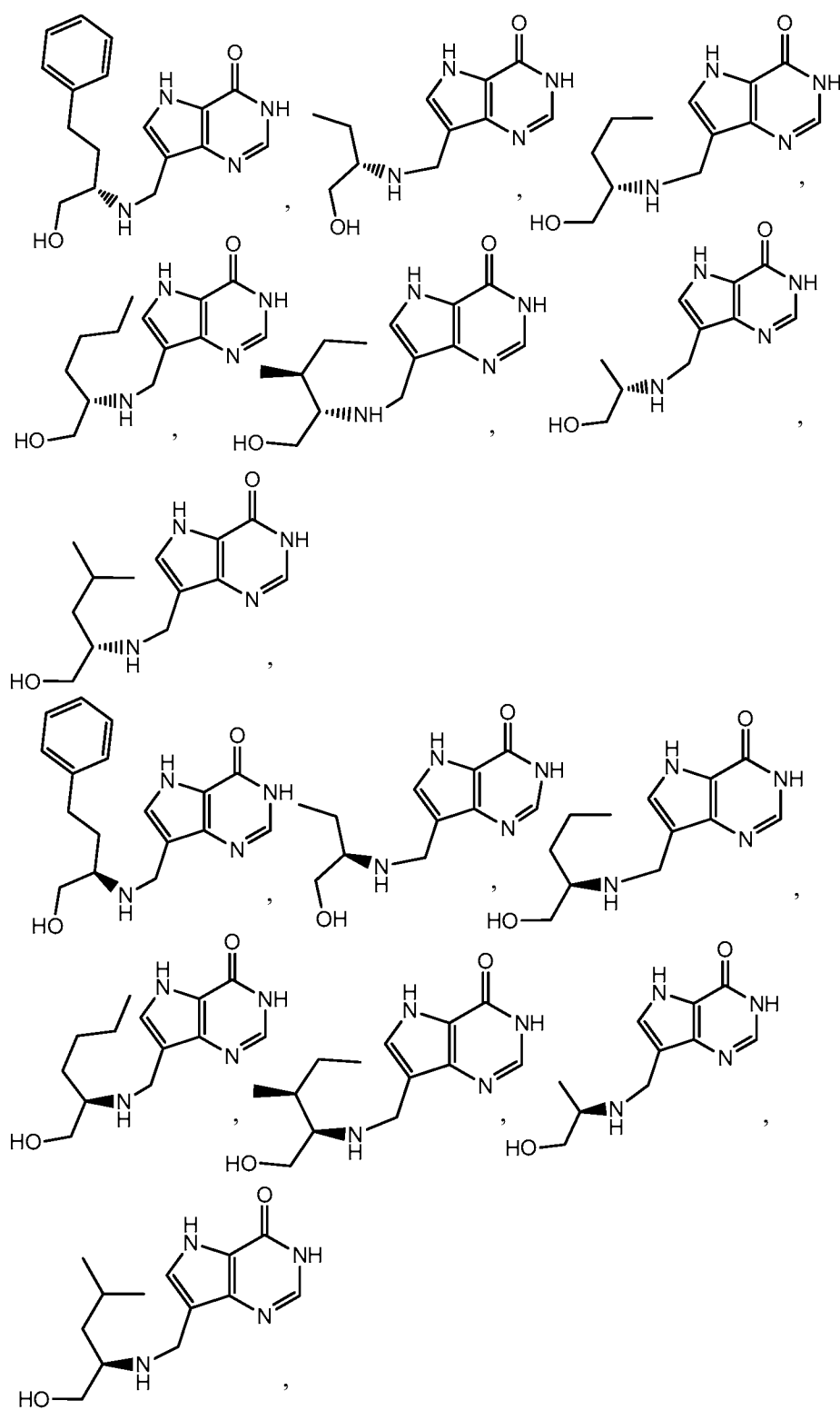


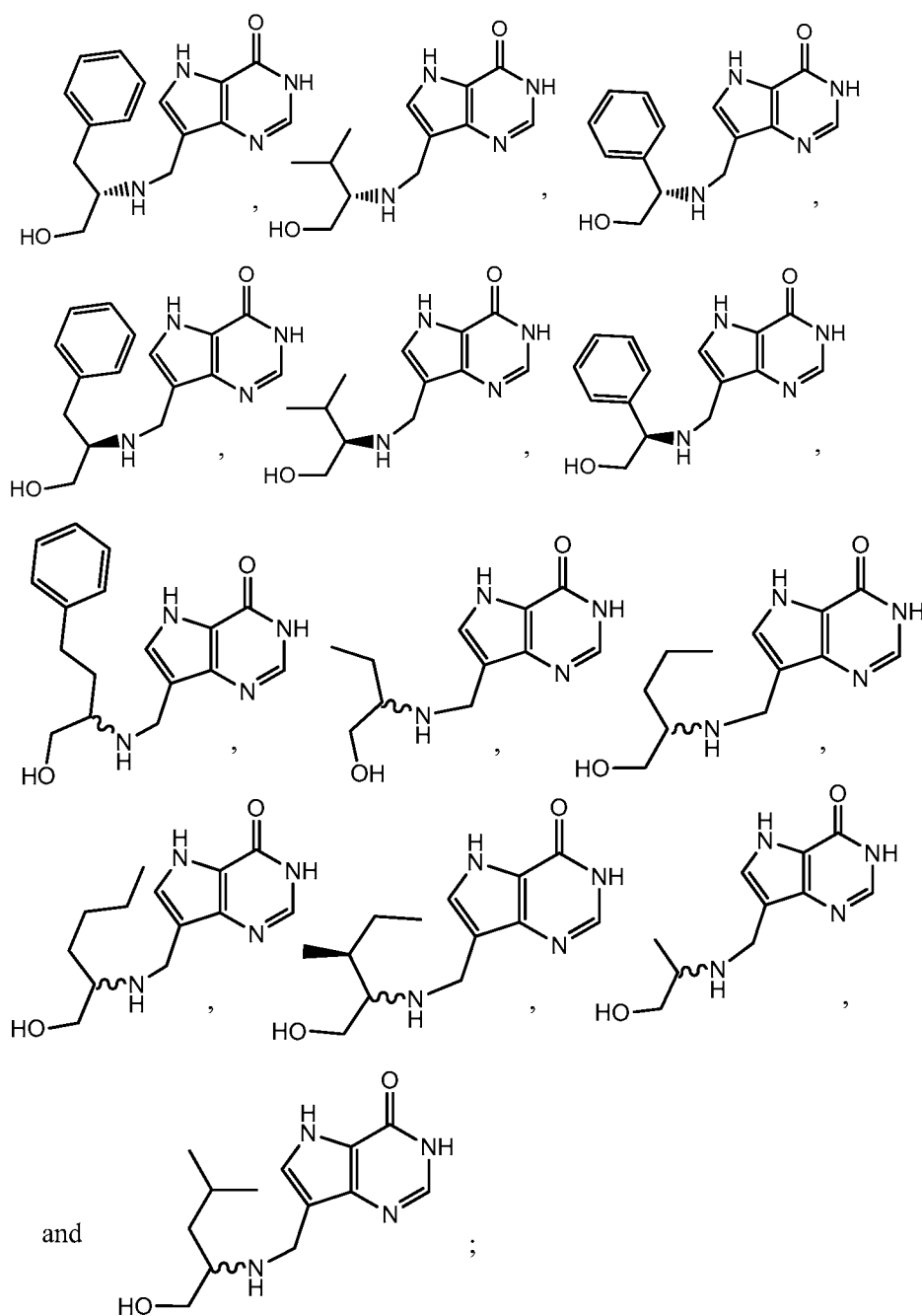












or a tautomer thereof; or a pharmaceutically acceptable salt thereof; or an ester thereof.

74. A pharmaceutical composition or an agrochemical composition comprising the compound of any of Claims 68-73 and a pharmaceutically or an agrochemically acceptable carrier.

75. The composition of any of Claims 58-61 or 74, comprising 5'-methylthioinosine and/or 5'-methylthioadenosine.

76. The method of any of Claims 1-57, comprising coadministering the 5'-methylthioinosine phosphorylase (MTIP) inhibitor with 5'-methylthioinosine and/or 5'-methylthioadenosine.

