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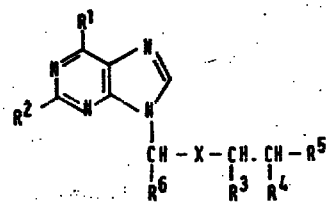
(54) PURINE COMPOUNDS AND SALTS THEREOF

(71) We, THE WELLCOME FOUNDATION LIMITED, of 183—193 Euston Road, London, N.W.1., a company incorporated in England do hereby declare the invention which was communicated from Burroughs Wellcome Co. of 3030 Cornwallis Road, Research Triangle Park, North Carolina 27709, being incorporated in The State of North Carolina, United States of America for which we pray that a Patent may be granted to us and the method by which it is to be performed, to be particularly described in and by the following statement:—

This invention relates to substituted purine compounds and their pharmaceutically acceptable salts and to methods of preparing them. In particular this invention relates to the 9 - (2 - hydroxyethoxymethyl) derivatives of purines such as adenine, guanine, thioguanine and 2,6 - diaminopurine and the pharmaceutically acceptable salts of these compounds. In 1971, Schaeffer, *et al.* {*J. Med. Chem., 14,* 367 (1971)} reported the syntheses of several purine acyclic nucleosides in a study of adenosine deaminase enzyme-substrate interaction. 9 - (2 - Hydroxyethoxymethyl)adenine in particular was reported and its substrate activity with adenosine deaminase measured.

It has now been discovered that substituted purines of formula (I)

(I)



wherein X is oxygen or sulphur and R^1, R^2, R^3, R^4, R^5 and R^6 are various substituents as hereinafter defined, have antiviral activity against various classes of DNA viruses both in *in vitro* and *in vivo* experiments. In particular the compounds are active against cytomegalovirus, adenovirus, in particular adenovirus 5, rhino virus, Mengo virus and Sindbis virus. They are especially active against vaccinia, and herpes viruses, including simplex, zoster and varicella, in mammals, which cause such diseases as for example herpetic keratitis in rabbits and herpetic encephalitis in mice. In addition they may be useful in the treatment of infectious mononucleosis.

According to the present invention there is provided a compound of formula (I) wherein X is sulphur or oxygen; R^1 is hydrogen, halogen, hydroxy, alkoxy, azide, mercapto, alkylthio, amino, alkylamino or dialkylamino; R^2 is hydrogen, halogen, amino or azide; R^3 is hydrogen, straight or branched chain or cyclic alkyl, hydroxyalkyl, benzyloxyalkyl or phenyl; R^4 is hydrogen, hydroxy, or alkyl; R^5 is hydrogen, hydroxy, amino, alkyl, hydroxyalkyl, benzoyloxy, benzoyloxymethyl, benzyloxy, sulphamoyloxy, phosphate, carboxypropionyloxy, acetoxy, or substituted carbamoyl group of formula NH . CO—Z wherein Z is alkyl, aryl or aralkyl optionally substituted by one or more of sulphonyl, amino, carbamoyl, halogen; R^6 is hydrogen, alkyl, provided that when X is oxygen, R^2, R^3, R^4 and R^6 are hydrogen, and R^5 is hydroxy, hydrogen or benzyloxy, R^1 is not amino or methylamino; when R^2 is hydrogen R^1 is not chlorine; or a salt thereof, especially a pharmaceutically acceptable salt thereof.

Compounds of formula (I) as defined above wherein X is oxygen; R^1 is hydrogen, halogen, hydroxy, alkoxy, mercapto, alkylthio, amino, alkylamino, dialkylamino or azide; R^2 is hydrogen, halogen, amino or azide; R^3 is hydrogen, straight or branched chain or cyclic alkyl, hydroxyalkyl or phenyl; R^4 is hydrogen, or hydroxy; R^5 is hydrogen, hydroxy, benzoyloxy, hydroxyalkyl, amino, carboxypropionyloxy, acetoxy, benzyloxy, benzoyloxymethyl phosphate, sulphamoyloxy, substituted carbamoyl group of formula NH . CO—Z where Z is alkyl, aryl or aralkyl optionally

SPECIFICATION AMENDED - SEE ATTACHED SLIP

substituted by one or more of sulphonyl, amino, carbamoyl, halogen; R^6 is hydrogen, alkyl, provided that R^5 is hydroxy only when R^1 is amino, hydroxy, alkylamino, alkylthio or dialkylamino, and R^2 is amino and R^6 is hydrogen; R^5 is hydroxyalkyl only when R^1 is hydroxy; R^5 is hydrogen only when R^1 is hydroxy or halo; when R^5 is benzoyloxy R^2 is not halogen; R^5 is acetoxy only when R^1 is hydroxy or amino and R^2 is amino or both R^1 and R^2 are amino or both halogen; R^5 is a substituted carbamoyl of formula NH . CO—Z wherein Z is a group

$$CH(NH_2)CH_2C_6H_5$$

only when R^1 is dialkylamino; except that when R^5 is hydroxy and R^1 is alkylamino then R^2 is not hydrogen; R^1 is chlorine only when R^2 is not hydrogen; R^1 is not amino or alkylamino when R^5 is benzyloxy and R^2, R^3, and R^6 are hydrogen; or a salt thereof, especially a pharmaceutically acceptable salt thereof are preferred.

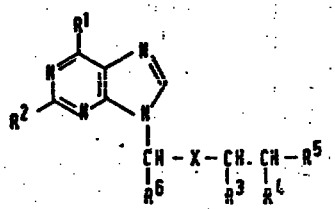
In particular compounds of formula (I), as hereinbefore defined, where X is oxygen, R^1 is halogen, amino, hydroxy or alkylthio; R^2 is amino; R^5 is hydroxy, benzoyloxy, carboxypropionyloxy, acetoxy or hydroxyalkyl and R^3, R^4 and R^6 are hydrogen, provided that R^5 is hydroxyalkyl only when R^1 is hydroxy and R^6 is acetoxy only when R^1 is amino, are more preferred and have been found to be highly active. Compounds where X is oxygen, R^1 is amino, mercapto or hydroxy, R^2 is amino, R^3, R^4 and R^6 are each hydrogen and R^5 is hydroxy are most preferred. Compounds where X is sulphur, R^1 is halogen, amino or alkylamino and R^2, R^3, R^4, R^5 and R^6 are hydrogen have also been found to be highly active.

The preferred halogen substituent is chlorine. As used herein and throughout the specification the term alkyl is denoted to mean 1 to 12 carbon atoms, preferably 1 to 8 carbon atoms.

Salts which are especially convenient for therapeutic use are acid addition salts of pharmaceutically acceptable organic acids such as lactic, acetic, malic, or p - toluenesulfonic acid as well as salts of pharmaceutically acceptable mineral acids such as hydrochloric or sulfuric acid.

In a second aspect of the present invention there is provided a method of preparing a substituted purine of formula (I)

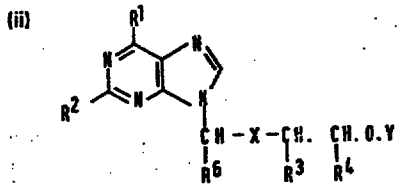
(i)



Wherein X, R^1, R^2, R^3, R^4, R^5, and R^6 are as hereinbefore defined, or a salt thereof, especially a pharmaceutically acceptable salt thereof, characterised in that

(a) a protecting group Y is cleaved from a compound of formula (II)

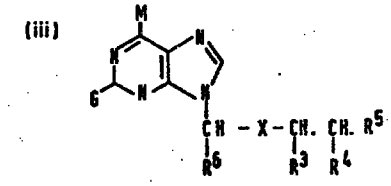
(ii)



by hydrolysis or hydrogenolysis to give a compound of formula (I) wherein R^5 is hydroxy; or

(b) a compound of formula (III)

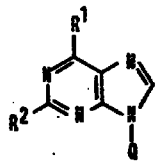
(iii)



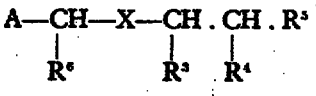
wherein either or both of M and G represent a halogen atom, amino, hydroxy, azido, aralkoxy or mercapto group is converted into a compound of formula (I) by methods known per se; or

(c) a compound of formula (IV)

(iv)



is reacted with a compound of formula (V)



(V)

wherein A is a leaving group or Q is hydrogen or a leaving group; or

(d) a blocking group is linked to R^1 and/or R^2 in a compound of formula (I), the blocking group or groups being removed by solvolysis or alcoholysis; and where the product of said reaction is a base, optionally converting a compound of formula (I) into an acid addition salt thereof, or where the product is a salt of a compound of formula (I), optionally converting said salt into its base or another salt thereof.

