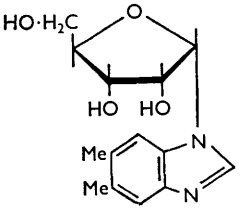
845. *The Synthesis of 7-Glycosylpurines. Part I. Syntheses from Purines.*

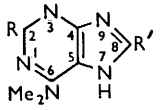
By G. M. Blackburn and A. W. Johnson.

Condensation of mercurichloride derivatives of several purines with acylhalogeno-sugars has been studied and a possible mechanism for the reaction is suggested. Two 7-glycosylpurines have been synthesised and adenosine has been obtained by a modified route.

Several naturally occurring compounds are known which differ from vitamin B_{12} only in that the 5,6-dimethylbenzimidazole portion of the molecule is replaced by a purine, *e.g.*, pseudovitamin B_{12} [1] (adenine), factor A [2,3] (2-methyladenine), factor G [4] (hypoxanthine), factor H [3,4] (2-methylhypoxanthine) and un-named factors containing 2-methylthioadenine [5] and guanine [6] as well as certain biosynthetic compounds.[7] By analogy with the vitamin B_{12} hydrolytic fragment, α-ribazole [8] (I), 5,6-dimethyl-1α-D-ribofuranosylbenzimidazole, the structure of which has been confirmed by synthesis,[9,10] it was expected that the nucleoside of pseudovitamin B_{12} would have an α-glycosidic linkage. Moreover, on the basis of *X*-ray crystallographic studies, Hodgkin [11] predicted that the ribose would be attached to $N_{(7)}$ of the adenine molecule.



(I)



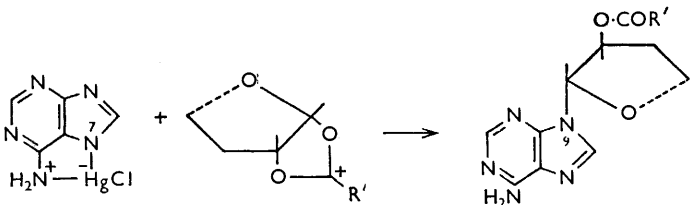
(II) R = R′ = H
(III) R = R′ = SMe
(IV) R = SMe, R′ = H

The name " pseudonucleoside " has been suggested [12] to describe a 7-glycosylpurine, and the present paper describes attempts to synthesise these compounds. The synthesis of purine nucleosides by the reaction of acetobromoglucose with the silver derivatives of purines [13] was modified by Davoll and Lowy [14] who substituted the chloromercuri-derivatives of the purines, and several of the natural nucleosides,[15] including adenosine,[14,16] have been prepared by this method. A feature of this reaction is that, with few exceptions, it is the 9-position of the purine which undergoes substitution. Thus, as in the case of 6-acetamidopurine, we have found that 6-chloropurine reacts with acetobromoglucose to give the 6-chloro-9-tetra-acetylglucopyranosylpurine, identified by conversion into the known 9-β-D-glucopyranosyladenine [13] by treatment with ammonia. The 9-ribosyl derivative of 6-chloropurine has also been described.[17] An early exception to this general

[1] Dion, Calkins, and Pfiffner, *J. Amer. Chem. Soc.*, 1952, **74**, 1108; *Fed. Proc.*, 1952, **11**, 269.
[2] Ford, Porter, *et al.*, *Biochem. J.*, 1952, **51**, v; *Nature*, 1953, **171**, 148.
[3] Brown, Cain, Gant, Parker, and Smith, *Biochem. J.*, 1955, **59**, 82.
[4] Brown and Smith, *Biochem. J.*, 1954, **56**, xxxiv.
[5] Friedrich and Bernhauer, *Chem. Ber.*, 1957, **90**, 1966.
[6] Friedrich and Bernhauer, *Angew. Chem.*, 1959, **71**, 311.
[7] Kon, Ciba Symposium on " Chemistry and Biology of Purines," J. and A. Churchill, London, 1957, p. 169.
[8] Brink, Holly, Shunk, Peel, Cahill, and Folkers, *J. Amer. Chem. Soc.*, 1950, **72**, 1866.
[9] Holly, Skunk, Peel, Cahill, Lavigne, and Folkers, *J. Amer. Chem. Soc.*, 1952, **74**, 4521.
[10] Johnson, Miller, Mills, and Todd, *J.*, 1953, 3061.
[11] Hodgkin, *Biochem. Soc. Symposia*, 1955, **13**, 28 (footnote).
[12] Friedrich and Bernhauer, *Chem. Ber.*, 1956, **89**, 2507.
[13] Fischer and Helferich, *Ber.*, 1914, **47**, 210.
[14] Davoll and Lowy, *J. Amer. Chem. Soc.*, 1951, **73**, 1650.
[15] Schaeffer and Thomas, *J. Amer. Chem. Soc.*, 1958, **80**, 3738, 4896.
[16] Brown and Weliky, *J. Org. Chem.*, 1958, **23**, 125.
[17] Brown and Weliky, *J. Biol. Chem.*, 1953, **204**, 1019.

