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PROCESS FOR PREPARING INHIBITORS OF NUCLEOSIDE PHOSPHORYLASES AND NUCLEOSIDASES

5 TECHNICAL FIELD

This invention relates to a process for the preparation of certain nucleoside analogues. In particular, the invention relates to a process that includes the reaction of formaldehyde, or a formaldehyde equivalent, with a cyclic amine and
10 a heteroaromatic compound to give methylene linked cyclic amine deazapurines.

BACKGROUND

Nucleoside analogues that are potent inhibitors of purine nucleoside phosphorylase (PNP) and purine phosphoribosyltransferases (PPRT) are useful
15 in treating parasitic infections, T-cell malignancies, autoimmune diseases and inflammatory disorders {see e.g. V.L. Schramm, Biochimica et Biophysica Acta, 1587 (2002) 107-117}. The analogues are also useful for immunosuppression in organ transplantation.

20 Related nucleoside analogues that are potent inhibitors of 5'-methylthioadenosine phosphorylase (MTAP) and 5'-methylthioadenosine nucleosidase (MTAN) are useful:

- (a) as anti-microbial compounds, and in decreasing the virulence of
25 microbial infections by decreasing production of the quorum sensing pathway;
- (b) as agents for treating parasitic infections such as malaria that infects red blood cells {see e.g. G.A. Kicska et al., J. Biol. Chem., 277 (2002) 3226-3231}; and
- (c) as anti-tumour compounds, potentially in combination therapy with
30 methotrexate or azaserine.

The applicants have previously disclosed potent inhibitors of such enzymes in a class called the Immucillins, based upon deazapurines covalently linked directly



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to aza-sugar moieties (US Patent Nos. 5,985,848 and 6,066,722, "Inhibitors of Nucleoside Metabolism"; and WO 02/19371, "Nucleoside Metabolism Inhibitors").

In the search for new and improved nucleoside analogues that are potent inhibitors of the aforementioned nucleoside phosphorylases and nucleosidases, the applicants have also discovered a new class of compounds that are potent inhibitors of these nucleoside phosphorylases and hydrolases (PCT Patent Application PCT/NZ03/00186, "Inhibitors of Nucleoside Phosphorylases and Nucleosidases").

The preparation of such nucleoside analogues is by way of multi-step chemical syntheses. Consequently, the time and cost required for each synthesis can be considerable. There is therefore a need for more efficient and cost effective methods of preparing compounds in this new class.

The Mannich reaction is a condensation reaction between three components, namely an amine, formaldehyde and a compound with an active hydrogen atom such as a heteroaromatic compound e.g. indole (pages 812-814, *Vogel's Textbook of Practical Organic Chemistry*, 4th Edition, revised by B.S. Furniss, A.J. Hannaford, V. Rogers, P.W.G. Smith and A.R. Tatchell, Longmans, London, 1978).

Mannich reactions have been used to assemble compounds that incorporate a 9-deazapurine moiety linked via a methylene group to aliphatic and alicyclic amines [G.A. Modnikova et al., "Pyrrolo[3,2-d]pyrimidines. III. 7-Aminomethyl-substituted pyrrolo[3,2-d]pyrimidines", *Khim.-farm. Zh.*, 1983, 352-356 (English translation)]. Compounds that incorporate a pyrimidine moiety linked via a methylene group to a cyclic secondary amine have also been assembled using Mannich reactions. [V.V. Filichev and E.B. Pedersen, "Synthesis of 1'-aza-C-nucleosides from (3R,4R)-4-(hydroxymethyl)pyrrolidin-3-ol", *Tetrahedron*, 57 (2001) 9163-9168].

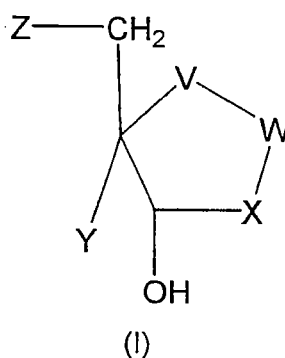
The applicants have now found that a Mannich reaction can be used to prepare compounds that incorporate a 9-deazapurine or an 8-aza-9-deazapurine moiety (or their 2-aza-analogues) linked via a methylene group to a cyclic secondary

amine. These compounds are described as potent inhibitors, or potentially potent inhibitors, of nucleoside phosphorylases and nucleosidases in PCT Patent Application PCT/NZ03/00186.

5 It is therefore an object of the present invention to provide a process for preparing these compounds, or at least to provide a useful choice.

STATEMENTS OF INVENTION

10 In a first aspect, the invention provides process for preparing a compound of the formula (I)



15 wherein:

V is selected from CH₂ and NH, and W is NR¹; or
V is NR¹, and W is selected from CH₂ and NH;

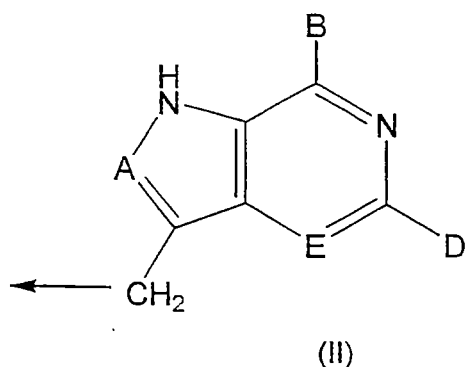
20 X is selected from CH₂ and CHOH in the R or S-configuration, except where W is selected from NH and NR¹, then X is CH₂;

Y is selected from hydrogen, halogen and hydroxy, except where V is selected from NH and NR¹, then Y is hydrogen;

25

Z is selected from hydrogen, halogen, hydroxy, a sulfonate leaving group, SQ, OQ and Q, where Q is an optionally substituted alkyl, aralkyl or aryl group; and

R¹ is a radical of the formula (II)



wherein:

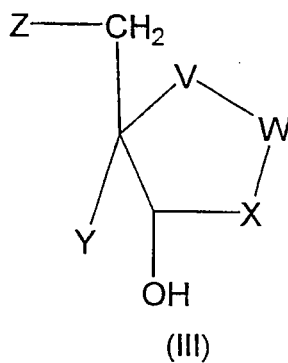
A is selected from N, CH and CR², where R² is selected from halogen, optionally substituted alkyl, aralkyl or aryl, OH, NH₂, NHR³, NR³R⁴ and SR⁵, where R³, R⁴ and R⁵ are each optionally substituted alkyl, aralkyl or aryl groups;

B is selected from OH, NH₂, NHR⁶, SH, hydrogen and halogen, where R⁶ is an optionally substituted alkyl, aralkyl or aryl group;

D is selected from OH, NH₂, NHR⁷, hydrogen, halogen and SCH₃, where R⁷ is an optionally substituted alkyl, aralkyl or aryl group; and

E is selected from N and CH;

including reacting a compound of the formula (III)



wherein:

V is selected from CH_2 and NH , and W is NH ; or

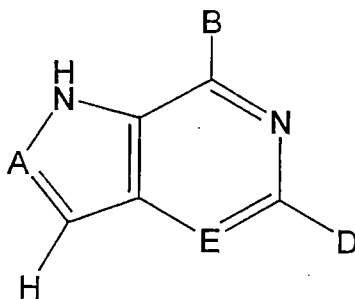
V is NH , and W is selected from CH_2 and NH ;

X is selected from CH_2 and CHOH in the R or S-configuration, except where W is NH , then X is CH_2 ;

Y is selected from hydrogen, halogen and hydroxy, except where V is selected from NH , then Y is hydrogen; and

Z is selected from hydrogen, halogen, hydroxy, a sulfonate leaving group, SQ, OQ and Q, where Q is an optionally substituted alkyl, aralkyl or aryl group;

with a compound of the formula (IV)



wherein A, B, D, and E are as defined above;

and with formaldehyde or a formaldehyde equivalent.

Preferably, Z is hydrogen, halogen, hydroxy, SQ or OQ, where Q is an optionally substituted alkyl, aralkyl or aryl group. It is also preferred that A is CH . It is further preferred that Y is H.

Preferably W is NR¹, V is CH₂ and X is CH₂. It is also preferred that R¹ is a radical of formula (II) as defined in claim 1, where A is CH and E is N.

5 It is further preferred that D is H or NH₂. Additionally, it is preferred that B is NH₂, OH or Cl.

Preferred processes of the invention include those where Z in the compound of formula (I) is methanesulfonate, *p*-toluenesulfonate or trifluoromethanesulfonate. Most preferably Z is methanesulfonate.

10

Preferred processes of the invention also include those where the compounds of formula (III) and (IV) are reacted with formaldehyde. Alternatively it is preferred that the compounds of formula (III) and (IV) are reacted with a formaldehyde equivalent such as paraformaldehyde.

15

The more preferred processes of the invention include those where the compound of formula (I) is:

(3R,4R)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-hydroxymethyl-pyrrolidine;

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(3R,4R)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(hydroxymethyl)-pyrrolidine;

(3R,4S)-4-(benzylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine;

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(3R,4S)-4-(4-chlorophenylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine;

(3R,4R)-1-[(6-chloro-9-deazapurin-9-yl)methyl]-3-hydroxy-4-(hydroxymethyl)-pyrrolidine

(3R,4R)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(methanesulfonyl)-pyrrolidine;

30

(3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(methylthiomethyl)-pyrrolidine;

(3R,4S)-4-(ethylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine;

(3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(propylthiomethyl)-pyrrolidine;

(3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(isopropylthiomethyl)-pyrrolidine;

5 (3R,4S)-4-(butylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine;

(3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(phenylthiomethyl)-pyrrolidine;

10 (3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-4-(4-fluorophenylthiomethyl)-3-hydroxy-pyrrolidine;

(3R,4S)-4-(3-chlorophenylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine;

(3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(cyclohexylthiomethyl)pyrrolidine;

15 (3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(4-pyridylthiomethyl)-pyrrolidine;

(3R,4R)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(methoxymethyl)-pyrrolidine;

20 (3R,4R)-4-(benzyloxymethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine;

(3R,4R)-1-[(9-deazaguanin-9-yl)methyl]-3-hydroxy-4-hydroxymethyl-pyrrolidine;

(3R,4S)-1-[(9-deazahypoxanthin-9-yl)-3-hydroxy-4-(propylthiomethyl)-pyrrolidine;

25 (3R,4S)-4-(butylthiomethyl)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxy-pyrrolidine;

(3R,4S)-1-[(9-deaza-6-chloro-purin-9-yl)methyl]-3-hydroxy-4-(2-phenylethyl)pyrrolidine;

(3R,4S)-1-[(9-deazaadenin-9-yl)methyl]-3-hydroxy-4-propyl-pyrrolidine;

30 (3R,4S)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-propyl-pyrrolidine; or

(3R,4S)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(methylthiomethyl)-pyrrolidine.

The invention also provides compound of formula (I) when prepared by a process according to claim 1.

In one embodiment, the invention provides a process for preparing a compound of formula (I) as defined in claim 1 including:

- (i) reacting a compound of formula (III) as defined in claim 1 with a compound of formula (IV) as defined in claim 1 and with formaldehyde or a formaldehyde equivalent, where any one or more of V, W, X, Y and Z of the compound of formula (III) is protected with a suitable protecting group; and
- (ii) removing the one or more protecting groups to give the compound of formula (I).

In another embodiment, the invention provides a process for preparing a compound of formula (I) as defined in claim 1 including:

- (i) reacting a compound of formula (III) as defined in claim 1 with a compound of formula (IV) as defined in claim 1 and with formaldehyde or a formaldehyde equivalent, where any one or more of A, B, D and E of the compound of formula (IV) is protected with a suitable protecting group; and
- (ii) removing the one or more protecting groups to give the compound of formula (I).