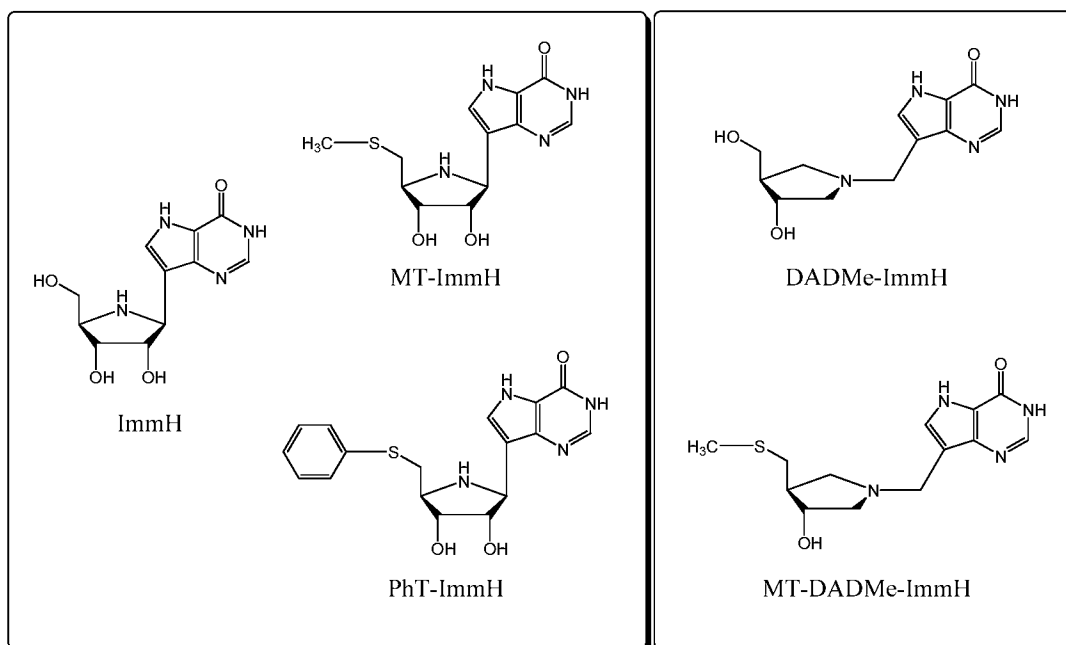


FIGURE 1

2/3

1YR3 6 -FSHNPLFCIDI I KTKPD-FTPRVAFILGSGILGALAD-QIENAVASIEKLPGFVYSTVGHAGELVLGHLQGVVVCMKGR-----GHFYEGRGM
 1N3I 9 --DELARAAQVIADRTG-IGEDHVAVLGSGWLPAVAALGSPITVLPQAEPLGFPVPTAAGHAGELLSPVIGAHRLVLVLAGR-----I HAYEGHDL
 3KHS 3 -DYDLAKETAAWLNKQLQ--IRPVLGIVCGSGIKIGD-SLETSTIVAYSIDIPNFPVSGVKGHAGSLIFGSGVNGVSCVCMKGR-----F HLYEGHTA
 2P4S 90 YTYDTLQEIATYLLERTE--LRPKVGIIICGSLGLTAE-QLTDDVDFDIETIPHPVYSTVAGHVGRVLFGYLAGVPVCMQGR-----F HLYEGYPL
 1VMK 1 -MMKIEEARTFISERTN--LSPDILIIILSGFGPFIE-KVEDPVIIDYKDIPHPQPTVEGSHGKLVFGRLSDKPMIMAGR-----F HLYEGHDP
 1A9T 5 YTYEDYQDTAKWLLSHT--QRQVAVICGSGIGGLVN-KLTQATQFDYSEIPNFPSTVPGHAGRLVFGFLNGRACVMMQGR-----F HMYEGYPF
 1RR6 5 YTYEDYKNTAEWLLSHTK--HRQVAVICGSGIGGLTD-KLTQAIQFDYSEIPNFPSTVPGHAGRLVFGFLNGRACVMMQGR-----F HMYEGYPL
 3OZB 17 -----VYAIIG--GTGLTQLEGLTSESL--PIETPSTGAPS--APLQGRGYAGREVLFARHGHGPHRFPHPQV-----
 1V4N 7 -----EKASIGIIG--GSGLYDPQIILNVKEI--KVYTPYGEPS--DNIILGELEGRKVAFLPRHGRGHRIPPHKI-----
 1K27 11 -----KIGIIGGTGLDD-PE--ILEGRTEK--YVDTPTFGKPS--DALILGKIKNVDCVLLARHGRGQHTIMPSKV-----
 1WTA 3 -----EITR--PPG--VRAHVGVIG--GSGLYDPGIVENPEV--KVSTPYGNPS--DFIVVGDVAGVKVAFLPRHGRGHRIPPHAI-----
 2A8Y 2 -----IEQN-----EKASIGIIGGSGLYDP-G--IFSESKEI--KVYTPYQPS--DFITIGKIGNKSVAFLEPRHGRGHRIPPHKI-----