rule was glucosyltheophylline [13] in which the sugar reacts at the 7-position of the purine, but later, in their synthetic work on puromycin,[18] Baker and his co-workers showed that the mercurichlorides of 6-dimethylaminopurine (II) and 6-dimethylamino-2,8-dimethylthiopurine (III) react with acetobromoglucose to give 7-glucosyl derivatives but that 6-dimethylamino-2-methylthiopurine (IV) gives the 9-glucosyl derivative. However, 2,3,5-tri-*O*-benzoyl-D-ribofuranosyl chloride reacts with the purines (II) and (IV) to give the 9-derivatives but with the thio-compound (III) to give a mixture of the 9- and the 7-isomer. By analogy with the structure of the cupric adenine complex,[19] it seems probable that the monomercurichlorides of adenine should have the preferred planar structure (V). If this is so, then the complex must react at position 9 with the orthoester cation (VI) of the acylhalogeno-sugar by an $S_{E2'}$ mechanism, and furthermore according to Baker [20] the nucleoside formed (VII) has a 1,2-*trans*-configuration (*e.g.*, β- from glucose, α- from arabinose, etc.) in the sugar regardless of the original configuration at positions 1 and 2.

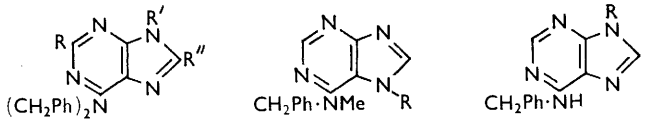
It is suggested that the 6-dimethylamino-group, because of its size, hinders the formation of the mercury–nitrogen bond at $N_{(7)}$ and this is therefore formed at $N_{(9)}$, where there is greater electron-availability than at $N_{(1)}$ or $N_{(3)}$, and leads to the formation of a 7-β-glucoside. However, when the mercurichloride derivative of the 6-dimethylaminopurine



(V) (VI) (VII)

is treated with the tribenzoylribofuranosyl cation, the increased bulk of the latter prevents its approach to $N_{(7)}$ and this ion therefore reacts by displacing the mercury at $N_{(9)}$, *e.g.*, by an S_{E1} mechanism. In order to verify this hypothesis, it was necessary to observe the effect of increasing size of alkyl substituents on the 6-amino-group, and as these should be capable of removal for the synthesis of purine 7-nucleosides, benzyl groups were used in the first instance.

The first dibenzylaminopurine was prepared by treating 2,6,8-trichloropurine [21] with dibenzylamine. The 6-chloro-group is the most reactive [22] and the 2,8-dichloro-6-dibenzylaminopurine (VIII; R = R'' = Cl, R' = H) was treated with acetobromoglucose. The product absorbed at 284 mμ which showed it to be the 9-tetra-acetylglucosyl derivative [VIII; R = R'' = Cl, R' = $C_6H_7O(OAc)_4$] by comparison with 2,8-dichloro-6-dibenzyl-



(VIII) (IX) (X)

amino-9-methylpurine (VIII; R = R'' = Cl, R' = Me) which absorbs at 283 mμ (this compound was prepared from 2,6,8-trichloro-9-methylpurine [22] and dibenzylamine).