In still another embodiment, the invention provides a process of preparing a compound of formula (I) as defined in claim 1 including:

- (i) reacting a compound of formula (III) as defined in claim 1 with a compound of formula (IV) as defined in claim 1 and with formaldehyde or a formaldehyde equivalent, where any one or more of V, W, X, Y and Z of the compound of formula (III) is protected with a suitable protecting group and where any one or more of A, B, D and E of the compound of formula (IV) is protected with a suitable protecting group; and
- (ii) removing the one or more protecting groups to give the compound of formula (I).

DETAILED DESCRIPTION

The present invention provides a useful synthetic route to compounds that are potential inhibitors of PNP, PPRT, MTAN, MTAP and/or nucleoside hydrolases (NH). Such compounds may find use in the treatment of parasitic infections, T-cell malignancies, autoimmune diseases and inflammatory disorders. These compounds may also be used as antimicrobial agents, as antitumour agents, or as agents for treating parasitic infections.

Previous synthetic routes to compounds of formula (I) have been time-consuming and costly. In contrast, the present synthesis is a facile route to this useful class of compounds. The synthetic procedure involves using a Mannich reaction to couple a 9-deazapurine or an 8-aza-9-deazapurine moiety (or their 2-aza-analogues) to a cyclic secondary amine.

The applicants have therefore found that the desired compounds of formula (I) are advantageously prepared in good yield in a one-step synthesis.

It will be appreciated that the representation of a compound of formula (I), where B and/or D 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 use of the enol-type tautomeric representation is simply to allow fewer structural formulae to represent the compounds of the invention.

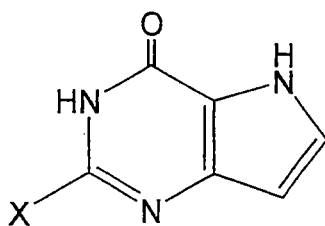
Similarly, it will be appreciated that the representation of a compound of formula (I), where B and/or D is a thiol group, is of the thioenol-type tautomeric form of a corresponding thioamide, and this will largely exist in the thioamide form. The use of the thioenol-type tautomeric representation is simply to allow fewer structural formulae to represent the compounds of the invention.

As used herein, the term "sulfonate leaving group" means an alkyl or aryl sulfonate such as methanesulfonate or benzenesulfonate, or a substituted form thereof such as bromobenzenesulfonate, trifluoromethanesulfonate or *p*-toluenesulfonate.

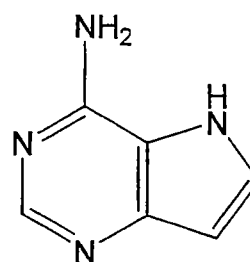
As used herein, the term "protecting group" means a group that selectively protects an organic functional group, temporarily masking the chemistry of that functional group and allowing other sites in the molecule to be manipulated without affecting the functional group. Suitable protecting groups are known to those skilled in the art and are described, for example, in *Protective Groups in Organic Synthesis* (3rd Ed.), T. W. Greene and P. G. M. Wuts, John Wiley & Sons Inc (1999).

Compounds of the formula (III) defined above may be prepared by known methods, as described in PCT Patent Application PCT/NZ03/00186 and the references cited therein. Procedures for the preparation of selected compounds of formula (III) are described herein.

Compounds of formula (IV) defined above may be prepared by known methods. In particular, processes for the preparation of the compounds 3*H*,5*H*-pyrrolo[3,2-*d*]pyrimidin-4-one (9-deazahypoxanthine) and 2-amino-3*H*,5*H*-pyrrolo[3,2-*d*]pyrimidin-4-one (9-deazaguanine), compounds **1** and **2** shown below, are described in PCT Patent Application PCT/NZ00/00048, "Process for Preparing Inhibitors of Nucleoside Metabolism and Substrates" and in R.H. Furneaux and P.C. Tyler, J. Org. Chem., 64 (1999) 8411-8412. Further, 9-deazaadenine (**3**) can be prepared by treatment of 9-deazahypoxanthine (**1**) with POCl₃ and then with ethanolic ammonia.



1 X = H
2 X = NH₂



3

One advantage of the applicants' new process is that neither the amine nor the heterocyclic component needs to have protecting groups on the functional groups

that are not directly involved in the reaction chemistry. Nevertheless, there may be occasions where it is advantageous to utilize a protected form of a compound of formula (III) and/or formula (IV) as components in the reaction.

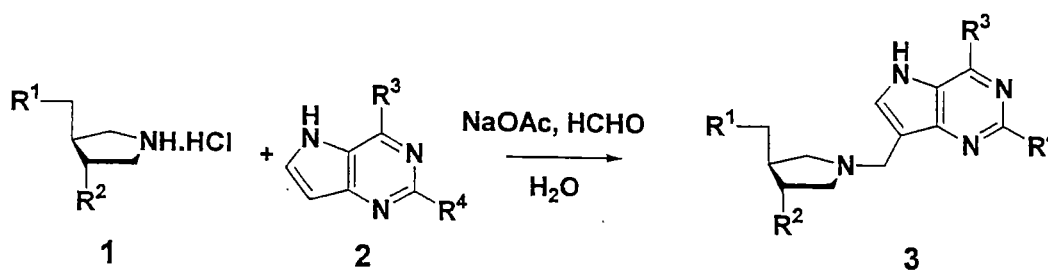
Suitably protected forms of compounds of formula (III) are described in US Patent Nos. 5,985,848 and 6,066,722, "Inhibitors of Nucleoside Metabolism" and WO 02/19371, "Nucleoside Metabolism Inhibitors". It is essential that suitably protected forms of compounds of the formula (IV) have a proton at position-9 of the 9-deazapurine or 8-aza-9-deazapurine moiety (or their 2-aza-analogues).

Suitably protected forms of compounds of formula (IV) are described in PCT Patent Application PCT/NZ03/00186, "Inhibitors of Nucleoside Phosphorylases and Nucleosidases". It is essential that protected forms of compounds of the formula (III) have an unprotected ring amino-group.

EXAMPLES

The following examples further illustrate the invention. It is to be appreciated that the invention is not limited to the examples.

Example 1: Mannich Reaction - General Procedure



Scheme 1

General procedure for the preparation of compounds of formula (3) using the Mannich reaction shown in Scheme 1; Pyrrolidine hydrochlorides of formula (1) (1.0 mol equiv.; as listed in Table 1 as "Amine reactant" unless otherwise specified) and sodium acetate (1.0 mol. equiv.) were dissolved in water

and 1,4-dioxane (4:1 v/v, 2 mL per mmol) and to the solution were added aqueous formaldehyde (1.0 – 1.5 mol. equiv.) and the substituted 9-deazapurine of formula (2) (0.8 – 1.5 mol equiv.). The reaction was stirred at the temperature and for the time shown in Table 1. Silica gel (1.0 g per mmol of 1) was added and the mixture was evaporated to dryness. Purification by chromatography on silica gel, using gradient elution with CH₂Cl₂: MeOH: NH₄OH (95:5:1→80:20:1 v/v/v) as the eluent, afforded the compound of formula (3) as detailed in Table 1 as the free base or partial acetic acid salt, which was converted to the HCl salt by addition and evaporation of excess conc. HCl [Evans, G.B.; Furneaux, R.H.; Tyler, P.C.; Schramm, V.L. *Org. Lett.* **2003**, 5, 3639-3640.] The preparations of the pyrrolidine hydrochlorides of formula (1) are exemplified in the Preparative Examples.

Table 1: Compounds Prepared via the Mannich Reaction General Procedure

Cpd No.	Temp (° C)	Amine reactant	Reaction Time (h)	Substituents				Yield (%)
				R ¹	R ²	R ³	R ⁴	
4	95	5	16	OH	OH	OH	H	47
6	95	5	1	OH	OH	NH ₂	H	65
7	95	41	1	S _{Bn}	OH	NH ₂	H	72
8	95	39	1	S _{Ph-p-Cl}	OH	NH ₂	H	72
9	95	5	3	OH	OH	Cl	H	78
10	90	53	1	OSO ₂ Me	OH	NH ₂	H	39
11	95	32	1	S _{Me}	OH	NH ₂	H	39
12	85	33	2	S _{Et}	OH	NH ₂	H	60
13	90	34	3	S _{n-Pr}	OH	NH ₂	H	59
14	95	35	3	S _{iso-Pr}	OH	NH ₂	H	38
15	90	36	3	S _{Bu}	OH	NH ₂	H	52
16	95	37	1	S _{Ph}	OH	NH ₂	H	57
17	90	38	1	S _{Ph-p-F}	OH	NH ₂	H	27
18	90	40	1.5	S _{Ph-m-Cl}	OH	NH ₂	H	52
19	95	44	3	S _{Chx}	OH	NH ₂	H	34
20	85	45	2	S-4-pyridyl	OH	NH ₂	H	45
21	85	42	1.5	OMe	OH	NH ₂	H	59
22	90	43	1.5	OBn	OH	NH ₂	H	22

23	95	5	12	OH	OH	OH	NH ₂	57
24	90	34	3	SPr	OH	OH	H	78
25	90	36	3	SBu	OH	OH	H	72
26	95	49	12	C-Bn	OH	Cl	H	38
27	95	52	2.5	C-Et	OH	NH ₂	H	31
28	95	52	5	C-Et	OH	OH	H	36

Example 1.01

(3R,4R)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-hydroxymethyl-pyrrolidine (4). Starting with 9-deazahypoxanthine (Furneaux and Tyler, *J. Org. Chem.*, **1999**, 64, 8411-8412) and (3R,4R)-3-hydroxy-4-hydroxymethyl-pyrrolidine hydrochloride (5) (Evans et al, *J. Med. Chem.*, **2003**, 46 5271-5276), the Mannich reaction general procedure (above) was followed to afford compound 4 as the acetic acid salt. After conversion to the HCl salt and ¹H and ¹³C NMR spectra analysis, the compound was found to be identical in all respects with that previously reported (Evans et al *J. Med. Chem.* **2003**, 46, 5271-5276).

Example 1.02

(3R,4R)-1-[(9-Deaza-adenin-9-yl)methyl]-3-hydroxy-4-(hydroxymethyl)-pyrrolidine (6). Starting with 9-deaza-adenine (Preparative Example 3.01) and (3R,4R)-3-hydroxy-4-hydroxymethyl-pyrrolidine (5), the Mannich reaction general procedure (above) was followed to afford compound 6 as the acetic acid salt. ¹H NMR (d₄-MeOH) δ 8.20 (s, 1H), 7.65 (s, 1H), 4.27 (s, 1H), 4.22 (quintet, J = 3.0 Hz, 1H), 3.59 (m, 2H), 3.46 (dd, J = 11.1, 8.3 Hz, 1H), 3.26 (dd, J = 11.4, 5.7 Hz, 1H), 3.11 (dd, J = 11.4, 3.0 Hz, 1H), 2.95 (dd, J = 11.2, 6.8 Hz, 1H), 2.37 (brs, 1H), 1.82 (s, 3H). ¹³C NMR (d₄-MeOH) 152.9, 151.9, 147.1, 132.0, 115.8, 108.2, 73.6, 63.1, 61.9, 56.0, 50.8, 49.5, 23.7. HRMS (MH⁺) calc for C₁₂H₁₈N₅O₂: 264.1461. Found 264.1457.