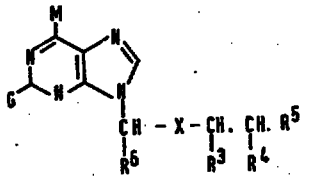
In the case of method a) the 9-ether chain is blocked terminally by a protecting group Y which may be an ester, e.g. an acyl group,

that is a carboxylic acid radical RCO, R being aliphatic, or an arylmethyl group e.g. benzyl. In the first instance the acyl group may aliphatic e.g. acetyl, or aromatic e.g. benzoyl, both types of acyl group being removed by mild basic hydrolysis. In general warming with aqueous methylamine will suffice to bring about the deblocking.

Arylmethyl blocking groups, such as benzyl, are removed by hydrogenolysis, either catalytically, as by hydrogen and Raney nickel or palladium-on-charcoal, or chemically, as by sodium in liquid ammonia. When sodium in liquid ammonia is used, an excess of ammonia serves as solvent. For catalytic hydrogenolysis an alkanol is the preferred solvent, although a number of inert, e.g. non-halogen and non-sulphide or mercapto containing solvents may be used provided they dissolve the acyl-blocked substrate, e.g. such solvents as benzene, tetrahydrofuran or dioxane.

Conversion of a compound of formula (III)

(iii)



into a compound of formula (I), by method (b), can be achieved in numerous different ways. For example one or both of M and G can be converted into a halogen by halogenation; into hydroxy by hydrolysis; into amino or alkylamino by aminolysis; into mercapto or alkylthio by sulphuration; into hydrogen by reduction or desulphuration; or by formation of the azide. All such methods are well known and can be found in "Hetrocyclic compounds—Fused Pyrimidines Part II Purines Ed. by D. J. Brown (1971) published Wiley-Interscience".

Alternatively conversion can be brought about by using enzymes, for example adenosine deaminase efficiently converts a 2,6 - diamino compound to guanine R^1=OH, R^2=NH_2, in an aqueous suspension at about 37°C and initial pH of about 7.0.

Those compounds of formula (III) that can be converted into compounds of formula (I) can be considered as intermediates in the synthesis of those compounds.

In method c) the leaving group A is a reactive residue of an organic or inorganic acid, and may therefore be halogen, or carboxylate, and Q is hydrogen or acyl (as hereinbefore defined). The preferred method comprises the condensation of a purine having the desired 2- and 6-substitution with an acyl or aralkyl - blocked 2 - haloalkoxyethanol for instance 2 - benzoyloxyethoxymethyl chloride, in a strong polar solvent such as DMF or hexamethylphosphoramide, and in the presence of a base such as triethylamine or potassium carbonate. The reaction is preferably carried out at room temperature over an extended period of time i.e. several days may be required to give reasonable yields.

Alternatively a thermal condensation, i.e. fusion reaction, may be carried out to give the product directly. For this reaction a suitably substituted purine is heated together with an acyloxy - alkoxymethyl carboxylate e.g. 2 - oxa - 1 - 4 - butanediol diacetate, in the presence of a catalytic amount of a strong acid such as sulphuric acid. Temperatures in excess of 100°C are generally required, but they should preferably not be greater than about 200°C in order to minimise decomposition. The temperature should be selected such that the mixture of reactants fuse, i.e. melt, before they undergo decomposition.

The fusion reaction may also be carried out under substantially the same conditions as above, with perhaps somewhat lower temperatures, between a 9 - acylpurine and the alkoxymethyl carboxylate or halide. Alternatively the fusion reaction can be carried out using the diester, for instance 2 - acetoxyethoxymethyl acetate.

In method d) substituents R^1 and R^2 may be blocked by for instance trimethylsilyl groups. Such a compound will be the product of the condensation of a trimethylsilylated purine and an ester or diester as in method c). Such blocking groups are very labile and can be removed by solvolysis with alcoholic or aqueous ammonia, or by alcoholysis.

Alternatively the mercuric chloride salt of a purine can be prepared in the presence of alkali and then condensed with a haloether in solvent of the aromatic organic type. Prior to preparation of the salt however all reactive substituents on the purine must be blocked, and therefore the last step in this method is the unblocking of the blocked substituents.

In another method, which combines methods a) and b) solvolytic deblocking may be effected at the same time as replacement of a purine ring leaving group, so that when one of R^1 and R^2 is hydrogen or one or both of R^1 and R^2 is halogen, mercapto, azido, aralkoxy, said substituent is converted into amino, hydroxy, alkylamino or dialkylamino. One example is when a leaving group, such as halogen, on the purine nucleus is replaced by amino at the same time as deblocking the side chain, by reaction with liquid ammonia.

Acid Addition salts may be prepared by simply mixing a compound of formula (I), for example 9 - hydroxyethoxymethyl adenine or 6 - methylamino - 9 - (2 - hydroxyethyoxymethyl) purine, with a pharmaceutically acceptable acid.

In another aspect of the invention there is provided a pharmaceutical composition or preparation comprising a compound of formula (I), wherein X is sulphur or oxygen; R^1 is

halogen, hydroxy, mercapto, alkoxy, azide, alkylthio, amino, alkylamino or dialkylamino; R^2 is hydrogen, halogen, amino or azide; R^3 is hydrogen, straight or branched chain or cyclic alkyl, hydroxyalkyl, benzyloxyalkyl or phenyl; R^4 is hydrogen, hydroxy, or alkyl; R^5 is hydrogen, hydroxy, amino, alkyl, hydroxyalkyl, benzoyloxy, benzoyloxymethyl, benzyloxy, sulphamoyloxy, phosphate, carboxypropionyloxy, acetoxy, or substituted carbamoyl group of formula NH . CO—Z where Z is alkyl, aryl or aralkyl optionally substituted by one or more of sulphonyl, amino, carbamoyl, or halogen; R^6 is hydrogen, alkyl, hydroxyalkyl; wherein when R^6 is alkyl group it has from 1 to 8 carbon atoms and in all other cases when the substituents have an alkyl moiety it has from 1 to 4 carbon atoms; or a pharmaceutically acceptable salt thereof; together with a pharmaceutically acceptable carrier therefor. In a particular aspect the pharmaceutical composition comprises a compound of formula (I) in effective unit dosage form.

As used herein the term "effective unit dosage" or "effective unit dose" is denoted to mean a predetermined antiviral amount sufficient to be effective against the viral organisms *in vivo*. Pharmaceutically acceptable carriers are materials useful for the purpose of administering the medicament, and may be solid, liquid or gaseous materials, which are otherwise inert and medically acceptable and are compatible with the active ingredients.

These pharmaceutical compositions may be given parenterally, orally, used as a suppository or pessary, applied topically as an ointment, cream, aerosol, powder, or given as eye or nose drops etc., depending on whether the preparation is used to treat internal or external viral infections.

For internal infections the compositions are administered orally or parenterally at dose levels, calculated as the free base, of about 0.1 to 250 mg per kg, preferably 1.0 to 50 mg per kg, of mammal body weight, and are used in man in a unit dosage form, administered a few times daily in the amount of 1 to 250 mg per unit dose.

For oral administration, fine powders or granules may contain diluting, dispersing and/or surface active agents, and may be presented in a draught, in water or in a syrup; in capsules or sachets in the dry state or in a non-aqueous solution or suspension, wherein suspending agents may be included; in tablets, wherein binders and lubricants may be included; or in a suspension in water or a syrup. Where desirable or necessary, flavouring, preserving, suspending, thickening or emulsifying agents may be included. Tablets and granules are preferred, and these may be coated.

For parenteral administration or for administration as eye or nose drops, as for eye infections, the compounds may be presented in aqueous solution in a concentration of from about 0.1 to 10%, more preferably 0.1 to 7%, most preferably 0.2% w/v. The solution may contain antioxidants, buffers, etc.

Alternatively for infections of the eye, or other external tissues e.g. mouth and skin the compositions are preferably applied to the infected part of the body of the patient as a topical ointment or cream. The compounds may be presented in an ointment, for instance with a water soluble ointment base, or in a cream, for instance with an oil in water cream base, in a concentration of from about 0.1 to 10%, preferably 0.1 to 7%, most preferably 1% w/v.

In a further aspect of the invention there is provided a method of preparing a pharmaceutical composition comprising bringing into association a substituted purine of formula (I) as hereinbefore defined, or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier therefor.