[18] Baker *et al.*, *J. Org. Chem.*, 1954, **19**, 1780; *J. Amer. Chem. Soc.*, 1955, **77**, 18.
[19] Harkins and Freiser, *J. Amer. Chem. Soc.*, 1958, **80**, 1132.
[20] Baker, Joseph, Schaub, and Williams, *J. Org. Chem.*, 1954, **19**, 1786; Baker, Ciba Symposium on "Chemistry and Biology of Purines," J. and A. Churchill, London, 1957. p. 120.
[21] Davoll and Lowy, *J. Amer. Chem. Soc.*, 1951, **73**, 2936.
[22] Fischer. *Ber.*, 1897, **30**, 2220.

6-Dibenzylaminopurine (VIII; R = R′ = R″ = H), from 6-chloropurine [23] and dibenzylamine, gave the 9-tetra-acetylglucosyl derivative [VIII; R = R″ = H, R′ = $C_6H_7O(OAc)_4$], which absorbed at 278 mμ, in agreement with the absorption of 6-dimethylamino-9-methylpurine [24] at 277·5 mμ. Sodium in liquid ammonia has been used for the removal of benzyl groups from *N*-benzyl compounds, even in the presence of an *N*-glycosyl group.[25] In the present work catalytic hydrogenation failed to remove the benzyl groups and reaction with sodium and liquid ammonia resulted in removal of only one benzyl group from the 6-dibenzylamino-group. Thus, both the 6-dibenzylaminopurine glycosides [VIII; R = R″ = Cl, R′ = $C_6H_7O(OAc)_4$; and R = R″ = H, R′ = $C_6H_7O(OAc)_4$] gave the same product 6-benzylamino-9-D-glucopyranosylpurine; and 2,8-dichloro-6-dibenzylaminopurine was reduced to the known 6-benzylaminopurine [26] (X; R = H) which confirms the structure of the compound (VIII; R = R″ = Cl, R′ = H).

It seemed probable that 6-(*N*-benzyl-*N*-methylamino)purine would show orientation effects intermediate between those of 6-dimethylamino- and 6-dibenzylamino-purine. In a personal communication, Dr. Stokstad has confirmed that the melting point in his paper [27] of the *N*-benzyl-*N*-methylamino-derivative was misquoted as 114·5—115° and should have read 214·5—215°, as we have found. When this product was treated with acetobromoglucose, the light absorption of the crude product showed a maximum at 302 mμ and a strong inflexion at 280 mμ. This indicated the presence of both $N_{(7)}$- and $N_{(9)}$-glycosyl residues as 7-benzyl-6-dimethylaminopurine absorbed at 300 mμ and 6-dimethylamino-9-ethylpurine at 277·5 mμ.[28] Pure 6-*N*-benzyl-*N*-methylamino-7-(tetra-acetylglucopyranosyl)purine [IX; R = $C_6H_7O(OAc)_4$] was isolated from the mixture and had light absorption maxima at 233 and 305 mμ; the optical rotation, $[\alpha]_D^{22}$ −15°, compares with the values of other tetra-acetyl-β-D-glucopyranosylpurines.[13] However, when 6-(*N*-benzyl-*N*-methylamino)purine reacted with tri-*O*-benzoyl-D-ribofuranosyl chloride, no evidence was obtained for the formation of the 7-ribosyl compound in that the first material eluted from the chromatogram absorbed at 277 mμ and showed no shoulder at 300 mμ.