Example 1.03

(3R,4S)-4-(Benzylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine (7). The Mannich reaction general procedure (above) was followed to afford compound 7 as the acetic acid salt. The acetic acid salt was converted to the free base via ion exchange chromatography. ¹H NMR (d₄-MeOH) 8.17 (s, 1H), 7.46 (s, 1H), 7.26 – 7.16 (m, 5H), 3.93 – 3.90 (m, 1H), 3.83 – 3.74 (m, 2H),

3.68 (s, 2H), 3.03 – 2.97 (m, 1H), 2.80 (dd, $J = 10.2, 6.4$ Hz, 1H), 2.66 – 2.58 (m, 2H), 2.38 (dd, $J = 12.5, 8.9$ Hz, 1H), 2.30 (dd, $J = 9.5, 7.2$ Hz, 1H), 2.20 – 2.14 (m, 1H). ^{13}C NMR ($\text{d}_4\text{-MeOH}$) 152.5, 151.4, 147.4, 140.4, 130.4, 130.4, 129.8, 115.5, 112.9, 77.3, 62.7, 59.2, 49.3, 48.6, 37.5, 35.6. HRMS (MH^+) calc for $\text{C}_{19}\text{H}_{24}\text{N}_5\text{OS}$: 370.1702. Found 370.1694.

Example 1.04

(3R,4S)-4-(4-Chlorophenylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-

hydroxy-pyrrolidine (8). The Mannich reaction general procedure (above) was followed to afford compound **8** as the acetic acid salt. ^1H NMR ($\text{d}_4\text{-MeOH}$) 8.25 (s, 1H), 7.84 (s, 1H), 7.35 – 7.23 (m, 5H), 4.54 (s, 2H), 4.30 (m, 1H), 3.74 (dd, $J = 11.9, 7.9$ Hz, 1H), 3.59 (dd, $J = 12.2, 5.6$ Hz, 1H), 3.40 – 3.15 (m, 4H), 2.89 (dd, $J = 13.5, 9.1$ Hz, 1H), 2.47 (brs, 1H), 1.98 (s, 3H). ^{13}C NMR ($\text{d}_4\text{-MeOH}$) 153.0, 151.8, 146.1, 135.7, 134.0, 133.2, 132.2, 130.7, 115.7, 105.5, 74.6, 60.4, 57.3, 49.2, 47.7, 36.1, 23.0. HRMS (MH^+) calc for $\text{C}_{18}\text{H}_{21}\text{ClN}_5\text{OS}$: 390.1155. Found 390.1264.

Example 1.05

(3R,4R)-1-[(6-Chloro-9-deazapurin-9-yl)methyl]-3-hydroxy-4-

(hydroxymethyl)-pyrrolidine (9). Starting with 6-chloro-9-deazapurine (K. Imai, *Chem. Pharm. Bull.*, **1964**, 12, 1030) and (3R,4R)-3-hydroxy-4-hydroxymethyl-pyrrolidine, the Mannich reaction general procedure (above) was followed to afford compound **9** as the acetic acid salt. ^1H NMR (D_2O) 8.34 (s, 1H), 7.98 (s, 1H), 4.48 (s, 2H), 4.31 (m, 1H), 3.68 (dd, $J = 12.1, 8.3$ Hz, 1H), 3.53 (d, $J = 5.9$ Hz, 2H), 3.45 (dd, $J = 12.6, 5.5$ Hz, 1H), 3.32 (dd, $J = 12.6, 2.5$ Hz, 1H), 3.13 (dd, $J = 12.0, 7.4$ Hz, 1H), 2.40 (brs, 1H), 1.82 (s, 3H). ^{13}C NMR ($\text{d}_4\text{-MeOH}$) 149.7, 148.6, 143.4, 137.6, 124.8, 104.5, 71.3, 60.7, 59.8, 54.4, 48.0, 47.8, 23.5. HRMS (MH^+) calc for $\text{C}_{12}\text{H}_{16}\text{ClN}_4\text{O}_2$: 283.0962. Found 283.0973.

Example 1.06

(3R,4R)-1-[(9-Deaza-adenin-9-yl)methyl]-3-hydroxy-4-(methanesulfonyl)-

pyrrolidine (10). The Mannich reaction general procedure (above) was followed to afford compound **10**. ^1H NMR ($\text{d}_4\text{-MeOH}$) 8.17 (s, 1H), 7.52 (s, 1H), 4.30 – 3.82 (m, 5H), 3.10 – 3.00 (m, 1H), 3.06 (s, 3H), 2.94 (dd, $J = 10.3, 6.3$ Hz, 1H),

2.71 (dd, $J = 10.3, 4.1$ Hz, 1H), 2.53 (dd, $J = 10.1, 6.7$ Hz, 1H), 2.43 – 2.34 (m, 1H). ^{13}C NMR (d_4 -MeOH) 152.6, 151.5, 147.2, 130.7, 115.6, 112.0, 73.8, 71.8, 62.4, 56.0, 49.4, 49.1, 37.5.

5 **Example 1.07**

(3R,4S)-1-[(9-Deaza-adenin-9-yl)methyl]-3-hydroxy-4-(methylthiomethyl)-pyrrolidine (11). The Mannich reaction general procedure (above) was followed to afford compound **11** as the acetic acid salt. After conversion to the HCl salt and ^1H and ^{13}C NMR spectra analysis, the compound was found to be identical in
10 all respects with that previously reported. (Evans et al., *J. Med. Chem.* **2003**, 46, 5271-5276).

Example 1.08

(3R,4S)-4-(Ethylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine (12). The Mannich reaction general procedure (above) was followed to afford compound **12**. ^1H NMR (d_4 -MeOH) 8.16 (s, 1H), 7.52 (s, 1H), 4.00 – 3.82 (m, 3H), 3.12 (dd, $J = 9.9, 7.9$ Hz, 1H), 2.92 (dd, $J = 10.5, 6.3$ Hz, 1H), 2.76 – 2.68 (m, 2H), 2.55 – 2.41 (m, 4H), 2.25 – 2.15 (m, 1H), 1.21 (t, $J = 7.4$ Hz, 3H). ^{13}C NMR (d_4 -MeOH) 152.5, 151.5, 147.3, 130.7, 115.6, 112.1, 77.0, 62.4, 59.1,
15 49.4, 48.8, 35.5, 27.2, 15.5. HRMS (MH^+) calc for $\text{C}_{14}\text{H}_{22}\text{N}_5\text{OS}$: 308.1540. Found 308.1535.

Example 1.09

(3R,4S)-1-[(9-Deaza-adenin-9-yl)methyl]-3-hydroxy-4-(propylthiomethyl)-pyrrolidine (13). The Mannich reaction general procedure (above) was followed to afford compound **13**. ^1H NMR (d_4 -MeOH) 8.17 (s, 1H), 7.50 (s, 1H), 4.00 – 3.79 (m, 3H), 3.08 (dd, $J = 9.8, 7.9$ Hz, 1H), 2.86 (dd, $J = 10.3, 6.4$ Hz, 1H), 2.72 – 2.62 (m, 2H), 2.50 – 2.38 (m, 4H), 2.22 – 2.12 (m, 1H), 1.55 (sextet, $J = 7.3$ Hz, 2H), 0.95 (t, $J = 7.3$ Hz, 3H). ^{13}C NMR (d_4 -MeOH) 152.5, 151.4, 147.4, 130.5,
25 115.6, 112.7, 77.2, 62.6, 59.2, 49.4, 49.0, 36.1, 35.6, 24.3, 14.1. HRMS (MH^+) calc for $\text{C}_{15}\text{H}_{24}\text{N}_5\text{OS}$: 322.1696. Found 322.1709.

Example 1.10

(3R,4S)-1-[(9-Deaza-adenin-9-yl)methyl]-3-hydroxy-4-(isopropylthiomethyl)-pyrrolidine (14). A variation of the Mannich reaction general procedure (above) using 20% 1,4-dioxane in water as the solvent and 0.9 mol equiv. of 9-deazaadenine afforded the crude title compound **14** (386 mg, 80%) after column chromatography on silica eluting with CH₂Cl₂ : MeOH : NH₄OH (8 : 1.8 : 0.2). Residual impurities could be removed by column chromatography on silica eluting with CH₂Cl₂ : NH₃ (7N) in MeOH to afford title compound **14** (183 mg, 38%). ¹H NMR (MeOH-d₄): δ ppm: 8.16 (s, 1H), 7.49 (s, 1H), 3.99-3.94 (m, 1H), 3.82 (dd, J = 18.7, 13.4 Hz, 1H), 3.04 (dd, J = 9.7, 7.9 Hz, 1H), 2.95-2.82 (m, 2H), 2.75 (dd, J = 12.5, 6.0 Hz, 1H), 2.66 (dd, J = 10.3, 4.2 Hz, 1H), 2.50 (dd, J = 12.5, 9.1 Hz, 1H), 2.38 (dd, J = 9.7, 7.1 Hz, 1H), 2.21-2.10 (m, 1H), 1.23, 1.22 (2s, 3H each). ¹³C NMR (MeOH-d₄): δ ppm: 152.48, 151.38, 147.40, 130.45, 115.54, 112.90, 77.29, 62.66, 59.26, 49.32, 49.09, 36.49, 34.66, 24.19.

Example 1.11

(3R,4S)-4-(Butylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine (15). The Mannich reaction general procedure (above) was followed to afford compound **15**. ¹H NMR (d₄-MeOH) 8.16 (s, 1H), 7.50 (s, 1H), 3.99 – 3.79 (m, 3H), 3.08 (dd, J = 9.7, 7.9 Hz, 1H), 2.87 (dd, J = 10.3, 6.4 Hz, 1H), 2.75 – 2.69 (m, 2H), 2.51 – 2.38 (m, 4H), 2.22 – 2.12 (m, 1H), 1.55 – 1.32 (m, 4H), 0.90 (t, J = 7.3 Hz, 3H). ¹³C NMR (d₄-MeOH) 152.5, 151.4, 147.4, 130.5, 115.6, 112.6, 77.1, 62.6, 59.2, 49.4, 49.0, 36.1, 33.3, 33.2, 23.3, 14.4. HRMS (MH⁺) calc for C₁₈H₂₆N₅OS: 336.1853. Found 336.1850.

Example 1.12

(3R,4S)-1-[(9-Deaza-adenin-9-yl)methyl]-3-hydroxy-4-(phenylthiomethyl)-pyrrolidine (16). The Mannich reaction general procedure (above) was followed to afford compound **16**. ¹H NMR (d₄-MeOH) 8.22 (s, 1H), 7.74 (s, 1H), 7.33 – 7.15 (m, 2H), 4.43 (s, 2H), 4.26 (m, 1H), 3.62 (dd, J = 11.7, 7.9 Hz, 2H), 3.48 (dd, J = 12.0, 5.6 Hz, 1H), 3.25 (t, dd, J = 12.0, 3.3 Hz, 1H), 3.15 (m, 2H), 2.85 (dd, J = 13.5, 9.1 Hz, 1H), 2.43 (m, 1H). ¹³C NMR (d₄-MeOH) 152.9, 152.1, 146.8, 136.8, 132.7, 131.4, 130.6, 128.1, 115.8, 106.3, 74.9, 60.6, 57.4, 49.7, 47.7, 36.3. HRMS (MH⁺) calc for C₁₈H₂₅N₅OS: 356.1545. Found 356.1542.