Of the compounds of formula (I) 9 - (2 - hydroxyethoxymethyl)guanine (R^1=OH, R^2=NH_2) and 2 - amino - 9 - (2 - hydroxyethoxymethyl)adenine are most preferred, particularly because of their extremely high antiviral activity against herpes viruses. Additionally 2 - amino - 6 - chloro - 9 - {(2 - benzoyloxyethoxy)methyl}purine, 9 - (2 - benzoyloxyethoxymethyl)guanine, 9 - (3 - hydroxypropoxymethyl)guanine, 2 - amino - 6 - methylthio - 9 - (2 - hydroxyethoxymethyl)purine, 9 - {2 - (3 - carboxypropionyloxy)ethoxymethyl}guanine, 9 - (2 - acetoxyethoxymethyl) - 2,6 - diamino purine, 6 - chloro - 9 - ethylthiomethyl purine, 9 - ethylthiomethyladenine, 9 - ethylthiomethyl - 6 - methylamino - purine also show high activity against herpes viruses and vaccinia.

In yet a further aspect of the invention there is provided a method of treating viral infections in mammals which comprises the administration of an effective antiviral amount, as hereinbefore defined, of a substituted purine of formula (I), or a pharmaceutically acceptable salt thereof. Administration is preferably by topical application or by the oral or parenteral route.

The invention will now be illustrated with reference to the following Examples.

Example 1

6 - Chloro - 9 - (2 - benzoyloxyethoxymethyl)-purine

A solution of benzonitrile (103 g) in ethylene glycol (310 g) was heated at reflux under substantially anhydrous conditions for 3 days. The reaction mixture was cooled and added to a mixture of ice and water (about 300 ml). The resulting mixture was extracted with ether (3×300 ml) and the combined ether extract back-washed with water

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(2×300 ml) and then with a saturated sodium chloride solution (300 ml). The ether solution was dried over anhydrous sodium sulfate. The ether was evaporated and the residual oil distilled to give 108 g (65% of theoretical) of ethylene glycol monobenzoate, b.p. 132—135°C/1.5 mm Hg.

A cold (0°C) mixture of ethylene glycol monobenzoate (166 g) and paraformaldehyde (30 g) in dry dichloroethane was saturated with dry HCl with stirring for 3 hours. The pinkish red liquid was dried over calcium chloride and the volatile components removed on a rotary evaporator at 30°C to give 1 - benzoyloxy - 2 - chloromethoxyethane (215 g). The Infra-Red spectrum indicated the absence of a hydroxyl group.

Following substantially the procedure of Schaeffer, *et al. supra* but with minor modifications, 1 - benzoyloxy - 2 - chloromethoxyethane (4.4 g) was added with stirring to a dry solution of 6 - chloropurine (3.1 g) and triethylamine (6.5 ml) in dimethylformamide (50 ml). The reaction was exothermic, immediately precipitating triethylamine hydrochloride. The reaction mixture was stirred at room temperature for 24 hours and filtered. The filtrate was evaporated on a rotary evaporator at 70°C. The remaining thick amber oil was dissolved in the minimum amount of benzene and purified by column chromatography using a silica gel column.

Benzene elution removed a trace of an unidentified material. Elution with ether first removed a small amount of ethylene glycol monobenzoate and then 6 - chloro - 9 - (2 - benzoyloxyethoxymethyl)purine. Recrystallization from ether gave a white material, m.p. 108.5—111°C.

Example 2

9 - (2 - Hydroxyethoxymethyl)adenine (I; R^1=NH_2, R^2=H)

A solution of 6 - chloro - 9 - (2 - benzoyloxyethoxymethyl)purine (50 g) and ammonia (31 g) in methanol (120 ml) was heated in a bomb at 95°C for 18 hours. The reaction mixture was removed from the bomb and the solvent evaporated under reduced pressure at 50°C. The resulting solid was triturated first with water and then, after drying, with chloroform. The solid, 9 - (2 - hydroxyethoxymethyl)adenine, was retained, and the aqueous triturate was extracted several times with chloroform. The aqueous extracts were treated with a strongly basic ion exchange resin and then evaporated to dryness under reduced pressure. The remaining solid was combined with that from the trituration step and recrystallized from isopropanol to give 22 g (70% of theoretical) of white 9 - (2 - hydroxyethoxymethyl)adenine, m.p. 199.5—200°C.

Example 3

2 - Chloro - 9 - (2 - hydroxyethoxymethyl)-adenine

Following the procedure of Example 1, 2,6 - dichloropurine was condensed with 1 - benzoyloxy - 2 - chloromethoxyethane to give a 41% yield of 2,6 - dichloro - 9 - (2 - benzoyloxyethoxymethyl)purine, m.p. 121—125°C. This was treated according to the procedure of Example 2 to give simultaneous ammonolysis of the 6 - chloro and benzoyloxy groups yielding 2 - chloro - 9 - (hydroxyethoxymethyl)adenine, in 94% yield, m.p. 188—190°C after recrystallization from isopropanol.

Example 4

2 - Amino - 9 - (2 - hydroxyethoxymethyl)-adenine I; R^1=R^2=NH_2

2 - Chloro - 9 - (2 - hydroxyethoxymethyl)-adenine (0.83 g) was added with stirring to 95% hydrazine (11 ml) and the resulting solution stirred 1 hour under nitrogen. Excess hydrazine was removed by distillation in a rotary evaporator at a bath temperature of 30°C followed by co-evaporation twice with ethyl acetate. The residue was thoroughly triturated with methanol to give a quantitative yield (0.8 g) of white to cream colored 2 - hydrazino - 9 - (2 - hydroxyethoxymethyl)-adenine, m.p. 220—222°C.

A solution of this hydrazine derivative (0.85 g) in 5% aqueous acetic acid (30 ml) was chilled to 5—8°C, and a cold (0°C) solution of sodium nitrite (0.3 g) in water (65 ml) was added in one portion. The reaction mixture was stirred in an ice bath for one hour and then filtered, washed with cold water and dried at room temperature to give 2 - azido - 9 - (2 - hydroxyethoxymethyl)-adenine (0.86 g). Recrystallization from water gave a light mauve product, m.p. 191—192°C.

A mixture of 2 - azido - 9 - (2 - hydroxyethoxymethyl)adenine (0.41 g) and 10% palladium on charcoal (80 mg) in ethanol (41 ml) was shaken under hydrogen at 50 psi for 3.5 hours. The catalyst was removed by filtration through a celite pad and washed thoroughly with ethanol and water. The solvent was removed by evaporation under reduced pressure giving a quantitative yield (0.37 g) of nearly pure (one spot on TLC) 2 - amino - 9 - (2 - hydroxyethoxymethyl)-adenine, m.p. 183—184°C after recrystallization from n - propanol.

Example 5

9 - (2 - Hydroxyethoxymethyl)guanine (I; R^1=OH, R^2=NH_2)

Solid sodium nitrite (0.97 g) was added at room temperature with stirring over a period of one hour to a solution of 2 - chloro - 9 - (2 - hydroxyethoxymethyl)adenine (0.5 g) in glacial acetic acid (10 ml). The reaction mixture was stirred for an additional 4½

hours. The white solid was removed by filtration, washed with cold acetic acid and then well triturated with cold water to remove the sodium acetate present. The solid product was retained. The combined acetic acid filtrate and wash was evaporated at reduced pressure and 40°C bath temperature and the residual oil triturated with cold water. The resulting solid material was combined with the previously isolated solid and the combined solids dried and recrystallized from ethanol to give 2 - chloro - 9 - (2 - hydroxyethoxymethyl)hypoxanthine (0.25 g), m.p. >310°C. Elemental analysis and NMR spectrum were consistent with this structure.

A mixture of 2 - chloro - 9 - (2 - hydroxyethoxymethyl)hypoxanthine (0.375 g) and methanol (80 ml) saturated with anhydrous ammonia was heated in a bomb at 125°C for 5 hours. The bomb was cooled in an ice bath and the reaction mixture removed. Solvent and excess ammonia were removed under reduced pressure at 50°C. After the residue was triturated with cold water to remove the ammonium chloride formed, the remaining solid was dried and then recrystallized from methanol to give pure 9 - (2 - hydroxyethoxymethyl)guanine (0.24 g), m.p. 256.5—257°C.