 1YR3 95 TIMTDAIRTFKLLGCELLFCTNAAGSLRPEVGGAGSLVALKDHINTM--PGTPWVGL-NDDRFGERFTSLANAYDAEYRALLQKVAKEEGFP--LTEG
 1N3I 98 RYVHPVRAARAAGAQIMVLTNAAGGLRADLQVGQPVLSIDHNLTA--RSPLVGG--EFVLDTDAYSPLRLRELARQSDP-----QLAEG
 3KHS 91 ARATFPMRVFKALGVKIVLVTNAAGGLNRYRPGDFMVVRDHIINLPLAGANPLTGP-NDDTEGERFPSMTSVYDKTLRKYAI SAARELGMSYATHEG
 2P4S 179 AKCAMPVRVMHLIGCTHLIATNAAGGANPKYRVDIMLIKDHINLMGFAGNNPLOGP-NDERFGPRFGMANTYDPKLNQOAKVIARQIGIENELREG
 1VMK 89 ATVAFVYLAKYVGVKVVVNAAGAINPEFKPEIILVRDIINFMF--RNPLRGP-NDEKIGPRFPMSSVVDPEWARKIQERLS-----LKEG
 1A9T 94 WKVTFPVVRVRLGLVETLVVTNAAGGLNPNFEVGDIMLIRDHINLPLGFGSGENPLRGP-NEERFGVRFPAMSDAYDRDMRQKAHSTWKQMGEORELOEG
 1RR6 94 WKVTFPVVRVRLGLVETLVVTNAAGGLNPNFEVGDIMLIRDHINLPLGFGSGENPLRGP-NDERFGDRFPAMSDAYDRMTQRALSTWKQMGEORELOEG
 3OZB 79 -NYRANIWALKQAGAEAVI AVNAVGGIHAAMGTGHLCPHQI IDYTSG-REHTYFAG--DIEHVTHIDFESHYPYDEPLRQRLIEALRALGL-AHSSHG
 1V4N 72 -NYRANIWALKSLGVKRWI AVSAGSLRLDYKPGDFVVPNQFIDMTKG-RTYTFDG----PTVAHVSMADPFCEHLRSIILDSAKDLGI-TTHDKG
 1K27 73 -NYQANIWALKEEGCTHVITTACGSLREEIQPGDIVIIDQFIDRTM-RPQSFYDGSCHSARGVChIPMAEPFCPTREVL IETAKKLGL-RCHSKG
 1WTA 75 -NYRANIWALKALGVKRWI SVSAGSLREDYRPGDFVVPDQFIDMTKNRRHYTFD----GPVTvhVSMADPFCEDLRQRLIDSGRRGLG-TVHERG
 2A8Y 71 -NYRANIWALKELGVWRVVISV SAVGSLRMDYKLGDFVIPDQFIDMTKN-REYSFFD----GPVVAHVSMADPFCEDLRQRLIDSGRRGLG-TVHERG