6-Benzylaminopurine [26] was caused to react with tri-*O*-acetylribopyranosyl chloride [29] and, after purification, the product, 6-benzylamino-9-triacetylribopyranosylpurine [X; R = $C_5H_6O(OAc)_3$], although not obtained crystalline, showed absorption maxima at 208 and 271 mμ. Deacetylation of this compound gave crystalline 6-benzylamino-9-β-D-ribopyranosylpurine (X; R = $C_5H_9O_4$) which absorbed at 209 and 268 mμ. This, when compared with 6-butylamino-9-methylpurine [30] (absorption maximum at 269 mμ), confirms the 9-glycosyl structure of both compounds. Thus 6-benzylaminopurine, like 6-acetamido- and 6-benzamido-purine,[31] presumably forms the chloromercuri-derivative at position 7 and reacts with the acylhalogeno-sugar to give a 9-glycosyl derivative. On the other hand, 6-dimethylamino- and 6-(*N*-benzyl-*N*-methylamino)-purine form mercury complexes at position 9 as steric hindrance prevents reaction at position 7. These purines react with acetobromoglucose to give 7-glycosyl derivatives by an $S_{E2'}$ reaction although the greater steric hindrance in the latter purine is shown by the formation of some 9-derivative. Similarly 6-dibenzylamino- and 2,8-dichloro-6-dibenzylamino-purine form complexes at position 9 but the size of the 6-substituent prevents reaction at $N_{(7)}$ and consequently reaction with the acetohalogeno-sugars by an S_{E1} reaction again yields the 9-derivatives. The change from acetobromoglucose to benzoylchlororibose also yields the 9-glycosyl derivative for steric reasons, but in this case the dominating feature is the size of the protecting groups

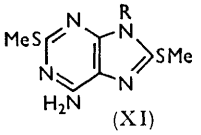
[23] Bendich, Russell, and Fox, *J. Amer. Chem. Soc.*, 1954, **76**, 6073.
[24] Montgomery and Holum, *J. Amer. Chem. Soc.*, 1958, **80**, 404.
[25] Shaw, *J. Amer. Chem., Soc.*, 1958 **80**, 3899.
[26] Skinner and Shive, *J. Amer. Chem. Soc.*, 1955, **77**, 6692.
[27] Bullock, Hand, and Stokstad, *J. Amer. Chem. Soc.*, 1956, **78**, 3693.
[28] Baker, Schaub, and Joseph, *J. Org. Chem.*, 1954, **19**, 638.
[29] Zinner, *Chem. Ber.*, 1950, **83**, 153.
[30] Montgomery and Temple, *J. Amer. Chem. Soc.*, 1957, **79**, 5238.
[31] Recondo and Rinderknecht, *Helv. Chim. Acta*, 1959, **42**, 1171.

on the sugar rather than that of the 6-substituent on the purine. A solvent of low dielectric constant, xylene, was used in all these reactions in order to favour reaction at $N_{(7)}$ according to the above interpretation of the reaction mechanism.

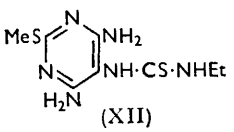
When the purines also contain methylthio-substituents, it is more difficult to assess the factors controlling the position of entry of the sugar. Thus introduction of a methylthio-group into 6-dimethylaminopurine causes reaction with acetobromoglucose to occur at $N_{(9)}$ rather than $N_{(7)}$,[18] and alkylation of these purines in alkaline solution gives a mixture of 18% of the 7- and 31% of the 9-alkyl derivatives from the 6-dimethylamino-2,8-dimethylthiopurine, but mainly the 7-alkyl derivative from 6-dimethylamino-2-methylthiopurine.[18] It seemed that the electronic effects of the methylthio-groups might outweigh steric considerations and the reactions of the methylthiopurines with acetohalogeno-sugars were examined in the hope that they might lead to 7-glycosylpurines after removal of the methylthio-group with Raney nickel. 6-Dibenzylamino-2-methythiolpurine was synthesised for such studies, but sufficient quantities could not be obtained and attention was turned to derivatives of adenine (unsubstituted 6-amino-group). The above dibenzylaminopurine was obtained from 6-amino-4-chloro-2-methylthiopyrimidine [32] by reaction with dibenzylamine to yield the 4-dibenzylamino-compound. Nitrosation, reduction, and cyclisation then gave the required purine in poor yield.

2,8-Dimethylthioadenine (XI; R = H) was prepared from 4,5,6-triamino-2-methylthiopyrimidine [32] by cyclisation with carbon disulphide and methylation of the resulting 8-mercaptopurine. The corresponding 9-methyl derivative (XI; R = Me) was prepared by a similar cyclisation of 5,6-diamino-4-methylamino-2-methylthiopyrimidine [32] and was identical with the sole methylation product of the purine (XI; R = H).