Example 6

9 - (2 - Hydroxyethoxymethyl)guanine (I; R^1=OH, R^2=NH_2)

A mixture of guanine (2.0 g), ammonium sulfate (1.5 g), and hexamethyldisilizane (126 g) was heated at reflux under nitrogen overnight. Excess hexamethyldisilazane was removed by distillation at reduced pressure. Dry benzene (10 ml) was added to the residual oil and any remaining ammonium sulfate removed by filtration. To this solution was added triethylamine (4 ml) and a solution of 2 - benzoyloxyethoxymethyl chloride (2.8 g) in dry benzene (15 ml) and the mixture heated at reflux under nitrogen overnight. The solvent was evaporated in a rotary evaporator at reduced pressure and the residue dissolved in 95% ethanol. The solution was warmed on a steam bath for ½ hour to effect hydrolysis of the silyl groups. The ethanol was then evaporated and the residual solid thoroughly washed with water, filtered and dried. Recrystallization from methanol and then from water (residual guanine was insoluble in the hot solvents and was removed by filtration) afforded 9 - (2 - benzoyloxyethoxymethyl)guanine (0.58 g, 14% of theroetical), m.p. 222—226°C. A later condensation of the tris - (trimethylsilyl)guanine with a 60% excess of 2 - benzoyloxyethoxymethyl chloride gave a 32% yield of 9 - (2 - benzoyloxyethoxymethyl)guanine.

9 - (Benzoyloxyethoxymethyl)guanine (0.58 g) and methanol (80 ml) saturated with ammonia were heated in a bomb at 80°C for 16 hours. The reaction mixture was removed from the bomb and the solvent evaporated under reduced pressure. The residue was thoroughly washed with ether and then recrystallized from methanol to give 9 - (2 - hydroxyethoxymethyl)guanine (0.31 g, 75% of theoretical), m.p. 256.5—257°C.

Example 7

2 - Amino - 9 - (2 - hydroxyethoxymethyl)-adenine (I; R^1=R^2=NH_2)

A mixture of 2 - chloro - 9 - (2 - hydroxyethoxymethyl)adenine (4.4 g) and liquid ammonia (40 ml) was heated at 120°C in a bomb for 19 hours. The reaction mixture was removed from the bomb and excess ammonia removed *in vacuo*. The residue was partitioned between ether and water and the aqueous layer washed twice more with ether and once with chloroform. The aqueous solution was then treated with an excess of a strongly basic ion exchange resin to convert the ammonium chloride to ammonia and evaporated to dryness *in vacuo*. The residue was dissolved in a minimal amount of ethanol, Florisil (Registered Trade Mark) (10 g) added, and the mixture evaporated in dryness. The residue was transferred to a prepared column of Florisil in chloroform and eluted with 5% methanol/95% chloroform (v/v). Fractions (30 ml) were collected and examined by TLC (silica gel plates developed in 10% methanol/90% chloroform) for the presence of product. The first 40 fractions contained starting material but no product and were discarded. Succeeding fractions which were found to contain the desired product were combined and evaporated to dryness. The residue was recrystallized from ethanol (with activated charcoal treatment) and from n - propanol to give 2 - amino - 9 - (2 - hydroxyethoxymethyl)adenine (0.5 g, 12% of theoretical), m.p. 183—184°C.

Florisil is an activated magnesium silicate widely used in column chromatograph.

Example 8

2 - Amino - 6 - chloro - 9 - (2 - benzoyloxyethoxymethyl)purine

A suspension of potassium carbonate (4.1 g) and 2 - amino - 6 - chloropurine (5.0 g) in dry dimethylformamide (120 ml) was stirred for 20 minutes. 2 - Benzoyloxyethoxymethyl - chloride (6.3 g) was added and the resulting pink suspension stirred at room temperature for 6 days. The reaction mixture was then poured into a mixture of ice and water (210 ml) with vigorously stirring. The aqueous mixture was extracted with chloroform (3×200 ml), and the chloroform solution obtained was washed with a 5% acetic acid solution and then twice with water. The chloroform solution was dried over anhydrous sodium sulfate and evaporated *in vacuo*. The residue was redissolved in a minimal amount of chloroform and the

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resulting solution applied to a column containing silica gel (200 g) in chloroform. The column was eluted with 2% methanol/98% chloroform and the eluant collected in 30 ml cuts. The fractions containing 2 - amino - 6 - chloro - 9 - (2 - benzoyloxyethoxymethyl)-purine (determined by TLC) were combined, evaporated and the residue recrystallized from benzene to give 2 - amino - 6 - chloro - 9 - (2 - benzoyloxyethoxymethyl)purine (2.0 g, m.p. 130—133.5°C).

Example 9

9 - (2 - Hydroxyethoxymethyl)thioguanine (I; R^1=SH, R^2=NH_2)

Thiourea (0.28 g) was added to a refluxing solution of 2 - amino - 6 - chloro - 9 - (2 - benzoyloxyethoxymethyl)purine (1.27 g) in isopropanol (40 ml). The reaction mixture was heated at reflux for 1½ hours and then chilled. 9 - (2 - benzoyloxyethoxymethyl)-thioguanine (0.58 g, m.p. 199—202°C) was removed by filtration. This was added to 40 ml of aqueous ammonia (about 7*N*) and the mixture heated on a steam bath for 10 minutes and then stirred overnight at room temperature. The water and ammonia were removed *in vacuo* and the residue triturated with acetone and then with ether to remove benzamide. The remaining solid was recrystallized twice from water to give yellow flakes of 9 - (2 - hydroxyethoxymethyl)thioguanine (0.26 g, m.p. 251—254°C).

Example 10

9 - (2 - Hydroxyethoxymethyl)guanine (I; R^1=OH, R^2=NH_2)

To a solution of 2 - amino - 9 - (2 - hydroxyethoxymethyl)adenine (0.22 g) in water (30 ml) was added a suspension of adenosine deaminase in aqueous ammonium sulfate (0.44 ml, containing 4.4 mg of the enzyme). The reaction mixture, which initially had a pH of 7.0, was heated at 37°C for 18 hours, at which time the pH was 8.5 and TLC (silica gel plates developed in 15% methanol—85% chloroform) indicated a single product different from starting material. The reaction mixture was thoroughly chilled in an ice bath and the resulting white solid removed by filtration and thoroughly washed with cold water. The product was dried at 100°C/0.1 mm Hg for 16 hours to give 0.20 g of 9 - (2 - hydroxyethoxymethyl)guanine. ¼ H_2O, the structure of which was confirmed by m.p., TLC, U.V., NMR and mass spectroscopy analysis. Recrystallization from methanol gave the anhydrous 9 - (2 - hydroxyethoxymethyl)-guanine.

Example 11

2 - Amino - 9 - (2 - hydroxyethoxymethyl)-adenine hydrochloride

2 - Amino - 9 - (2 - hydroxyethoxymethyl)-adenine (0.25 g) was dissolved in hot ethanol (50 ml) and the solution cooled in an ice bath. To the cool solution was added sufficient HCl-saturated ethanol to give a pH of 1.0. Dry ether (50 ml) was then added and the mixture thoroughly chilled. The resulting flesh-colored solid was removed by filtration and washed with cold ethanol. Recrystallization from methanol yielded 2 - amino - 9 - (2 - hydroxyethoxymethyl)adenine hydrochloride (0.21 g, 74% of theoretical), m.p. 207—211°C. Elemental analysis and NMR support this structure.

Example 12

Properties of 9 - (2 - hydroxyethoxymethyl)-guanine)

9 - (2 - Hydroxyethoxymethyl)guanine was determined by U.V. to be soluble in either water or 0.1N aqueous hydrochloric acid to the extent of about .2%. It was stable in either 0.01N or 0.1N aqueous hydrochloric acid at 26°C and at 37°C, showing no indication of hydrolysis after one week. However, in 1N aqueous hydrochloric acid at either 26°C or 37°C, it slowly hydrolyzed to guanine within one week.

Example 13

Preparation of 2 - amino - 9 - (2 - hydroxy-ethoxymethyl)adenine

2,6 - dichloro - 9 - (2 - benzoyloxyethoxy-methyl)purine 10 g, sodium azide 3.5 g and 54 ml of 1:1 (v/v) ethanol - water were refluxed with stirring for 3½ hours at 110—120°C. TLC of reaction mixture shows complete reaction. On overnight cooling the oil solidified and was filtered, washed with ethanol and water and recrystallised from ethanol. Quantitative yield m.p. 124—125°C. Compound is labile in aqueous base and sensitive to sunlight.

1.2 g of 2,6 - diazido - 9 - (2 - benzoyloxy-ethoxymethyl)purine was dissolved in 150 ml 1:1 methanol tetrahydrofuran and the solvent was put in Paar hydrogenation apparatus containing 33 mg of 10% palladium charcoal which had been wetted with water and shaken under 50 psi hydrogen for 4.5 hours. The catalyst was filtered and washed well with water and methanol. TLC showed complete reduction. Solution was evaporated to dryness giving a yield of 87% of 2,6 - diamino intermediate.