Example 1.13**(3R,4S)-1-[(9-Deaza-adenin-9-yl)methyl]-4-(4-fluorophenylthiomethyl)-3-**

hydroxy-pyrrolidine (17). The Mannich reaction general procedure (above) was followed to afford compound **17**. ¹H NMR (d₄-MeOH) 8.16 (s, 1H), 7.46 (s, 1H), 7.40 – 7.30 (m, 2H), 7.00 – 6.90 (m, 2H), 4.02 – 3.97 (m, 1H), 3.86 – 3.75 (m, 2H), 3.11 (dd, *J* = 12.9, 5.9 Hz, 1H), 3.00 (t, *J* = 8.7 Hz, 1H), 2.90 – 2.75 (m, 2H), 2.65 – 2.59 (m, 1H), 2.41 – 2.32 (m, 1H), 2.20 – 2.10 (m, 1H). ¹³C NMR (d₄-MeOH) 165.5, 162.0, 152.5, 151.4, 147.4, 134.1, 134.0, 133.1, 130.4, 117.4, 117.1, 115.5, 112.9, 77.2, 62.7, 59.0, 49.3, 48.8, 39.1. HRMS (MH⁺) calc for C₁₈H₂₁N₅OFS: 374.1445. Found 374.1438.

Example 1.14**(3R,4S)-4-(3-Chlorophenylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-**

hydroxy-pyrrolidine (18). The Mannich reaction general procedure (above) was followed to afford compound **18**. ¹H NMR (d₄-MeOH) 8.16 (s, 1H), 7.46 (s, 1H), 7.25 – 7.05 (m, 4H), 4.01 – 3.97 (m, 1H), 3.87 – 3.76 (m, 2H), 3.18 (dd, *J* = 12.9, 5.9 Hz, 1H), 2.99 (dd, *J* = 9.8, 7.9 Hz, 1H), 2.94 – 2.86 (m, 2H), 2.64 (dd, *J* = 10.2, 4.3 Hz, 1H), 2.41 (dd, *J* = 9.9, 7.0 Hz, 1H), 2.26 – 2.15 (m, 1H). ¹³C NMR (d₄-MeOH) 152.5, 151.4, 147.4, 140.7, 136.1, 131.6, 130.4, 129.8, 128.6, 127.4, 115.5, 112.8, 77.1, 62.6, 58.9, 49.3, 48.7, 37.4. HRMS (MH⁺) calc for C₁₈H₂₁N₅OCIS: 390.1150. Found 390.1142.

Example 1.15**(3R,4S)-1-[(9-Deaza-adenin-9-yl)methyl]-3-hydroxy-4-**

(cyclohexylthiomethyl)pyrrolidine (19). A variation of the Mannich reaction general procedure (above) using 20% 1,4-dioxane in water as the solvent and 0.9 mol equiv. of 9-deazaadenine afforded the crude title compound **19** (333 mg, 79%) after column chromatography on silica eluting with CH₂Cl₂ : MeOH : NH₄OH (8:1.8:0.2 v/v/v). Residual impurities could be removed by column chromatography on silica eluting with CH₂Cl₂ : NH₃ (7N) in MeOH (9:1 v/v) to afford title compound **19** (144 mg, 34%). ¹H NMR (MeOH-d₄): δ ppm: 8.15 (s, 1H), 7.50 (s, 1H), 3.97-3.92 (m, 1H), 3.82 (dd, *J* = 19.1, 13.4 Hz, 2H), 3.06-3.00 (m, 1H), 2.84 (dd, *J* = 10.3, 6.4 Hz, 1H), 2.75 (dd, *J* = 12.5, 5.9 Hz, 1H), 2.67-2.58

(m, 2H), 2.48 (dd, $J = 12.5, 9.3$ Hz, 1H), 2.37 (dd, $J = 9.8, 7.2$ Hz, 1H), 2.20-2.08 (m, 1H), 1.94-1.92 (m, 2H), 1.74-1.72 (m, 2H), 1.60-1.58 (m, 1H), 1.36-1.19 (m, 5H). ^{13}C NMR (MeOH- d_4): δ ppm: 152.48, 151.35, 147.32, 130.50, 115.48, 112.74, 77.21, 62.62, 59.18, 49.38, 49.26, 45.10, 35.27, 35.20, 34.18, 27.48, 27.39.

Example 1.16

(3R,4S)-1-[(9-Deaza-adenin-9-yl)methyl]-3-hydroxy-4-(4-pyridylthiomethyl)-pyrrolidine (20). The Mannich reaction general procedure (above) was followed to afford compound **20**. ^1H NMR (D_2O) 8.43 (d, $J = 7.2$ Hz, 1H), 8.42 (s, 1H), 8.00 (s, 1H), 7.77 (d, $J = 7.2$ Hz, 1H), 4.64 (s, 2H), 4.52 – 4.47 (m, 1H), 3.94 (dd, $J = 12.1, 8.0$ Hz, 1H), 3.67 (dd, $J = 12.6, 5.7$ Hz, 1H), 3.50 – 3.15 (m, 4H), 2.78 – 2.64 (m, 1H). ^{13}C NMR (D_2O) 163.9, 150.2, 144.6, 139.5, 135.4, 122.8, 113.2, 102.7, 73.0, 59.0, 55.9, 48.1, 44.4, 31.5. HRMS (MH^+) calc for $\text{C}_{17}\text{H}_{21}\text{N}_6\text{OS}$: 357.1492. Found 357.1509.

Example 1.17

(3R,4R)-1-[(9-Deaza-adenin-9-yl)methyl]-3-hydroxy-4-(methoxymethyl)-pyrrolidine (21). The Mannich reaction general procedure (above) was followed to afford compound **21**. ^1H NMR (d_4 -MeOH) 8.19 (s, 1H), 7.63 (s, 1H), 4.18 – 4.05 (m, 3H), 3.40 – 2.28 (m, 3H), 3.30 (s, 3H), 3.10 (dd, $J = 11.0, 5.7$ Hz, 1H), 2.95 (dd, $J = 11.0, 3.3$ Hz, 1H), 2.77 (dd, $J = 10.8, 6.7$ Hz, 1H), 2.41 – 2.29 (m, 1H). ^{13}C NMR (d_4 -MeOH) 152.7, 151.8, 147.2, 131.6, 115.7, 109.5, 74.3, 74.1, 62.2, 59.6, 56.5, 49.4, 49.0. HRMS (MH^+) calc for $\text{C}_{13}\text{H}_{20}\text{N}_5\text{O}_2$: 278.1612. Found 278.1626.

Example 1.18

(3R,4R)-4-(Benzyloxymethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine (22). The Mannich reaction general procedure (above) was followed to afford compound **22**. ^1H NMR (d_4 -MeOH) 8.17 (s, 1H), 7.55 (s, 1H), 7.30 – 7.20 (m, 5H), 4.46 (bs, 2H), 4.10 – 4.00 (m, 3H), 3.55 – 3.38 (m, 2H), 3.23 – 3.18 (m, 1H), 2.98 (dd, $J = 10.7, 5.8$ Hz, 1H), 2.85 (dd, $J = 10.7, 3.4$ Hz, 1H), 2.68 (dd, $J = 10.4, 6.9$ Hz, 1H), 2.38 – 2.30 (m, 1H). ^{13}C NMR (d_4 -MeOH) 152.6, 151.7,

147.2, 139.9, 131.3, 129.8, 129.3, 129.1, 115.6, 110.4, 74.5, 74.3, 71.9, 62.3, 56.6, 49.4, 49.0.

Example 1.19

5 **(3R,4R)-1-[(9-Deazaguanin-9-yl)methyl]-3-hydroxy-4-hydroxymethyl-pyrrolidine (23).** (3R,4R)-3-Hydroxy-4-hydroxymethyl-pyrrolidine hydrochloride (**5**) (154 mg, 1.0 mmol) and sodium acetate (82 mg, 1.0 mmol) were dissolved in water (2 mL) and to the solution were added aqueous formaldehyde (82 μ L, 1.0 mmol) and 9-deazaguanine (Furneaux and Tyler, *J. Org. Chem.*, **1999**, 64, 8411-10 8412) (120 mg, 0.8 mmol). The reaction was stirred at 95 °C for 12 h. Silica gel (1.0 g) was added and the mixture was evaporated to dryness. Purification by chromatography on silica gel, using CH₂Cl₂: MeOH: NH₄OH (5:4:1 v/v/v) as the eluent, afforded title compound **23** as the acetic acid salt. After conversion to the HCl salt and ¹H and ¹³C NMR spectra analysis, the compound was found to be 15 identical in all respects with that previously reported (Evans et al., *J. Med. Chem.* **2003**, 46, 5271-5276).

Example 1.20

20 **(3R,4S)-1-[(9-Deazahypoxanthin-9-yl)-3-hydroxy-4-(propylthiomethyl)-pyrrolidine (24).** The Mannich reaction general procedure (above) was followed to afford compound **24**. ¹H NMR (d₄-MeOH/D₂O) 7.99 (s, 1H), 7.53 (s, 1H), 4.06 – 3.98 (m, 1H), 3.92 – 3.80 (m, 2H), 3.06 (dd, *J* = 9.8, 8.0 Hz, 1H), 2.90 (dd, *J* = 10.5, 6.5 Hz, 1H), 2.79 – 2.65 (m, 2H), 2.52 – 2.38 (m, 4H), 2.22 – 2.15 (m, 1H), 1.57 (sextet, *J* = 7.3 Hz, 2H), 0.95 (t, *J* = 7.3 Hz, 3H). ¹³C NMR (d₄-MeOH) 145.7, 25 143.7, 131.0, 113.6, 77.0, 62.1, 58.6, 48.9, 48.5, 35.9, 35.6, 24.2, 14.2.

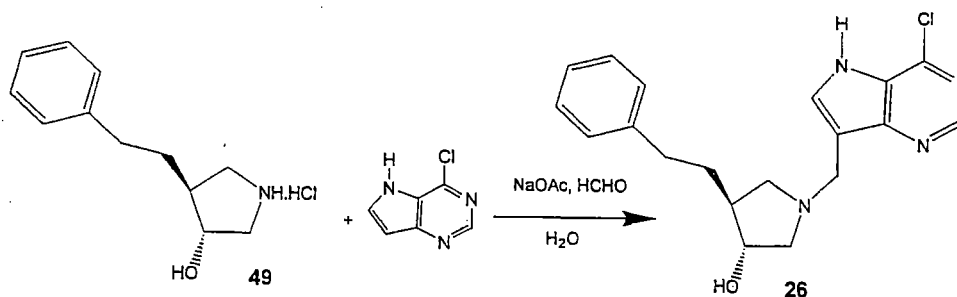
Example 1.21

30 **(3R,4S)- 4-(Butylthiomethyl)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxy-pyrrolidine (25).** The Mannich reaction general procedure (above) was followed to afford compound **25**. ¹H NMR (d₄-MeOH/CDCl₃) 7.86 (s, 1H), 7.38 (s, 1H), 4.00 – 3.92 (m, 1H), 3.82 – 3.75 (m, 2H), 3.07 (dd, *J* = 9.8, 8.0 Hz, 1H), 2.85 – 2.70 (m, 3H), 2.55 – 2.42 (m, 3H), 2.37 – 2.15 (m, 2H), 1.60 – 1.32 (m, 4H), 0.90 (t, *J* = 7.3 Hz, 3H). ¹³C NMR (d₄-MeOH/D₂O) 156.6, 145.6, 142.9, 129.9, 119.6,

114.7, 77.4, 63.0, 59.6, 49.4, 49.1, 36.6, 33.6, 33.3, 23.5, 14.9. HRMS (MH^+)
calc for $C_{16}H_{25}N_4O_2S$: 337.1693. Found 337.1684.