An attempted unambiguous synthesis of compound (XI; R = Et) by cyclisation of the condensation product (XII) of ethyl isothiocyanate and 4,5,6-triamino-2-methylthiopyrimidine gave 8-mercapto-2-methylthioadenine by elimination of ethylamine instead of the product of loss of ammonia. Synthesis of the 7-methyl derivative also proved difficult but was eventually accomplished by reaction of 2,8-dichloro-7-methyladenine [33] with potassium hydrogen sulphide and subsequent methylation of the 2,8-dimercapto-7-methyladenine. Methanethiol could not be used directly, as we had observed that with 2,8-dichloro-9-methyladenine [34] this thiol caused replacement of only the 8-chloro-substituent. In another attempt to synthesise 7-methyl-2,8-dimethylthioadenine, 4,6-diamino-5-methylamino-2-methylthiopyrimidine (XIII) was obtained in variable yield by reduction of the



(XI)



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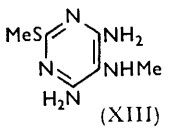


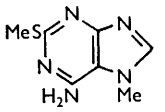
(XIII)

corresponding 5-formamido-compound [32] with lithium aluminium hydride. Although cyclisation of compound (XIII) with carbon disulphide could not be achieved, its structure was confirmed by cyclisation with formamide, which afforded 7-methyl-2-methylthioadenine (XIV). A comparison of the absorption spectra of this product (XIV) with those of 2-methylthiopseudoadenosine [5] affords positive evidence that the latter is a 7-glycosylpurine.

When the mercurichloride derivative of 2,8-dimethylthioadenine (XI; R = H) reacted with acetobromoglucose, the glycosyl derivative isolated was shown to be the 9-isomer (XI; R = $C_6H_{11}O_5$) as the absorption spectrum was almost identical with that of the 9-methyl derivative (max. at 230 and 290 mμ). A similar reaction with 2,3,5-tri-*O*-acetyl-D-ribofuranosyl chloride gave a crude product which after chromatographic separation

[32] Baddiley, Lythgoe, McNeil, and Todd, *J.*, 1943, 383.
[33] Fischer, *Ber.*, 1898, **31**, 104.
[34] Fischer, *Ber.*, 1899, **32**, 267.

yielded a fraction which absorbed at 297 mμ, followed by the 9-tri-*O*-acetylribofuranosyl compound [XI; R = $C_5H_6O(OAc)_3$] ($\lambda_{max.}$ 233 and 290 mμ) as the bulk of the crude product. The first fraction was further purified but could not be crystallised; however, the close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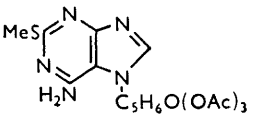
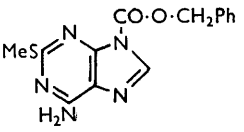

(XV)

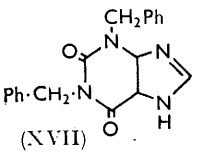

(XVI)

resemblance of its absorption spectrum ($\lambda_{max.}$ at 232 and 297 mμ) to that of 7-methyl-2,8-dimethylthioadenine indicated that this substance was the acetylated 7-nucleoside (XV). Partition chromatography on Celite with 10% aqueous methanol as the stationary phase was not satisfactory for the separation.

When product (XV) was treated with Raney nickel under desulphurising conditions, the resulting solution absorbed at 273 mμ with an inflection at 286 mμ. 7-Methyladenine [35] absorbs at 272 mμ at pH 7, and 7-methyl-2-methylthioadenine (XIV) in water at 286 mμ. After hydrolysis with dilute acid, the desulphurisation product absorbed at 264 mμ (cf. adenine at pH 2, 263 mμ [36]) and D-ribose was detected in the hydrolysate by chromatography.

When the mercurichloride of 2,8-dimethylthioadenine (XI; R = H) reacted with 2,3,5-tribenzoylribofuranosyl chloride, the 9-glycosyl derivative [XI; R = $C_5H_6O(OBz)_3$] ($\lambda_{max.}$ 234 and 292 mμ) was obtained with no detectable trace of the 7-isomer. This would accord with the view that the chloromercuri-complex is formed at $N_{(9)}$. Deacetylation of both the 9-triacetyl- and 9-tribenzoyl-ribosylpurines gave the same 2,8-dimethylthio-9-β-D-ribofuranosyladenine [XI; R = $C_5H_6O(OH)_3$]. Its structure was confirmed by desulphurisation of the tribenzoyl compound with Raney nickel and subsequent debenzoylation, adenosine being obtained in 25% yield.