 1YR3 190 VFVSPGPNFETAEEIRMMQI-IGGDVVGM SVVPEVISARHCDLKVAVASAITNMAEGL--SDVKLSHAQTLAAAEELSKQNFNLICGFLRKIA---
 1N3I 182 VYAGLPGPHYETPAEIRMLQT-LGADLVGMSTVHETIAARAAGAEVLGVS LVNTNLAAGI--TGEPLSHA EVLAAAGAA SATRMGALLADVIARF---
 3KHS 191 VYCCVNGPSFETPAECKJLRL-MGSDAVGMSTAPETIIVAKHGMRCCLAVSLISNVITASNCETPAEPTHEEVL RAGEEASARMTALVKLVIEKIRGE-
 2P4S 279 VYTCLGGPNFETVAEVKMLSM-LGVDAIGMSTVHEIITARHCGMTCFAFSLITNMCTMSYEEEECHDSIVGVGKNREKTLGFEVSRIVKHIHYEA
 1VMK 179 VYIGVLGPNFETPAEIRVFEK-LGADLVGMSTVPEVIAAKHCGKLVVFCVNTNMAAG--ITHGRLSHEEVVTRTKMAQQKIEKALT TAVEVF---
 1A9T 194 TYVMLGPNFETVAECRLRN-LGADAVGMSTVPEVIVARHCGLRVFGFSLITNKVIMDTESQOKANHEEVL EAGKQAAQKLEQFVSLIMASI---
 1RR6 194 TYVMVAGPSFETVAECRVLQK-LGADAVGMSTVPEVIVARHCGLRVFGFSLITNKVIMDTESLEKANHEEVL EAGKQAAQKLEQFVSLIMASI---
 3OZB 174 VYATQGPRL ETVAEIA RLER-DGNDIVGMTCMPEAALARELDIPYACALALVNPAAG-K-SAGIITmaeIEQALHDGKREVLRVLA-----
 1V4N 165 TYICIEGPRFSTRAESIVMKEVFKADIIGMTLVPEVNLAACEAEMCYSGVMGTDYDV-F-ADIPVtaEEVTKVMAENTAKVKKLLYEVRRLP---
 1K27 171 TMVTTIEGPRFSSRAESFMFRT-WGADV INNTVPEVWLAKAGICYASIAMATDYDC-WKEHEEA VSVDRVLKTLKENANKAKSLLLTIPQIGSTE
 1WTA 169 TYVTCIEGPRFSTRAESRVKDVFKADIIGMTLVPEVNLAACEAQLCYATIAMVTDYDV-W-ADRPVtaEEVERVMI SNVERARMLYDVIPKLAGE-
 2A8Y 164 TYICIEGPRFSTRAESRTWREVYKADIIGMTLVPEVNLAACEAQCYATIAMVTDYDV-F-AEIPVtaEEVTRVMAENTEKAKKLLYALIQLKPEK*