Example 1.22

(3R,4S)-1-[(9-Deaza-6-chloro-purin-9-yl)methyl]-3-hydroxy-4-(2-phenylethyl)pyrrolidine (**26**). A variation of the Mannich reaction general procedure (above) using 0.9 mol equiv. of 6-chloro-9-deazapurine (K. Imai, *Chem. Pharm. Bull.*, **1964**, 12, 1030) afforded the title compound **26** (Scheme 2). In comparison to the standard procedure the reaction mixture did not form a solution but a brown slurry which was diluted with 1,4-dioxane before being preabsorbed onto silica gel. Column chromatography eluting with CH_2Cl_2 : MeOH (4:1 v/v) followed by CH_2Cl_2 : MeOH : NH_4OH (5:4.5:0.5 v/v/v) gave **26** in 38% yield. 1H NMR (300 MHz, MeOH- d_4): δ ppm: 8.71 (s, 1H), 8.12 (s, 1H), 7.17 (s, 5H), 4.55 (s, 1H), 4.18 (m, 1H), 3.56 (m, 2H), 3.31 (m, 1H), 3.04 (dd, J = 11.6, 7.7 Hz, 1H), 2.64 (m, 2H), 2.21 (m, 1H), 1.87 (m, 1H), 1.61 (m, 1H). ^{13}C NMR (300 MHz, MeOH- d_4): δ ppm: 151.62, 151.35, 145.02, 142.99, 138.11, 129.84, 129.82, 127.46, 126.81, 107.73, 75.68, 61.00, 58.48, 49.51, 47.56, 35.26, 34.88.



Scheme 2

Example 1.23

(3R,4S)-1-[(9-Deazaadenin-9-yl)methyl]-3-hydroxy-4-propyl-pyrrolidine (**27**). A variation of the Mannich reaction general procedure (above) using 0.9 mol equiv. of 9-deazaadenine afforded the crude title compound **27** (136 mg, 73%) after column chromatography on silica eluting with CH_2Cl_2 : MeOH : NH_4OH (8:1.8:0.2 v/v/v). Residual impurities could be removed by column chromatography on silica eluting with MeCN : NH_4OH (4:1 v/v) to give **27** in 31%

yield. NMR (300 MHz, MeOH-d₄): δ ppm: 8.18 (s, 1H), 7.51 (s, 1H), 3.91-3.85 (m, 3H), 3.10 (dd, J = 9.6, 8.0 Hz, 1H), 2.82-2.72 (m, 2H), 2.22 (dd, J = 9.6, 8.0 Hz, 1H), 2.04-1.95 (m, 1H), 1.56-1.44 (m, 1H), 1.39-1.21 (m, 3H), 0.92-0.87 (m, 3H). ¹³C NMR (300 MHz, MeOH-d₄): δ ppm: 152.52, 151.45, 147.37, 130.59, 115.55, 112.50, 77.94, 62.63, 59.94, 49.55, 48.59, 36.89, 22.67, 14.91.

Example 1.24

(3R,4S)-1-[(9-Deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-propyl-pyrrolidine (28).

A variation of the Mannich reaction general procedure (above) using 0.9 mol equiv. of 9-deazahypoxanthine afforded the crude title compound **28** (90 mg, 61%) after column chromatography on silica eluting with CH₂Cl₂ : MeOH : NH₄OH (5:4.5:0.5 v/v/v). Residual impurities (9-deazahypoxanthine) could be removed by column chromatography on silica eluting with CH₂Cl₂ : MeOH : NH₄OH (8:1.8:0.2 v/v/v) to give **28** in 36 % yield. NMR (300 MHz, MeOH-d₄, ~30% CDCl₃): δ ppm: 7.86 (s, 1H), 7.39 (s, 1H), 3.89-3.76 (m, 3H), 3.09 (dd, J = 9.5, 7.9 Hz, 1H), 2.79-2.69 (m, 2H), 2.18-2.12 (m, 1H), 2.05-1.96 (m, 1H), 1.55-1.46 (m, 1H), 1.41-1.23 (m, 3H), 0.94-0.89 (m, 3H). ¹³C NMR (300 MHz, MeOH-d₄, ~30% CDCl₃): δ ppm: 145.59, 142.99, 129.93, 114.50, 78.14, 62.95, 60.24, 49.78, 48.94, 37.07, 22.80, 15.29.

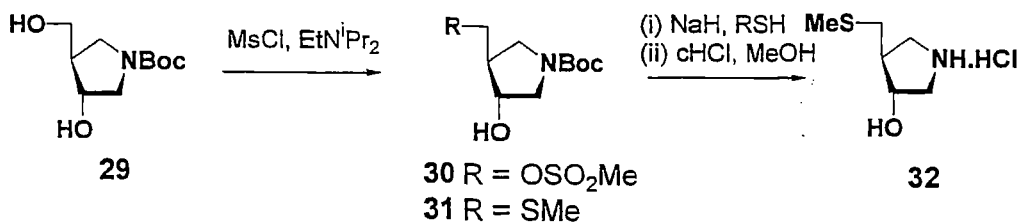
Preparative Example 1

Procedures for the synthesis of (3R,4S)-3-hydroxy-4-(alkyl-, aralkyl-, and aryl-thiomethyl)pyrrolidines.

General Preparative method. 4-Substituted-4-thiopyrrolidines were prepared essentially following the method detailed in Preparative Example 1.01 for compound **32**, but with the modifications noted for each Preparative Example and using the appropriate sodium thiolate. In cases when the sodium thiolate was not directly available it was pre-formed by treating a stirred mixture of NaH (2.85 mmol) in DMF (5 mL) at 0 °C with the appropriate thiol (2.85 mmol). After stirring the mixture for 10 min, a solution of the mesylate (450 mg, 1.53 mmol) was added as a solution in DMF (5 mL) and the mixture was stirred at RT until the complete consumption of the mesylate was observed (0.5 – 4 h) by TLC.

Preparative Example 1.01**(3R,4S)-3-Hydroxy-4-(methylthiomethyl)-pyrrolidine (32), Scheme 3.**

Methanesulfonyl chloride (0.950 mL, 12.3 mmol) was added to a solution of (3R,4R)-*N*-*tert*-butoxycarbonyl-3-hydroxy-4-hydroxymethyl-pyrrolidine (29) (Evans et al., *J. Med. Chem.*, **2003**, 46, 5271-5276) (2.16 g, 9.94 mmol) and diisopropylethyl amine (2.65 mL, 15.0 mmol) in DCM (40 mL) cooled to -78 °C over 5 min. After stirring at -78 °C for 40 min, aqueous 2M HCl was added, the organic phase was separated. The aqueous phase was extracted with DCM (x2). The combined organic extract was washed with sat. aqueous NaHCO₃ then brine and dried (MgSO₄). Normal processing and chromatography gave (1.99 g, 6.74 mmol, 68%) of mesylate **30** as a colourless glass. HRMS (MH⁺) calc for C₁₁H₂₁NO₆SNa: 318.0982. Found 318.0979. A solution of mesylate **30** (450 mg, 1.53 mmol) in DMF (5 mL) was added to a stirred solution of sodium thiomethoxide (200 mg, 2.85 mmol) in DMF (3 mL) and the mixture was stirred for 1 h. Toluene (50 mL) and H₂O (50 mL) were added and shaken, the phases separated, the organic layer dried (MgSO₄) and the solvent removed at reduced pressure. The crude product that was purified by chromatography on silica, eluting with 10 – 50% EtOAc/petroleum ether to afford intermediate **31** (210 mg, 0.849 mmol, 55%). A solution of this material in methanol (3 mL) was treated with cHCl (1 mL). After 1 h, the solution was concentrated to dryness to give title compound **32** as a solid residue (0.830 mmol, 98%). The solid residue was dissolved in H₂O (10 mL) or D₂O (for NMR samples) and the solvent removed (x3). ¹H NMR (D₂O) 4.40 (q, *J* = 3.1 Hz, 1H), 3.67 (dd, *J* = 12.0, 6.6 Hz, 1H), 3.50 (dd, *J* = 12.8, 5.1 Hz, 1H), 3.28 (dd, *J* = 12.8, 3.0 Hz, 1H), 3.22 (dd, *J* = 8.7, 3.4 Hz, 1H), 2.73 – 2.66 (m, 1H), 2.58 – 2.50 (m, 2H), 2.40 – 2.30 (m, 2H) 2.13 (s, 3H). ¹³C NMR (D₂O) 73.5, 51.5, 48.6, 45.2, 34.3, 14.9.

**Scheme 3**

Preparative Example 1.02

(3R,4S)-4-(Ethylthiomethyl)-3-hydroxy-pyrrolidine (33). Following the general procedure outlined above, mesylate **30** (260 mg, 0.880 mmol) was processed to afford title compound **33** (100 mg, 0.506 mmol, 58%). ¹H NMR (D₂O) 4.30 – 4.24 (m, 1H), 3.53 (dd, *J* = 12.3, 7.2 Hz, 1H), 3.37 (dd, *J* = 12.8, 5.2 Hz, 1H), 3.14 (dd, *J* = 12.8, 3.1 Hz, 1H), 3.07 (dd, *J* = 12.2, 5.7 Hz, 1H), 2.65 – 2.55 (m, 1H), 2.46 (q, *J* = 7.4 Hz, 2H), 2.40 – 2.30 (m, 2H) 1.09 (t, *J* = 7.4 Hz, 3H). ¹³C NMR (D₂O) 73.6, 51.5, 48.6, 45.6, 31.7, 26.0, 14.4. HRMS (MH⁺) calc for C₇H₁₆NOS: 162.0947. Found 162.0952.

Preparative Example 1.03

(3R,4S)-3-Hydroxy-4-(propylthiomethyl)-pyrrolidine (34). Following the general procedure outlined above, mesylate **30** (264 mg, 0.894 mmol) was processed to afford **34** (139 mg, 0.656 mmol, 73%). ¹H NMR (D₂O) 4.41 – 4.37 (m, 1H), 3.67 (dd, *J* = 12.3, 7.2 Hz, 1H), 3.50 (dd, *J* = 12.8, 5.2 Hz, 1H), 3.27 (dd, *J* = 12.8, 3.1 Hz, 1H), 3.21 (dd, *J* = 12.2, 5.6 Hz, 1H), 2.76 – 2.71 (m, 1H), 2.61 – 2.50 (m, 4H), 1.59 (sextet, *J* = 7.3 Hz), 0.95 (t, *J* = 7.3 Hz, 3H). ¹³C NMR (D₂O) 73.6, 51.5, 48.6, 45.7, 34.1, 32.1, 22.7, 13.1. HRMS (MH⁺) calc for C₈H₁₈NOS: 176.1104. Found 176.1106.