The residue was dissolved in 40% aqueous methylamine minimum amount and was heated for ½ hour on a steam bath, cooled and extracted 3 times with an equal volume of ether. The ether extracts were back-washed with water and the combined aqueous phase was evaporated to dryness. The residue was recrystallised from ethanol to give 89% yield of the compound 2 - amino - 9 - (2 - hydroxy-ethoxymethyl)adenine m.p. 182—183.5°C.

Example 14

Preparation of 2,6 - dichloro - 9 - (2 - Acetyloxyethoxymethyl)purine

2,6 - dichloro purine 5.5 g and 2 - oxa - 1,4 - butanediol diacetate 51.3 g were placed in a flask and partially evacuated and then heated to 138°C, initially the mixture was too thick to stir but gradually gave rise to a melt which was stirred, and after 20 minutes was completely melted and the reaction mixture heated for 10 minutes more (total heat time=30 minutes). The mixture was then cooled to room temperature and para - toluenesulphonic acid 150 mg was added, the vacuum reapplied and heating resumed with stirring. After 20 minutes heating vigorous bubbling was noted, cooled to room temperature, chloroform was added and the solution extracted once with saturated aqueous sodium bicarbonate and once with water. The chloroform phase was dried over anhydrous sodium sulphate and evaporated. The residual oil dissolved in benzene applied to column of 200 g of silica gel in benzene. Benzene elution removed acetate by products. Ether elution gave 9 isomer yield of 64%. Recrystallisation from benzene to give lustrous white flakes m.p. 96—99°C.

Example 15

2,6 - Diamino - 9 - (2 - benzoyloxyethoxymethyl)purine

A mixture of 2,6 - diaminopurine monohydrate (2.0 g), ammonium sulfate (1.32 g), and hexamethyldisilizane (100 g) was heated at reflux under nitrogen for 18 hours. The solvent was evaporated under reduced pressure and the residual oil dissolved in a minimal amount of benzene. To the benzene solution was added 2 - benzoyloxyethoxymethyl chloride (2.56 g), triethylamine (2 ml) and benzene (55 ml). This reaction mixture was heated at reflux under nitrogen for 18 hours. Additional 2 - benzoyloxyethoxymethyl chloride (2.56 g) and triethylamine (2 ml) were added, and heating at reflux was continued for an additional 6 hours. The solvent was evaporated under reduced pressure and the residue digested on a steam bath for 30 minutes in 95% ethanol (40 ml). The solvent was evaporated and the gummy residue recrystallized from ethanol, twice from methanol and finally from water to give 2,6 - diamino - 9 - (2 - benzoyloxyethoxymethyl)-purine as a yellow solid, m.p. 205°C, in 7.5% yield.

Example 16

9 - (2 - hydroxyethoxymethyl)guanine

A solution of sodium benzylate in benzyl alcohol was prepared from sodium (2.58 g) and benzyl alcohol (28 ml). The solution was heated to 120°C and 2 - amino - 6 - chloro - 9 - (2 - benzoyloxyethoxymethyl)purine (3.47 g) was added over a 10 minute period. The reaction mixture was heated overnight with stirring at 120—130°C and then poured into a mixture of ice and water. The resulting mixture was extracted thoroughly with chloroform. The aqueous phase was neutralized with acetic acid, giving a white precipitate which was recrystallized from methanol and then from water to give analytically pure 2 - amino - 6 - benzyloxy - 9 - (2 - hydroxyethoxymethyl)purine, m.p. 291—292°C (dec.) in 50% yield.

This compound is reductively cleaved, e.g. with hydrogen, palladium/charcoal in methanol, to give 9 - (2 - hydroxyethoxymethyl)guanine.

Example 17

9 - (2 - Hydroxyethoxymethyl)guanine

2 - Mercaptoethanol (0.75 ml) dissolved in 1M methanolic sodium methylate (7.5 ml) was added to 2 - amino - 6 - chloro - 9 - (2 - benzoyloxyethoxymethyl)purine (0.89 g) in methanol (150 ml). The reaction mixture was heated at reflux for 3 hours under nitrogen. The solvent was evaporated under reduced pressure and the residue dissolved in water. The aqueous solution was heated on a steam bath for 2 hours, chilled, and acidified to pH 5.0 with acetic acid. The resulting white solid was removed by filtration, washed well with ice cold water and ether, and then recrystallized from methanol to give a 45% yield of 9 - (2 - hydroxymethoxymethyl)guanine.

Example 18

9 - Ethylthiomethyladenine

A reaction mixture containing chloromethyl ethyl sulfide (5.53 g), 6 - chloropurine (7.73 g) and triethylamine (5.57) in dimethylformamide (50 ml) was allowed to stand at room temperature for three days. It was filtered and the filtrate evaporated. The semi-solid residue was dissolved in chloroform (ca. 80 ml), placed on a column of Florisil (activated magnesium silicate) (360 g) in chloroform, and eluted with chloroform. The initial eluate (ca. 500 ml) was discarded; the next 2.8 liters were collected and evaporated. Ligroin (ca. 50 ml) was added to the residual oil, the mixture chilled, and the resulting off white crystal of 6 - chloro - 9 - ethylthiomethylpurine collected by filtration and washed with ligroin; yield 3.3 g (m.p. 71—81°C). Recrystallization from ether - ligroin gave white needles, m.p. 81—82.5°C.

6 - Chloro - 9 - ethylthiomethylpurine (1.5 g) and liquid ammonia (20 ml) were placed in a bomb and heated at 60°C overnight. Evaporation of the ammonia gave a residue which was treated with cold water, filtered, and washed with cold water. This yielded 9 - ethylthiomethyladenine (1.17 g) as a white solid, m.p. 140—142°C. Recrystallization from ethanol gave white plates, m.p. 142—143°C.

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Example 19

9 - (2 - Hydroxyethylthiomethyl)adenine

2 - Acetoxyethanethiol was prepared according to the method of Miles and Owen, *J. Chem. Soc.*, 817 (1952). Acetic anhydride (102.1 g) was added over 1.25 hours to a mixture of 2 - mercaptoethanol (78.1 g) and a 10% solution of sulfuric acid in acetic acid (3 ml). External cooling was used during the addition to keep the reaction temperature below about 40°C. After the addition of acetic anhydride was complete, the reaction mixture was heated at 65°C for one hour and allowed to stand at room temperature overnight. Ether (500 ml) was added to the reaction mixture and the resulting solution washed with water (3×100 ml) and with brine (1×100 ml). The ether solution was dried over anhydrous sodium sulfate and sodium bicarbonate. Distillation gave 2 - acetoxyethanethiol (77.5 g, bp 57—67°C at 10 mm Hg).

Hydrogen chloride gas was introduced into a mixture of 2 - acetoxyethanethiol (24.0 g) and paraformaldehyde (6.0 g) at a moderate rate with external cooling for 3 hours. Calcium chloride (25 g) was added and the reaction mixture allowed to stand in a salt-ice bath for 4 hours. Methylene chloride (200 ml) was added, the reaction mixture filtered and the solvent evaporated with rigorous exclusion of moisture. The residual oil was distilled to give 2 - acetoxyethyl chloromethyl sulfide (18.5 g, bp 82—87°C at 3 mm Hg).

Following the procedure of Schaeffer, *et al. J. Med. Chem, 14,* 367 (1971), 2 - acetoxyethyl chloromethyl sulfide (3.37 g) was added to a mixture of 6 - chloropurine (3.09 g), triethylamine (2.23 g) and dimethylformamide (20 ml). The reaction mixture was stirred at room temperature for 90 hours and the solvent then removed under reduced pressure. The residual oil was dissolved in chloroform (30 ml) and placed on a column of Florisil (200 g) in chloroform. Elution with chloroform resulted in the desired product in 2.1 liters of eluate (after discarding the initial 300 ml collected). The solvent was evaporated to give a residual oil. Ether - pet.ether was added and the mixture chilled to give 9 - (2 - acetoxyethylthiomethyl) - 6 - chloropurine (1.4 g, m.p. 82—87°C). Recrystallization from ether gave white prisms m.p. 89—91°C).

9 - (2 - Acetoxyethylthiomethyl) - 6 - chloropurine (1.1 g) and ammonia (20 ml) were placed in a bomb and heated at 60°C for 24 hours. The reaction mixture was removed from the bomb and the ammonia allowed to evaporate. The residue was triturated with cold water, filtered and washed with additional cold water. The resulting crude 9 - (2 - hydroxyethylthiomethyl)adenine (0.73 g) melted at 166—169.5°C. Recrystallization from ethanol gave white plates, m.p. 170—172°C.