FIGURE 2

3/3

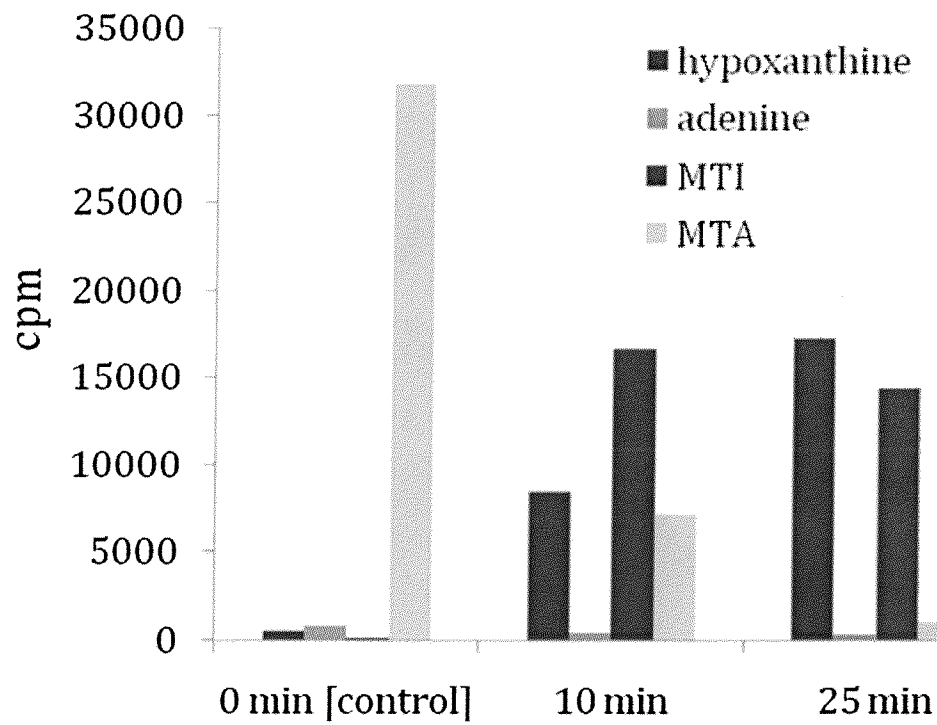


FIGURE 3

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 11/62208

A. CLASSIFICATION OF SUBJECT MATTER

IPC(8) - A61K 31/7076; A61K 31/41 (2012.01)

USPC - 514/46, 514/359, 536/27.8, 424/94.5

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

USPC: 514/46, 514/359, 536/27.8, 424/94.5

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

USPC: 424/94.1; 435/183, 193-194; 514/18.7, 27.21, 27.2, 27.13, 27.1, 22.1, 43, 45, 183; 536/27.6 (see search terms below)

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

PubWEST;PGPB, USPT, EPAB, JPAB, SureChem, GoogleScholar, Dialog

5'-methylthioinosine phosphorylase (MTIP), inhibitor, antagonist, quorum sensing, sub-growth inhibiting amount, Pseudomonas aeruginosa, Pseudomonas syringae, Xanthomonas campestris, 5'-methylthioadenosine etc.

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2008/030119 A1 (CLINCH et al.) 13 March 2008 (13.03.2008) pg 96, compound 7.3	73
X --- Y	US 5,985,848 A (FURNEAUX et al.) 16 November 1999 (16.11.1999) col 1, ln 45-col 2, ln 15, col 3, ln 30-40, col 3, ln 55-65, col 22, ln 10-15, col 25, compound examples 5-7	71 ----- 1-6, 58-60
X	US 7,553,839 B2 (EVANS et al.) 30 June 2009 (30.06.2009) col 3, ln 30-col 4, ln 30; col 27, compound 37	72
Y	US 6,562,785 B1 (SHAPIRO) 13 May 2003 (13.05.2003) Title; col 2, ln 40-45; col 9, ln 5-20	1-6, 58-60
Y	US 2008/0070860 A1 (SNYDER et al.) 20 March 2008 (20.03.2008) para [0003], [0005], [0008], [0010], [0068], [0074], [0144], [0172]	1-6
Y	Trelles et al. Nucleoside synthesis by immobilised bacterial whole cells. Journal of Molecular Catalysis B: Enzymatic, 2004, Vol 30, pp 219-227; pg 220, col 1, pg 222, col 2	5, 7

☒ Further documents are listed in the continuation of Box C.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search

9 March 2012 (09.03.2012)

Date of mailing of the international search report

23 APR 2012

Name and mailing address of the ISA/US

Mail Stop PCT, Attn: ISA/US, Commissioner for Patents
P.O. Box 1450, Alexandria, Virginia 22313-1450

Facsimile No. 571-273-3201

Authorized officer:

Lee W. Young

PCT Helpdesk: 571-272-4300
PCT OSP: 571-272-7774

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 11/62208

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	Dixon. Natural products and plant disease resistance. NATURE, 2001, Vol 411, pg 844, col 1	5, 7
A/P	Guan et al. Methylthioinosine Phosphorylase from Pseudomonas aeruginosa. Structure and Annotation of a Novel Enzyme in Quorum Sensing. Biochemistry, 22 February 2011, Vol 50(7), pp 1247-1254; Abstract	62-63
A	Hentzer et al. Inhibition of quorum sensing in Pseudomonas aeruginosa biofilm bacteria by a halogenated furanone compound. Microbiology, 2002, Vol 148, pp 87-102	1-7, 58-60, 62-63, 71-73

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 11/62208

Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. ☒ Claims Nos.: 8-57, 61, 64-70, 74-76
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of additional fees.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.
- ☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.
- ☐ No protest accompanied the payment of additional search fees.