Preparative Example 1.04

(3R,4S)-3-Hydroxy-4-(isopropylthiomethyl)-pyrrolidine hydrochloride (35). To a solution of 2-propanethiol (0.36 mL, 3.9 mmol) in DMF (10 mL) at 0 °C was added 60% NaH (145 mg, 3.6 mmol). After 10 min. of stirring a solution of the mesylate **30** (565 mg, 1.91 mmol) in DMF (10 mL) was added. Stirring was continued while the reaction was allowed to attain r.t. After completion, the reaction was quenched with aq. NaHCO₃, toluene was added and the reaction was processed normally to give the crude intermediate which was subjected to column chromatography on silica eluting with petroleum ether: EtOAc (4:1 → 1:1 v/v) to give the clean intermediate (3R,4S)-*N*-*tert*-butoxycarbonyl-3-hydroxy-4-(isopropylthiomethyl)-pyrrolidine as a colourless syrup (490 mg, 97%). NMR (300 MHz, CDCl₃): δ ppm: 4.18-4.11 (m, 1H), 3.72-3.60 (m, 2H), 3.28-3.18 (m, 1H), 3.13 (dd, *J* = 11.1, 6.7 Hz, 1H), 2.95 (sept., *J* = 6.7 Hz, 1H), 2.69-2.50 (m, 2H), 2.33-2.22 (m, 1H), 1.46 (s, 9H), 1.29, 1.27 (s, 3H each). ¹³C NMR (300 MHz,

CDCl₃): δ ppm: (note that some peaks are doubled due to slow conversion of rotamers) 154.93, 79.97, (75.48, 74.64), (52.81, 52.55), (49.69, 49.42), (46.13, 45.35), 35.71, 32.32, 28.87, 23.73, 23.69. A solution of this material in MeOH (10 mL) was treated with 12 N (conc.) HCl (4 mL) at 40 °C for 30 min. to give the crude title compound **35** which was used directly in a Mannich-type reaction.

Preparative Example 1.05

(3R,4S)-4-(Butylthiomethyl)-3-hydroxy-pyrrolidine (36). Following the general procedure outlined above, mesylate **30** (438 mg, 1.48 mmol) was processed to afford **36** (284 mg, 1.25 mmol, 84%). ¹H NMR (D₂O) 4.40 – 4.36 (m, 1H), 3.66 (dd, *J* = 12.3, 7.2 Hz, 1H), 3.48 (dd, *J* = 12.8, 5.2 Hz, 1H), 3.26 (dd, *J* = 12.8, 3.1 Hz, 1H), 3.21 (dd, *J* = 12.2, 5.6 Hz, 1H), 2.76 – 2.70 (m, 1H), 2.62 – 2.50 (m, 4H), 1.61 – 1.51 (m, 2H), 1.40 – 1.33 (m, 2H), 0.86 (t, *J* = 7.3 Hz, 3H). ¹³C NMR (D₂O) 73.5, 51.5, 48.6, 45.7, 32.2, 31.7, 31.3, 21.7, 13.3. HRMS (MH⁺) calc for C₉H₂₀NOS: 190.1260. Found 190.1257.

Preparative Example 1.06

(3R,4S)-3-Hydroxy-4-(phenylthiomethyl)-pyrrolidine (37). Following the general procedure outlined above, mesylate **30** (300 mg, 1.00 mmol) was processed to afford **37** (692 mg, 0.69 mmol). ¹H NMR (D₂O) 7.51 – 7.24 (m, 5H), 4.38 – 4.34 (m, 1H), 3.56 (dd, *J* = 12.2, 7.7 Hz, 1H), 3.45 (dd, *J* = 12.6, 5.2 Hz, 1H), 3.26 – 3.00 (m, 3H), 2.88 (dd, *J* = 13.7, 8.3 Hz, 1H), 2.48 – 2.37 (m, 1H). ¹³C NMR (D₂O) 134.5, 130.5, 129.9, 127.6, 73.4, 51.5, 48.4, 45.5, 34.3.

Preparative Example 1.07

(3R,4S)-(4-Fluorophenylthiomethyl)-3-hydroxy-4-pyrrolidine (38). Following the general procedure outlined above, mesylate **30** (281 mg, 0.951 mmol) was processed to afford **38** (160 mg, 0.607 mmol, 64%). ¹H NMR (D₂O) 7.49 – 7.42 (m, 2H), 7.19 – 7.06 (m, 2H), 4.41 – 4.36 (m, 1H), 3.60 (dd, *J* = 12.3, 7.7 Hz, 1H), 3.48 (dd, *J* = 12.8, 5.2 Hz, 1H), 3.27 – 3.16 (m, 2H), 3.07 (dd, *J* = 13.8, 6.9 Hz, 1H), 2.88 (dd, *J* = 13.8, 8.3 Hz, 1H), 2.45 – 2.38 (m, 1H). ¹³C NMR (D₂O) 164.2, 160.9, 133.7, 133.6, 129.4, 116.9, 116.6, 73.3, 51.5, 48.4, 45.5, 35.5. HRMS (MH⁺) calc for C₁₁H₁₅NOSF: 228.0853. Found 228.0856.

Preparative Example 1.08

(3R,4S)-(4-Chlorophenylthiomethyl)-3-hydroxy-4-pyrrolidine (39). Following the general procedure outlined above, mesylate **30** (245 mg, 0.83 mmol) was processed to afford **39** (212 mg, 0.76 mmol, 91%). ¹H NMR (d₄-MeOH) 7.51 – 7.39 (m, 2H), 7.35 – 7.31 (m, 2H), 4.38 – 4.33 (m, 1H), 3.59 (dd, *J* = 12.0, 7.6 Hz, 1H), 3.47 (dd, *J* = 12.4, 4.9 Hz, 1H), 3.28 – 3.18 (m, 3H), 2.93 (dd, *J* = 13.6, 9.0 Hz, 1H), 2.49 – 2.38 (m, 1H). ¹³C NMR (D₂O) 135.7, 134.1, 132.9, 130.8, 74.8, 52.9, 49.6, 47.5, 35.8.

Preparative Example 1.09

(3R,4S)-4-(3-Chlorophenylthiomethyl)-3-hydroxy-pyrrolidine (40). Following the general procedure outlined above, mesylate **30** (300 mg, 1.02 mmol) was processed to afford **40** (192 mg, 0.685 mmol, 67%). ¹H NMR (D₂O) 7.33 – 7.15 (m, 4H), 4.39 – 4.35 (m, 1H), 3.59 (dd, *J* = 12.2, 7.7 Hz, 1H), 3.47 (dd, *J* = 12.7, 5.1 Hz, 1H), 3.28 – 3.04 (m, 3H), 2.89 (dd, *J* = 13.7, 8.3 Hz, 1H), 2.49 – 2.41 (m, 1H). ¹³C NMR (D₂O) 136.9, 134.7, 131.0, 129.3, 128.1, 127.2, 73.4, 51.5, 48.4, 45.3, 34.0. HRMS (MH⁺) calc for C₁₁H₁₅NOSCl: 244.0557(4). Found 244.0556(8).

Preparative Example 1.10

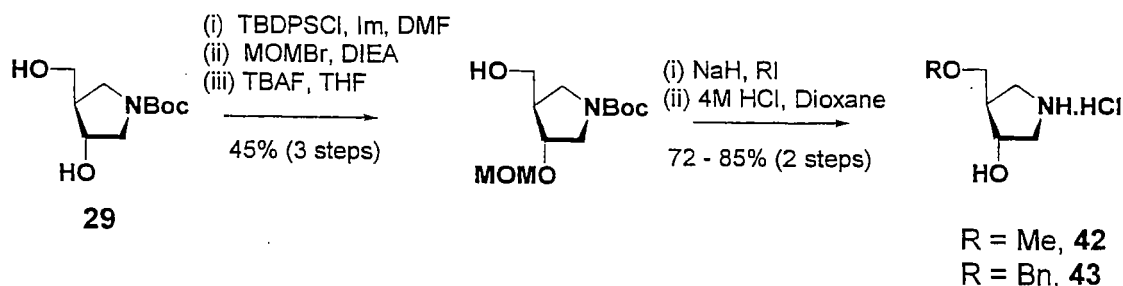
(3R,4S)-4-(Benzylthiomethyl)-3-hydroxy-pyrrolidine (41). Following the general procedure outlined above, mesylate **30** (1.10 g, 3.7 mmol) was processed to afford **41** (730 mg, 2.81 mmol, 76%). ¹H NMR (D₂O) 7.40 – 7.27 (m, 5H), 4.26 – 4.22 (m, 1H), 3.74 (s, 2H), 3.56 (dd, *J* = 12.4, 7.2 Hz, 1H), 3.37 (dd, *J* = 12.8, 5.2 Hz, 1H), 3.21 (dd, *J* = 12.8, 3.0 Hz, 3H), 2.07 (dd, *J* = 12.4, 5.5 Hz, 1H), 2.61 – 2.52 (m, 1H), 2.47 – 2.34 (m, 2H). ¹³C NMR (D₂O) 138.7, 129.5, 129.3, 127.9, 73.5, 51.5, 48.5, 45.4, 35.9, 31.8. HRMS (MH⁺) calc for C₁₂H₁₈NOS: 224.1109. Found 224.1102.

Preparative Example 1.11

(3R,4R)-3-Hydroxy-4-(methoxymethyl)-pyrrolidine (42), Scheme 4. The diol **29** was manipulated according to Scheme 4 to afford **42**. ¹H NMR (D₂O) 4.30 – 4.26 (m, 1H), 3.52 – 3.28 (m, 4H), 3.22 (s, 3H), 3.15 – 3.00 (m, 2H), 2.48 – 2.37

(m, 1H). ^{13}C NMR (D_2O) 72.1, 71.6, 58.8, 52.0, 46.7, 45.7. HRMS (MH^+) calc for $\text{C}_6\text{H}_{14}\text{NO}_2$: 132.1019. Found 132.1012.

5



Scheme 4

Preparative Example 1.12

(3R,4R)-4-(Benzyloxymethyl)-3-hydroxy-pyrrolidine (43), Scheme 4. The diol **29** was manipulated according to Scheme 4 to afford **43**. ^1H NMR (D_2O , free base) 7.32 – 7.15 (m, 5H), 4.36 (s, 2H), 3.92 – 3.85 (m, 1H), 3.35 (dd, $J = 9.8$, 7.0 Hz, 1H), 3.24 (dd, $J = 9.8$, 7.8 Hz, 1H), 2.97 (dd, $J = 11.8$, 7.9 Hz, 1H), 2.75 (dd, $J = 12.4$, 5.5 Hz, 1H), 2.57 (dd, $J = 12.4$, 3.4 Hz, 1H), 2.36 (dd, $J = 11.8$, 5.7 Hz, 1H), 2.15 – 2.05 (m, 1H). ^{13}C NMR (D_2O) 137.6, 129.0, 128.8, 128.6, 74.7, 73.1, 71.0, 53.2, 48.0, 47.6. HRMS (MH^+) calc for $\text{C}_{12}\text{H}_{18}\text{NO}_2$: 208.1332. Found 208.1329.

Preparative Example 1.13

(3R,4S)-4-(Cyclohexylthiomethyl)-3-hydroxy-pyrrolidine hydrochloride (44)

To a solution of cyclohexylmercaptan (0.47 mL, 3.84 mmol) in DMF (10 mL) at 0 °C was added 60% NaH (145 mg, 3.6 mmol). After 10 min. of stirring, a solution of the mesylate **30** (565 mg, 1.91 mmol) in DMF (10 mL) was added. Stirring was continued while the reaction was allowed to attain r.t. After completion, the reaction was quenched with aq. NaHCO_3 , toluene was added and the reaction was processed normally to give the crude intermediate which was subjected to column chromatography on silica eluting with petroleum ether : EtOAc (1:1 v/v) to give the clean intermediate (3R,4S)-*N*-tert-butoxycarbonyl-3-hydroxy-4-(cyclohexylthiomethyl)-pyrrolidine as a colourless syrup (428 mg, 71%). NMR (300 MHz, CDCl_3): δ ppm: 4.17-4.09 (m, 1H), 3.72-3.60 (m, 2H), 3.28-3.18 (m,

1H), 3.15-3.09 (m, 1H), 2.74-2.64 (m, 2H), 2.60-2.53 (m, 1H), 2.32-2.23 (m, 1H), 1.96 (m, 2H), 1.81-1.77 (m, 2H), 1.66-1.61 (m, 1H), 1.45 (s, 9H), 1.35-1.24 (m, 5H). ¹³C NMR (300 MHz, CDCl₃): δ ppm: (note that some peaks are doubled due to slow conversion of rotamers) 154.91, 79.94, (75.57, 74.72), (52.80, 52.55), (49.70, 49.43), (46.24, 45.41), 44.26, 33.99, 33.93, 31.90, 28.87, 26.43, 26.12. A solution of this material in MeOH (10 mL) was treated with 12 N (conc.) HCl (4 mL) at 40 °C for 30 min. to give the crude title compound **44** which was used directly in a Mannich-type reaction.