Example 20

9 - {2 - (3 - Carboxypropionyloxy)ethoxymethyl}guanine

A mixture of 9 - (2 - hydroxyethoxymethyl)guanine (0.25 g), succinic anhydride (0.55 g) and pyridine (50 ml) was heated under anhydrous conditions on a steam bath overnight. The solvent was evaporated under reduced pressure at <40°C, the last trace being removed azeotropically with toluene. The residue was triturated with acetone and the product removed by filtration. Recrystallization from methanol afforded 9 - {2 - (3 - carboxypropionyloxy)ethoxymethyl}-guanine, m.p. 203—207°C (sinter 190°C), in 44% yield.

Example 21

9 - Ethylthiomethyl - N - 6 - methyladenine

6 - Chloro - 9 - ethylthiomethylpurine (1.2 g) and methylamine (20 ml) were placed in a bomb and heated at 60°C for 24 hours. The excess methylamine was then allowed to evaporate at room temperature. The residue was triturated with ice water (10 ml) and filtered. Recrystallization of the tan solid from ether with charcoal (Darco 60, Registered Trade Mark) treatment gave 9 - ethylthiomethyl - N - 6 - methyladenine (0.64 g), m.p. 111.5—113°C.

Example 22

9 - (2 - Aminoethoxymethyl)adenine

To a stirred, ice-bath cooled dispersion of sodium hydride (4.0 g of a 60% dispersion in mineral oil) in dimethylformamide (500 ml) was added adenine (13.5 g). After one hour, the ice-bath was removed and the mixture stirred at ambient temperature for an additional 3 hours. A solution of N - (2 - chloromethoxyethyl)phthalimide (23.9 g) in dimethylformamide (100 ml) was added dropwise over 0.5 hour. The reaction mixture was stirred at ambient temperature for 18 hours and then poured with stirring over ice water (2 liters). The resulting solid was recrystallized from 2 - methoxyethanol and then from dimethylformamide to give 9 - (2 - phthalimidoethoxymethyl)adenine (15.0 g), m.p. 256—258°C.

A mixture of 9 - (2 - phthalimidoethoxymethyl)adenine (3.38 g), hydrazine hydrate (1.0 ml of an 85% aqueous solution) and ethanol (150 ml) was heated at reflux for 2 hours. 2 - Methoxyethanol (50 ml) was added and refluxing continued for an additional 2 hours. The solvent was removed under reduced pressure and 1N hydrochloric acid (50 ml) added. The mixture was stirred at ambient temperature for 30 minutes and allowed to stand at 0°C overnight. The solution was filtered and the filtrate diluted with water (100 ml) and stirred with excess basic ion exchange resin {Rexyn 201(OH)}

until a negative silver nitrate test was obtained. It was then filtered and most of the water removed under reduced pressure. The aqueous solution was cooled and the resulting solid removed by filtration. This was recrystallized from isopropanol to give 9 - (2 - aminoethoxymethyl)adenine (1.0 g), m.p. 170—171°C.

Example 23

2,6 - Diamino - 9 - (2 - acetoxyethoxymethyl)-purine

Following the procedure of Example *13*, 2,6 - dichloro - 9 - (2 - acetyloxyethoxymethyl)purine was converted to 2,6 - diazido - 9 - (2 - acetyloxyethoxymethyl)purine, m.p. 69—70°C (from ethanol water), in 83% yield; and this was catalytically reduced by hydrogen over 10% palladium on charcoal in methanol at 50 psi to give 2,6 - diamino - 9 - (2 - acetoxethoxymethyl)purine, m.p. 158—159°C (from ethanol), in 88% yield.

Example 24

2 - Amino - 6 - methylthio - 9 - (2 - hydroxyethoxymethyl)purine

A mixture 9 - (2 - Hydroxyethoxymethyl)-thioguanine (0.7 g), methyl iodide (0.2 ml), Dowex (Registered Trade Mark) HCO_3^- resin (1.7 g) and methanol (200 ml) was stirred overnight at room temperature. The resin was removed by filtration, washed with methanol and the combined filtration evaporated, and the residue recrystallized from isopropanol and then from acetone to give 2 - amino - 6 - methylthio - 9 - (2 - hydroxyethoxymethyl)purine, m.p. 188—191°C in 38% yield.

Example 25

9 - (3 - Hydroxypropoxymethyl)guanine

Sodium benzoate (96.32 g) in DMF (690 ml) was heated to 80°C, and 1 - chloro - 3 - hydroxypropane (63.06 g) was added over 15 minutes. The temperature increased to 135°C, and the reaction mixture was heated at 135—175°C for 3 hours. Filtration removed 38 g of sodium chloride (97% of theory). The filtration was partially evaporated at reduced pressure at <40°C. The concentrated filtrate was poured into ice water and extracted well with ether. The combined ether extracts were washed with water, dried over anhydrous sodium sulfate, and evaporated. The residual oil was distilled through a Vigreux column to give 3 - benzoyloxy - 1 - propanol (85.2 g), b.p. 124—132°C at 0.055 mm Hg.

Anhydrous hydrogen chloride was bubbled into a solution of 3 - benzoyloxy - 1 - propanol (15.02 g) and paraformaldehyde (2.49 g) in dichloromethane (35 ml) for 1 hour at 0°C. The solvent was evaporated under reduced pressure at <40°C giving a 92% yield of crude 3 - benzoyloxypropoxymethyl chloride which was used without purification.

A solution of trimethylsilylated guanine in benzene (25 ml) prepared as in Example 6 (from 2.0 g of guanine) containing triethylamine was heated at reflux and 3 - benzoyloxypropoxymethyl chloride (2.96 g) dissolved in benzene (15 ml) was added over a 3 hour period. The reaction mixture was heated at reflux under nitrogen overnight. The solvent was removed under reduced pressure, and 95% ethanol and methanol were added to the residual oil. The mixture was heated on a steam bath for several minutes and the solvent then evaporated under reduced pressure. Chloroform (200 ml) was added and the resulting solid removed by filtration. The solid was dissolved in a minimal amount of DMF, filtered (to remove any guanine present) and reprecipitated by the addition of water. Recrystallization from methanol (with charcoaling) gave 9 - (3 - benzoyloxypropoxymethyl)guanine (0.94 g) as a pale yellow solid, m.p. 198—201°C.

A mixture of 9 - (3 - benzoyloxypropoxymethyl)guanine (0.5 g) and aqueous 45% methylamine (10 ml) was stirred overnight at room temperature. Excess methylamine and water was evaporated at <30°C under reduced pressure and the residue recrystallized from ethanol to give 9 - (3 - hydroxypropoxymethyl)guanine (0.24 g), m.p. 223°C (with resolidification), as the .½ hydrate.

Example 26

Oil in Water Cream base

9 - (2 - hydroxyethoxymethyl)-guanine	5.0 g
Lanolin, Anhydrous	20.0 g
Polysorbate 60	4.0 g
Sorbitan Monopalmitate	2.0 g
Light Liquid Paraffin	4.0 g
Propylene Glycol	5.0 g
Methyl Hydroxybenzoate	0.1 g
Purified Water	to 100.0 g

Example 27

Water Soluble Ointment Base

2 - amino - 9 - (2 - hydroxy-ethoxymethyl)adenine	0.5 g
Glycerol	15.0 g
Macrogol 300	20.0 g
Polyethylene Glycol 1500	64.5 g

Example 28

Tablet—(Total weight 359 mg)

9 - (2 - hydroxyethoxymethyl)-guanine	100 mg
Lactose	200 mg
Starch	50 mg
Polyvinylpyrrolidone	5 mg
Magnesium Stearate	4 mg

WHAT WE CLAIM IS:—

1. A substituted purine of formula (I)

until a negative silver nitrate test was obtained. It was then filtered and most of the water removed under reduced pressure. The aqueous solution was cooled and the resulting solid removed by filtration. This was recrystallized from isopropanol to give 9 - (2 - aminoethoxymethyl)adenine (1.0 g), m.p. 170—171°C.

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A mixture of 9 - (3 - benzoyloxypropoxymethyl)guanine (0.5 g) and aqueous 45% methylamine (10 ml) was stirred overnight at room temperature. Excess methylamine and water was evaporated at <30°C under reduced pressure and the residue recrystallized from ethanol to give 9 - (3 - hydroxypropoxymethyl)guanine (0.24 g), m.p. 223°C (with resolidification), as the .½ hydrate.