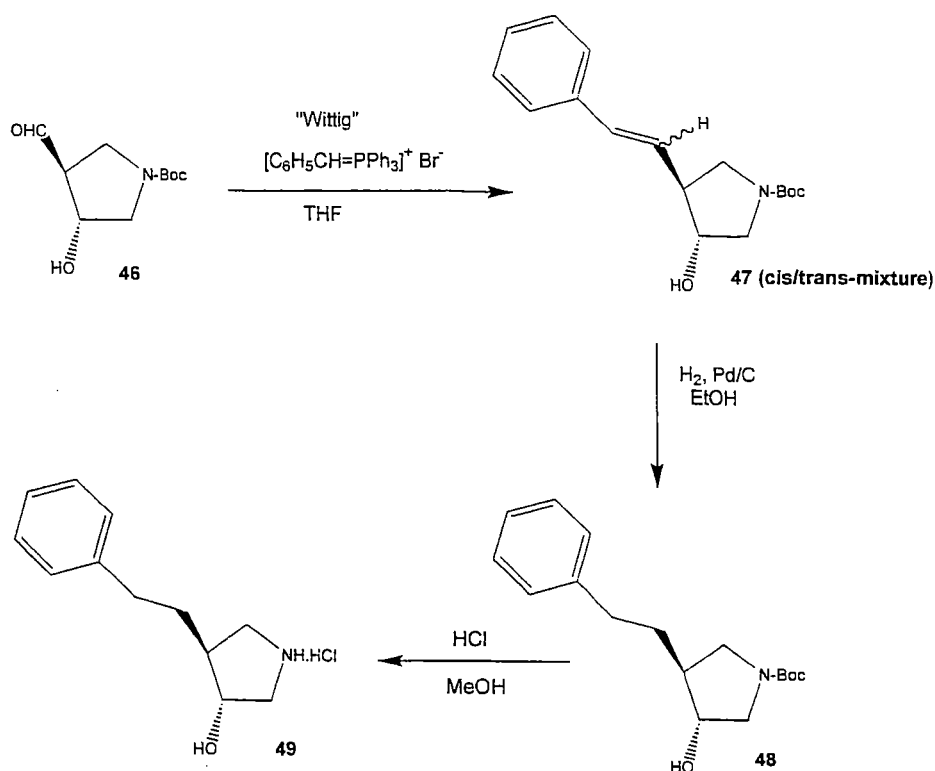
Preparative Example 1.14

(3R,4S)-3-Hydroxy-4-(4-pyridylthiomethyl)-pyrrolidine (45). Following the general procedure outlined above, mesylate **30** (348 mg, 1.18 mmol) was processed to afford **45** (105 mg, 0.426 mmol, 36%). ¹H NMR (D₂O) 8.42 (d, *J* = 7.2 Hz, 2H), 7.82 (d, *J* = 7.2 Hz, 2H), 4.51 – 4.46 (m, 1H), 3.74 (dd, *J* = 12.4, 7.8 Hz, 1H), 3.57 (dd, *J* = 12.8, 5.5 Hz, 1H), 3.44 (dd, *J* = 13.6, 7.3 Hz, 1H) 3.34 – 3.22 (m, 3H), 2.78 – 2.60 (m, 1H). ¹³C NMR (D₂O) 164.1, 139.4, 122.9, 73.4, 51.4, 48.4, 44.3, 31.3. HRMS (MH⁺) calc for C₁₀H₁₅N₂OS: 211.0900. Found 211.0908.

Preparative Example 2.01

(3R,4S)-3-Hydroxy-4-(2-phenylethyl)-pyrrolidine hydrochloride (49), Scheme 5. To a suspension of benzyltriphenylphosphonium bromide (1.75 g, 4.97 mmol) in dry THF (10 mL) under argon at 0 °C was added 1.6 M BuLi in THF (2.33 mL, 3.73 mmol) and the deep red solution left stirring without cooling for 10 min. After re-cooling to 0 °C, the aldehyde **46** (335 mg, 1.56 mmol) [G.B. Evans, R.H. Furneaux, A. Lewandowicz, V.L. Schramm, and P.C. Tyler, Second-Generation Transition State Analogues of Human Purine Nucleoside Phosphorylase, *J. Med. Chem.*, 46 (2003) 5271-5276] in THF (5 mL) was added and the mixture stirred at r.t. for 12 h. The reaction was then quenched with water (1 mL) and extracted with dichloromethane (100 mL). The organic phase was washed with sat. aq. NaHCO₃ (15 mL) then water (15 mL), dried (MgSO₄) and concentrated *in vacuo*. The residue was subjected to chromatography to afford a ca. 1:3 *cis/trans* mixture of (3R,4S)-*N*-*tert*-butoxycarbonyl-3-hydroxy-4-(2-phenylethenyl)-pyrrolidine (**47**) as a syrup (290 mg, 64 %). ¹H NMR (300 MHz, CDCl₃): δ ppm:

trans: 7.28 (m, 5H), 6.49 (d, J = 15.9 Hz, 1H), 6.03 (dd, J = 15.9, 8.1 Hz, 1H), 4.11 (m, 1H), 3.67 (m, 2H), 3.32 (m, 2H), 2.83 (m, 1H), 1.46 (s, 9H); **cis:** 7.27 (m, 5H), 6.58 (d, J = 11.6 Hz, 1H), 5.43 (dd, J = 11.6 Hz, 10.0 Hz, 1H), 4.11 (m, 1H), 3.65 (m, 2H), 3.21 (m, 2H), 2.88 (m, 1H), 1.44 (s, 9H). To a solution of intermediate **47** (290 mg, 1.00 mmol) in ethanol (20 mL) was added 10 % Pd/C (250 mg) and the suspension was stirred under an atmosphere of hydrogen for 12 h. After filtration, the solvent was removed *in vacuo* to give (3R,4S)-*N*-tert-butoxycarbonyl-3-hydroxy-4-(2-phenylethyl)pyrrolidine (**48**) as a syrup, 254 mg (87 %). ¹H NMR (300 MHz, CDCl₃): δ ppm: 7.10 (m, 5H), 4.00 (m, 1H), 3.47 (m, 2H), 3.07 (m, 2H), 2.67 (m, 2H), 2.04 (m, 1H), 1.83 (m, 1H), 1.54 (m, 1H), 1.45 (s, 9H). ¹³C NMR (300 MHz, CDCl₃): δ ppm (note that some peaks are doubled due to slow conversion of rotamers): 155.17, 142.03, 128.83, 128.71, 126.37, 79.88, (74.94, 71.26), (53.17, 52.90), (49.90, 49.34), (46.11, 45.52), 34.41, 33.69, 28.91. To a solution of intermediate **48** (254 mg, 0.87 mmol) in methanol (10 mL) was added 12N (conc.) HCl (4 mL) and the solution stirred at 40 °C for 30 min. After removal of the solvent *in vacuo* and azeotroping with toluene, the crude title compound (3R,4S)-3-Hydroxy-4-(2-phenylethyl)pyrrolidine hydrochloride **49** was obtained as a solid (202 mg, 0.89 mmol, 102 %). ¹H NMR (300 MHz, MeOH-d₄): δ ppm: 7.14 (m, 5H), 4.22 (m, 1H), 3.52 (dd, J = 11.8, 7.4 Hz, 1H), 3.39 (dd, J = 12.3, 4.9 Hz, 1H), 3.14 (dd, J = 12.3, 2.8 Hz, 1H), 3.02 (dd, J = 11.8 Hz, 1H), 2.71 (m, 2H), 2.20 (m, 1H), 1.84 (m, 1H), 1.62 (m, 1H). ¹³C NMR (300 MHz, MeOH-d₄): δ ppm: 142.94, 129.93, 129.89, 127.56, 75.56, 52.90, 48.55, 47.28, 35.18, 34.44.



Scheme 5

Preparative Example 2.02**(3R,4S)-3-Hydroxy-4-propylpyrrolidine hydrochloride (52)**

5 The synthesis of this compound follows the same general route outlined in scheme 5 [see Preparative Example 2.01]. To a suspension of ethyltriphenylphosphonium bromide (2.9 g, 6.93 mmol) in dry THF (15 mL) under argon at 0 °C was added 1.6 M BuLi in THF (4 mL, 6.40 mmol) and the deep red solution left stirring without cooling for 10 min. After re-cooling to 0 °C, the aldehyde **46** (580 mg, 2.69 mmol) in THF (10 mL) was added and the mixture stirred at r.t. for 12 h. The reaction was then quenched with water (1 mL) and extracted with dichloromethane (100 mL). The organic phase was washed with sat. aq. NaHCO_3 (15 mL) then water (15 mL), dried over MgSO_4 and concentrated *in vacuo*. The residue was subjected to chromatography to

10 afforded (3R,4S)-*N*-tert-butoxycarbonyl-3-hydroxy-4-propenylpyrrolidine (**50**) as a light yellow syrup (165 mg, 27 %). To a solution of intermediate **50** (165 mg, 0.73 mmol) in ethanol (10 mL) was added 10 % Pd/C (60 mg) and the

15

suspension was stirred under an atmosphere of hydrogen for 3 h. After filtration, the solvent was removed *in vacuo* to give (3R,4S)-*N*-*tert*-butoxycarbonyl-3-hydroxy-4-propyl-pyrrolidine (**51**) as a syrup (172 mg, 102%). ¹H NMR (300 MHz, CDCl₃): δ ppm: 3.98-3.96 (m, 1H), 3.61-3.57 (m, 3H), 3.22-3.18 (m, 1H), 3.10-3.01 (m, 1H), 2.03 (m, 1H), 1.45 (s, 9H), 1.41-1.31 (m, 2H), 1.24-1.12 (m, 1H), 0.94-0.89 (m, 3H). ¹³C NMR (300 MHz, CDCl₃): δ ppm (note that some peaks are doubled due to slow conversion of rotamers): 155.20, 79.69, (75.53, 74.76), (53.10, 52.80), (49.94, 49.38), (46.19, 45.67), 34.04, 28.83, 21.27, 14.47. To a solution of intermediate **51** (172 mg, 0.75 mmol) in methanol (10 mL) was added 12N (conc.) HCl (4 mL) and the solution stirred at 40 °C for 30 min. After removal of the solvent *in vacuo* and azeotroping with toluene, the crude title compound (3R,4S)-3-hydroxy-4-propyl-pyrrolidine hydrochloride (**52**) was obtained as a syrup (138 mg, 0.83 mmol, 111 %). ¹H NMR (300 MHz, MeOH-d₄): δ ppm: 4.20-4.16 (m, 1H), 3.59-3.52 (m, 1H), 3.44-3.39 (m, 1H), 3.19-3.14 (m, 1H), 3.05-2.99 (m, 1H), 2.23-2.17 (m, 1H), 1.55-1.28 (m, 4H), 0.98-0.94 (m, 3H). ¹³C NMR (300 MHz, MeOH-d₄): δ ppm: 75.65, 52.82, 50.30, 47.45, 34.69, 22.30, 14.78.

Preparative Example 3.01

9-Deaza-adenine. 6-Chloro-9-deazapurine (3 g, 19.5 mmol) was added to a saturated solution of ammonia in ethanol (30 mL). The resulting suspension was heated at 130 °C for 16 h in a sealed tube. The homogeneous solution was cooled, flash silica gel was added, and the suspension concentrated *in vacuo*. The resultant solid was loaded onto the top of a column of silica gel and eluted with methanol/CH₂Cl₂ (4:1 v/v) to afford 9-deaza-adenine as a pale yellow solid (2.16 g, 80%). ¹³C NMR (d₄-MeOH) δ ppm: 153.1, 149.9, 145.2, 131.3, 113.8, 101.6.

Preparative Example 4.01

(3R,4R)-3-Hydroxy-4-methanesulfonyloxy-pyrrolidine (53**).** A solution of HCl in 1,4-dioxane (4M, 2 mL) was added to a stirred solution of the mesylate **30** (169 mg, 0.572 mmol) in methanol (3 mL). After stirring at RT for 12 h, the solvents were removed *in vacuo* to give a residue to which methanol (x2) and then D₂O

was added and evaporated to give title compound **53** (120 mg, 0.518 mmol, 91%). ¹H NMR (D₂O) 4.55 – 4.35 (m, 3H), 3.74 (dd, *J* = 12.5, 8.4 Hz, 1H), 3.50 (dd, *J* = 12.7, 5.3 Hz, 1H), 3.35 – 3.20 (m, 2H), 3.25 (s, 3H), 2.82 – 2.67 (m, 1H). ¹³C NMR (D₂O) 71.3, 69.2, 52.0, 46.0, 45.3, 36.9. HRMS (MH⁺) calc for C₈H₁₄NO₄S: 196.0638. Found 196.0648.

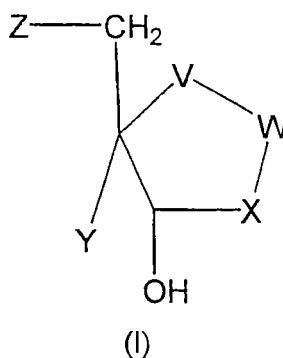
Although the invention has been described by way of example, it should be appreciated that variations or modifications may be made without departing from the scope of the invention. Furthermore, when known equivalents exist to specific features, such equivalents are incorporated as if specifically referred to in the specification.

INDUSTRIAL APPLICABILITY

The present invention provides a useful synthetic route to compounds that are inhibitors of PNP, PPRT, MTAN, MTAP and/or NH. The compounds may be useful in the treatment of diseases in which the inhibition of PNP, PPRT, MTAN, MTAP and/or NH is desirable. Such diseases include cancer, bacterial infection, protozoal infection or T-cell mediated diseases.

CLAIMS

1. A process for preparing a compound of the formula (I)



wherein:

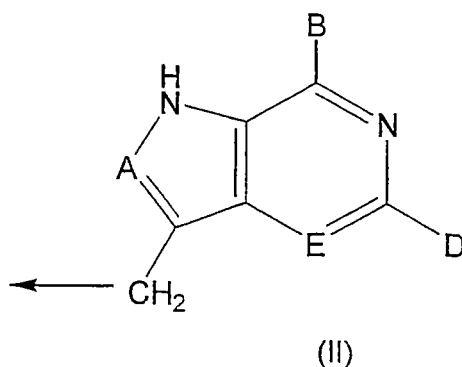
10 V is selected from CH₂ and NH, and W is NR¹; or
V is NR¹, and W is selected from CH₂ and NH;

15 X is selected from CH₂ and CHOH in the R or S-configuration, except
where W is selected from NH and NR¹, then X is CH₂;

Y is selected from hydrogen, halogen and hydroxy, except where V is
selected from NH and NR¹, then Y is hydrogen;

20 Z is selected from hydrogen, halogen, hydroxy, a sulfonate leaving
group, SQ, OQ and Q, where Q is an optionally substituted alkyl,
aralkyl or aryl group; and

R¹ is a radical of the formula (II)



wherein:

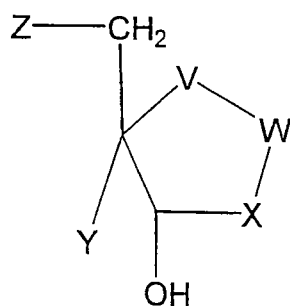
A is selected from N, CH and CR^2 , where R^2 is selected from halogen, optionally substituted alkyl, aralkyl or aryl, OH, NH_2 , NHR^3 , NR^3R^4 and SR^5 , where R^3 , R^4 and R^5 are each optionally substituted alkyl, aralkyl or aryl groups;

B is selected from OH, NH_2 , NHR^6 , SH, hydrogen and halogen, where R^6 is an optionally substituted alkyl, aralkyl or aryl group;

D is selected from OH, NH_2 , NHR^7 , hydrogen, halogen and SCH_3 , where R^7 is an optionally substituted alkyl, aralkyl or aryl group; and

E is selected from N and CH;

including reacting a compound of the formula (III)



wherein:

V is selected from CH₂ and NH, and W is NH; or

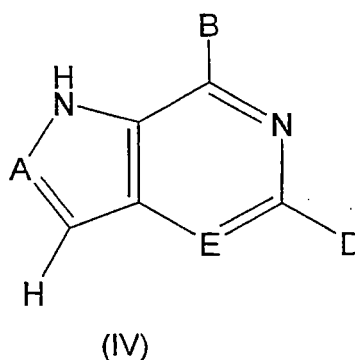
V is NH, and W is selected from CH₂ and NH;

X is selected from CH₂ and CHOH in the R or S-configuration, except where W is NH, then X is CH₂;

Y is selected from hydrogen, halogen and hydroxy, except where V is selected from NH, then Y is hydrogen; and

Z is selected from hydrogen, halogen, hydroxy, a sulfonate leaving group, SQ, OQ and Q, where Q is an optionally substituted alkyl, aralkyl or aryl group;

with a compound of the formula (IV)



wherein A, B, D, and E are as defined above;

and with formaldehyde or a formaldehyde equivalent.

2. A process as claimed in claim 1 wherein Z is hydrogen, halogen, hydroxy, SQ or OQ, where Q is an optionally substituted alkyl, aralkyl or aryl group.

3. A process as claimed in claim 1 or claim 2 wherein A is CH.
4. A process as claimed in any one of claims 1 to 3 wherein Y is H.
- 5 5. A process as claimed in any one of claims 1 to 4 wherein W is NR^1 , V is CH_2 and X is CH_2 .
6. A process as claimed in claim 5 wherein R^1 is a radical of formula (II) as defined in claim 1, where A is CH and E is N.
- 10 7. A process as claimed in any one of claims 1 to 6 wherein D is H or NH_2 .
8. A process as claimed in any one of claims 1 to 7 wherein B is NH_2 , OH or Cl.
- 15 9. A process as claimed in any one of claims 1 or 3 to 8, wherein Z is methanesulfonate, *p*-toluenesulfonate or trifluoromethanesulfonate.
- 20 10. A process as claimed in claim 9 wherein Z is methanesulfonate.
11. A process as claimed in any one of claims 1 to 10 wherein the compounds of formula (III) and (IV) are reacted with formaldehyde.
- 25 12. A process as claimed in any one of claims 1 to 10 wherein the compounds of formula (III) and (IV) are reacted with a formaldehyde equivalent which is paraformaldehyde.
- 30 13. A process as claimed in claim 1 wherein the compound of formula (I) is:
(3R,4R)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-hydroxymethyl-pyrrolidine;
(3R,4R)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(hydroxymethyl)-pyrrolidine;

- (3R,4S)-4-(benzylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine;
- (3R,4S)-4-(4-chlorophenylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine;
- 5 (3R,4R)-1-[(6-chloro-9-deazapurin-9-yl)methyl]-3-hydroxy-4-(hydroxymethyl)-pyrrolidine
- (3R,4R)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(methanesulfonyl)-pyrrolidine;
- (3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-
- 10 (methylthiomethyl)-pyrrolidine;
- (3R,4S)-4-(ethylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine;
- (3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(propylthiomethyl)-pyrrolidine;
- 15 (3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(isopropylthiomethyl)-pyrrolidine;
- (3R,4S)-4-(butylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine;
- (3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-
- 20 (phenylthiomethyl)-pyrrolidine;
- (3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-4-(4-fluorophenylthiomethyl)-3-hydroxy-pyrrolidine;
- (3R,4S)-4-(3-chlorophenylthiomethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine;
- 25 (3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(cyclohexylthiomethyl)pyrrolidine;
- (3R,4S)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(4-pyridylthiomethyl)-pyrrolidine;
- (3R,4R)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-4-(methoxymethyl)-
- 30 pyrrolidine;
- (3R,4R)-4-(benzyloxymethyl)-1-[(9-deaza-adenin-9-yl)methyl]-3-hydroxy-pyrrolidine;
- (3R,4R)-1-[(9-deazaguanin-9-yl)methyl]-3-hydroxy-4-hydroxymethyl-pyrrolidine;

(3R,4S)-1-[(9-deazahypoxanthin-9-yl)-3-hydroxy-4-(propylthiomethyl)-pyrrolidine;

(3R,4S)-4-(butylthiomethyl)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxy-pyrrolidine;

(3R,4S)-1-[(9-deaza-6-chloro-purin-9-yl)methyl]-3-hydroxy-4-(2-phenylethyl)pyrrolidine;

(3R,4S)-1-[(9-deazaadenin-9-yl)methyl]-3-hydroxy-4-propyl-pyrrolidine;

(3R,4S)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-propyl-pyrrolidine; or

(3R,4S)-1-[(9-deazahypoxanthin-9-yl)methyl]-3-hydroxy-4-(methylthiomethyl)-pyrrolidine.

14. A process for preparing a compound of formula (I) as defined in claim 1 including:

- (i) reacting a compound of formula (III) as defined in claim 1 with a compound of formula (IV) as defined in claim 1 and with formaldehyde or a formaldehyde equivalent, where any one or more of V, W, X, Y and Z of the compound of formula (III) is protected with a suitable protecting group; and
- (ii) removing the one or more protecting groups to give the compound of formula (I).

15. A process for preparing a compound of formula (I) as defined in claim 1 including:

- (i) reacting a compound of formula (III) as defined in claim 1 with a compound of formula (IV) as defined in claim 1 and with formaldehyde or a formaldehyde equivalent, where any one or more of A, B, D and E of the compound of formula (IV) is protected with a suitable protecting group; and
- (ii) removing the one or more protecting groups to give the compound of formula (I).

16. A process of preparing a compound of formula (I) as defined in claim 1 including:

- 5 (i) reacting a compound of formula (III) as defined in claim 1 with a compound of formula (IV) as defined in claim 1 and with formaldehyde or a formaldehyde equivalent, where any one or more of V, W, X, Y and Z of the compound of formula (III) is protected with a suitable protecting group and where any one or more of A, B, D and E of the compound of formula (IV) is protected
- 10 with a suitable protecting group; and
- (ii) removing the one or more protecting groups to give the compound of formula (I).