Example 26

Oil in Water Cream base

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Lanolin, Anhydrous	20.0 g
Polysorbate 60	4.0 g
Sorbitan Monopalmitate	2.0 g
Light Liquid Paraffin	4.0 g
Propylene Glycol	5.0 g
Methyl Hydroxybenzoate	0.1 g
Purified Water	to 100.0 g

Example 27

Water Soluble Ointment Base

2 - amino - 9 - (2 - hydroxy-ethoxymethyl)adenine	0.5 g
Glycerol	15.0 g
Macrogol 300	20.0 g
Polyethylene Glycol 1500	64.5 g

Example 28

Tablet—(Total weight 359 mg)

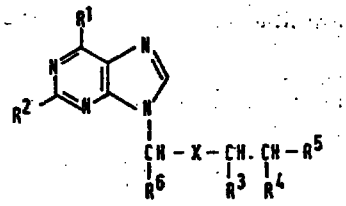
9 - (2 - hydroxyethoxymethyl)-guanine	100 mg
Lactose	200 mg
Starch	50 mg
Polyvinylpyrrolidone	5 mg
Magnesium Stearate	4 mg

WHAT WE CLAIM IS:—

1. A substituted purine of formula (I)

* C L M S *

(I)



wherein X is sulphur or oxygen; R^1 is halogen, hydroxy, hydrogen, mercapto, alkylthio, alkoxy, amino, alkylamino or dialkylamino or azide; R^2 is hydrogen, halogen, amino or azide; R^3 is hydrogen, straight or branched chain or cyclic alkyl, hydroxyalkyl, benzyloxyalkyl or phenyl; R^4 is hydrogen, hydroxy, or alkyl; R^5 is hydrogen, hydroxy, amino, alkyl, hydroxyalkyl, benzoyloxy, benzoyloxymethyl, benzyloxy, sulphamoyloxy, phosphate, carboxypropionyloxy, acetoxy, or substituted carbamoyl group of formula NH.CO—Z wherein Z is alkyl, aryl or aralkyl optionally substituted by one or more of sulphonyl, amino, carbamoyl, or halogen; R^6 is hydrogen, alkyl; provided that when X is oxygen, R^2, R^3, R^4 and R^6 are hydrogen, and R^5 is hydroxy, hydrogen or benzyloxy, R^1 is not amino or methylamino; when R^2 is hydrogen R^1 is not chlorine; or a salt thereof.

2. A substituted purine of formula (I) according to claim 1 wherein X is oxygen atom.

3. A substituted purine of formula (I) according to claim 2 wherein R^1 is hydrogen, halogen, hydroxy, alkoxy, mercapto, alkylthio, amino, alkylamino, dialkylamino or azide; R^2 is hydrogen, halogen, amino or azide; R^3 is hydrogen, straight or branched chain or cyclic alkyl, hydroxyalkyl or phenyl; R^4 is hydrogen, or hydroxy; R^5 is hydrogen, hydroxy, benzoyloxy, hydroxyalkyl, amino, carboxypropionyloxy, acetoxy, benzyloxy, benzoyloxymethyl, phosphate, sulphamoyloxy, substituted carbamoyl group or formula NH.CO—Z where Z is alkyl, aryl or aralkyl optionally substituted by one or more of sulphonyl, amino, carbamoyl, halogen; R^6 is hydrogen, alkyl provided that R^5 is hydroxy only when R^1 is amino, hydroxy, alkylamino, alkylthio or dialkylamino, and R^2 is amino and R^6 is hydrogen; R^5 is hydroxyalkyl only when R^1 is hydroxy; R^5 is hydrogen only when R^1 is hydroxy or halo; when R^5 is benzoyloxy R^2 is not halogen; R^5 is acetoxy only when R^1 is hydroxy or amino and R^2 is amino or both R^1 and R^2 are amino or both halogen; R^5 is a substituted carbamoyl of formula NH.CO—Z wherein Z is a group

$$CH(NH_2)CH_2C_6H_5$$

only when R^1 is dialkylamino; except that when R^5 is hydroxy and R^1 is alkylamino then R^2 is not hydrogen; R^1 is chlorine only when R^2 is not hydrogen; R^1 is not amino or alkylamino when R^5 is benzyloxy and R^2, R^3, R^6 are hydrogen; or a salt thereof.

4. A substituted purine of formula (I) according to claim 2 wherein R^1 is halogen, amino, hydroxy or alkylthio; R^2 is amino; R^5 is hydroxy, benzoyloxy, carboxypropionyloxy, acetoxy or hydroxyalkyl and R^3, R^4 and R^6 are all hydrogen atoms provided that R^5 is hydroxyalkyl only when R^1 is hydroxy and R^5 is acetoxy only when R^1 is amino; or a salt thereof.

5. A substituted purine of formula (I) according to claim 2 wherein R^1 is amino, mercapto or hydroxy when R^2 is amino, R^5 is hydroxy and R^3, R^4 and R^6 are each hydrogen; or a salt thereof.

6. A substituted purine of formula (I) according to claim 1 wherein X is sulphur.

7. A substituted purine of formula (I) according to claim 6 wherein R^1 is halogen, amino or alkylamino when R^2, R^3, R^4, R^5 and R^6 are each hydrogen; or a salt thereof.

8. A substituted purine of formula (I) according to any one of the preceding claims wherein the halogen atom is chlorine.

9. A substituted purine of formula (I) according to any one of the preceding claims wherein the alkyl group has from 1 to 12 carbon atoms.

10. A substituted purine of formula (I) according to claim 9 wherein the alkyl group has from 1 to 8 carbon atom.

11. A substituted purine of formula (I) according to claim 5 wherein R^1 and R^2 are both amino, R^5 is hydroxy and R^3, R^4 and R^6 are each hydrogen.

12. A substituted purine of formula (I) according to claim 5 wherein R^2 is amino, R^1 and R^5 are both hydroxy and R^3, R^4 and R^6 are each hydrogen.

13. A substituted purine of formula (I) according to claim 5 wherein R^1 is chlorine, R^2 is amino, R^5 is benzoyloxy and R^3, R^4 and R^6 are each hydrogen.

14. A substituted purine of formula (I) according to claim 4 wherein R^1 is hydroxy, R^2 is amino, R^5 is benzoyloxy and R^3, R^4 and R^6 are each hydrogen.

15. A substituted purine of formula (I) according to claim 4 wherein R^1 is hydroxy, R^2 is amino, R^5 is hydroxymethyl and R^3, R^4 and R^6 are each hydrogen.

16. A substituted purine of formula (I) according to claim 4 wherein R^1 is methylthio, R^2 is amino and R^5 is hydroxy and R^3, R^4 and R^6 are each hydrogen.

17. A substituted purine of formula (I) according to claim 4 wherein R^1 is hydroxy, R^2 is amino, R^5 is carboxypropionyloxy and R^3, R^4 and R^6 are each hydrogen.

18. A substituted purine of formula (I) according to claim 4 wherein R^1 and R^2 are each amino, R^5 is acetoxy and R^3, and R^4 and R^6 are each hydrogen.

19. A salt of a substituted purine of

formula (I), as claimed in any one of the preceding claims wherein the salt is a pharmaceutically acceptable salt.

20. A salt according to claim 19 wherein the salt is an acid addition salt.

21. A salt according to claim 20 wherein the acid is a pharmaceutically acceptable mineral acid.

22. A salt according to claim 20 wherein the acid is a pharmaceutically acceptable organic acid.

23. A pharmaceutical composition which comprises a substituted purine of the formula (I) where X is sulphur or oxygen; R^1 is halogen, azide, hydroxy, mercapto, hydrogen, alkoxy alkylthio, amino, alkylamino or dialkylamino; R^2 is hydrogen, halogen, amino or azide; R^3 is hydrogen, straight or branched chain or cyclic alkyl, hydroxyalkyl, benzyloxyalkyl or phenyl; R^4 is hydrogen, hydroxy, or alkyl; R^5 is hydrogen, hydroxy, amino, alkyl, hydroxyalkyl, benzoyloxy, benzoyloxymethyl, benzyloxy, sulphamoyloxy, phosphate, carboxypropionyloxy, acetoxy, or substituted carbamoyl group of formula NH.CO—Z where Z is alkyl, aryl or aralkyl optionally substituted by one or more of sulphonyl, amino, carbamoyl, or halogen; R^6 is hydrogen, alkyl, hydroxyalkyl; wherein when R^5 is alkyl group it has from 1 to 8 carbon atoms and in all other cases when the substituents have an alkyl moiety it has from 1 to 4 carbon atoms; or a pharmaceutically acceptable salt thereof; together with a pharmaceutically acceptable carrier therefor.

24. A pharmaceutical composition as claimed in claim 23 which comprises the compound of the formula as defined in any one of claims 2 to 21 or a pharmaceutically acceptable salt thereof; together with a pharmaceutically acceptable carrier therefor.

25. A pharmaceutical composition as claimed in claim 23 which comprises the compound of the formula as claimed in claim 11, or a pharmaceutically acceptable salt thereof; together with a pharmaceutically acceptable carrier therefor.

26. A pharmaceutical composition as claimed in claim 23 which comprises the compound of the formula as defined in claim 12, or a pharmaceutically acceptable salt thereof; together with a pharmaceutically acceptable carrier therefor.

27. A pharmaceutical composition as claimed in any one of claims 23 to 26 which is in the form of an ointment or cream.

28. A pharmaceutical composition as claimed in any one of claims 23 to 26 which is in the form of a tablet.

29. A pharmaceutical composition for oral or parenteral administration as claimed in any one of claims 23 to 26 comprising the compound in an amount of 1 to 250 mg per unit dose.

30. A pharmaceutical composition as claimed in any one of claims 23 to 26 for parenteral administration or eye or nose drops comprising the compound in a concentration from 0.1 to 10% w/v in an aqueous medium.

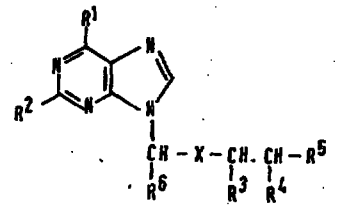
31. A pharmaceutical composition for topical administration as claimed in any one of claims 23 to 26 comprising the compound in a concentration from 0.1 to 10% w/v.

32. A pharmaceutical composition as claimed in claim 31 comprising the compound in a concentration of 1% w/v.

33. A method of preparing a pharmaceutical composition as claimed in any one of claims 22 to 31 which comprises bringing into association a substituted purine of formula (I) as defined in any one of claims 23 to 26, or a pharmaceutically acceptable salt thereof, and a pharmaceutically acceptable carrier therefor.

34. A process for preparing a substituted purine of formula (I)

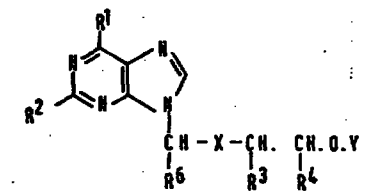
(i)



wherein X, R^1, R^2, R^3, R^4, R^5, and R^6 are defined in claim 1; or a salt thereof, characterised in that

(a) a protecting group Y is cleaved from a compound of formula (II)

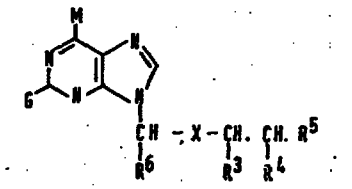
(ii)



by hydrolysis or hydrogenolysis to give a compound of formula (I) wherein R^5 is hydroxy; or

(b) a compound of formula (III)

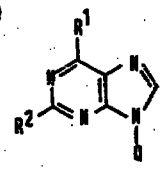
(iii)



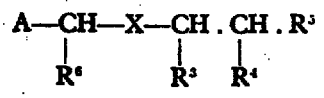
wherein either or both of M and G represent a halogen atom, amino, hydroxy, azido, aralkoxy or mercapto is converted into a compound of formula (I) by methods known per se; or

(c) a compound of formula (IV)

(iv)



is reacted with a compound of formula (V)



(V)

wherein A is a leaving group and Q is hydrogen or a leaving group; or

(d) a blocking group is linked to R^1 and/or R^2 in a compound of formula (I), the blocking group or groups being removed by solvolysis or alcoholysis,

and where the product of said reaction is a base optionally converting a compound of formula (I) into an acid addition salt thereof, or where the product is a salt of a compound of formula (I), optionally converting said salt into its base or another salt thereof.

35. A process as claimed in claim 34 a), c), or d) characterised in that following the formulation of a compound of formula (I), said compound is converted into another compound of formula (I) by the method as claimed in claim 34 (b).

36. A process as claimed in either claim 34 or claim 35 wherein X, R^1, R^2, R^3, R^4, R^5 and R^6 are as defined in claim 3, or a salt thereof.

37. A process as claimed in either claim 34 or claim 35 wherein X, R^1, R^2, R^3, R^4, R^5, and R^6 are defined in claim 4; or a salt thereof.

38. A process as claimed in either claim 34 or claim 35 wherein X is oxygen; R^1 is amino, mercapto or hydroxy when R^2 is amino, R^5 is hydroxy and R^3, R^4, and R^6 are each hydrogen; or a salt thereof.

39. A process as claimed in either claim 34 or claim 35 wherein X is sulphur; R^1 is halogen, amino or alkylamino when R^2, R^3, R^4, R^5 and R^6 are each hydrogen; or a salt thereof.

40. A process as claimed in either claim 34 or claim 35 wherein X is oxygen; R^1 and R^2 are both amino, R^5 is hydroxy; and R^3, R^4 and R^6 are each hydrogen.

41. A process as claimed in either claim 34 or claim 35 wherein X is oxygen; R^2, is amino; R^1 and R^5 are both hydroxy and R^3, R^4 and R^6 are each hydrogen.

42. A process as claimed in claim 41 wherein the protective group Y is aliphatic or aromatic acyl (as herein defined) or arylmethyl group.

43. A process as claimed in claim 42 wherein the acyl group is removed by mild basic hydrolysis.

44. A process as claimed in claim 42 wherein the arylmethyl group is removed by catalytic or chemical hydrogenolysis.

45. A process as claimed in any one of claims 34 and 35 to 41 wherein the leaving group A is a reactive residue of an organic or inorganic acid and Q is hydrogen or acyl (as herein defined).

46. A process as claimed in claim 45 wherein A is halogen, carboxylate.

47. A process as claimed in claim 46 wherein when A is halogen and Q is hydrogen, condensation is carried out in the presence of a base.

48. A process as claimed in claim 46 wherein when A is carboxylate and Q is hydrogen, or A is carboxylate or halogen and Q is acyl, fusion is carried out in the presence of a catalytic amount of a strong acid.

49. A process as claimed in any one of claims 34e and 35 to 41 wherein R^1 and/or R^2 is trimethylsilylated, and said grouping is removed by hydrolysis or alcoholysis.

50. A process as claimed in claims 34a and 34b and 35 to 41 wherein processes a) and b) are combined in a single step.

51. A process as claimed in claim 50 wherein the protective group Y is aliphatic or aromatic acyl (as herein defined) or arylmethyl group.

52. A process as claimed in claim 50 wherein one of R^1 and R^2 is hydrogen or one or both of R^1 and R^2 is halogen, mercapto, azido, aralkoxy, and said substituent(s) is converted into amino, hydroxy, alkylamino or dialkylamino.

53. A process as claimed in claim 50 wherein the conversion is carried out by ammonolysis.

54. A process for preparing an acid addition salt, which comprises mixing 9 - hydroxyethoxymethyl adenine with an acid to form a salt.

55. A process for preparing an acid addition salt, which comprises mixing 6 - methylamino - 9 - (2 - hydroxyethoxymethyl)-purine with an acid to form a salt.

56. A compound of formula I as claimed in claim 1 substantially as hereinbefore described with reference to any one of the foregoing Examples.

57. A pharmaceutical composition substantially as hereinbefore described with reference to any one of Examples 26 to 28.

58. A method of preparing a pharmaceutical composition substantially as hereinbefore described with reference to any one of the foregoing Examples.

59. A method of preparing a compound of formula (I) as claimed in claim 1 substantially as hereinbefore described with

reference to any one of the foregoing Examples.

60. A method of treating viral infections in a mammal other than a human being comprising the administration of a substituted purine of formula (I), as defined in claim 1, or a pharmaceutically acceptable salt thereof.

61. A method as claimed in claim 60 wherein administration is by topical application.

62. A method as claimed in claim 60 wherein administration by the oral or parenteral route.

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PATENTS ACT 1949

SPECIFICATION NO 1523865

The following amendments were allowed under Section 29 on 16 August 1979:

Page 1, line 70, page 11, line 21, *after* chlorine *insert* provided also that when x is oxygen, R^2, R^3, R^4 and R^5 are hydrogen and R^6 is methyl then R^1 is not ethylamino or amino

Page 2, line 20, *after* hydrogen *insert* provided also that when R^2, R^3, R^4, R^5 are hydrogen and R^6 is methyl then R^1 is not ethylamino or amino

Page 11, line 57, *after* hydrogen *insert* provided also that when x is oxygen, R^2, R^3, R^4, R^5 are hydrogen and R^6 is methyl then R^1 is not ethylamino or amino

Page 12, line 79, *for* 22 to 31 *read* 23 to 32

Page 13, line 49, *delete* claim *insert* any one of claims 34 to

Page 13, line 76, *for* 34e *read* 34d

THE PATENT OFFICE
18 September 1979

Bas 71163/11