In another synthetic approach to the 7-glycosylpurines, it was proposed to utilise the observation [37] that reaction of deoxyguanylic acid with methyl iodide leads to the transitory formation of the quaternary 7-methiodide which rapidly hydrolyses to 7-methylguanine. Application of such a method to the synthesis of 7-glycosylpurines necessitates the presence of a labile substituent at position 9 which can be removed without hydrolysis of the 7-sugar substituents, and the benzyloxycarbonyl group was selected for this purpose. As inosine has been shown [25] to be benzylated at position 1, a 2-methylthio-group was also included to ensure quaternisation at position 7. 9-Benzyloxy-2-methylthioadenine (XVI) ($\lambda_{max.}$ 235 and 277 mμ) was synthesised from the sodium salt of the purine and benzyl chloroformate. It proved to be unstable in hot methanol and after a short time the parent purine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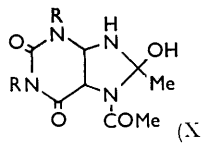
(XVIII)

was recovered. When a suspension of the ester (XVI) in anhydrous acetone reacted with 2,3,5-tribenzoylribofuranosyl chloride, three products were isolated. The principal product was 2-methylthioadenine; some unchanged (XVI) was also obtained and finally 1,3,5-tri-*O*-benzoylribofuranose.[38] It was clear that, even if reaction had occurred between the purine and the halogeno-sugar, any intermediate formed was very sensitive to hydrolysis.

[35] Prasad and Robins, *J. Amer. Chem. Soc.*, 1957, **79**, 6401.
[36] Cavalieri, Bendich, Tinker, and Brown, *J. Amer. Chem. Soc.*, 1948, **70**, 3075.
[37] Lawley, *Proc. Chem. Soc.*, 1957, 290.
[38] Ness and Fletcher, *J. Amer. Chem. Soc.*, 1956, **78**, 4710.

The difficulty of forming quaternary salts from 9-substituted purines was emphasised by the failure of 9-benzyl-2,8-dimethylthioadenine to give a methiodide, from which it had been hoped to prepare the 7-methyl compound by reduction with sodium in liquid ammonia. It appears from the above results that the preparation of purine 7-nucleosides by this method requires the presence of a 6-substituent of moderate size on the purine ring, sufficiently large to allow the formation of the mercury complex at $N_{(9)}$ but not large enough to prevent the approach of the sugar to $N_{(7)}$. The substituent must also be capable of easy conversion into amino- or hydroxy-substituents.

An attempt was also made to utilise the observation [13] that acetobromoglucose and theophylline give the 7-derivative. The factors governing the course of this reaction were undoubtedly different from those obtaining for the 6-dimethylaminopurines, as theophylline possesses a cyclic amide nucleus by virtue of the 1- and 3-alkyl groups. In order to preserve the latter feature, 1,3-dibenzylxanthine (XVII) was synthesised as it was hoped that the benzyl groups might be removed by hydrogenolysis after reaction with the sugar derivative. This compound (XVII) was obtained by benzylation of 4,5-diaminouracil " diacetate " (XVIII; R = H) which is known to be methylated at positions 1 and 3. The resulting compound (XVIII; R = CH_2Ph) was cyclised by heating it with formamide, whereupon the desired product (XVII) ($\lambda_{max.}$ 274 mμ similar to that of theophylline) was obtained, but the yield was low and this approach was abandoned in favour of the methods described earlier.

Experimental

Absorption spectra are recorded for ethanolic solutions except where otherwise stated.

" 4,5-*Diamino*-1,3-*dibenzyluracil Diacetate* " (9-*Acetyl*-1,3-*dibenzyl*-1,2,3,4,8,9-*hexahydro*-8-*hydroxy*-8-*methyl*-2,4-*dioxopurine*) (XVIII; R = CH_2Ph).—" 4,5-Diaminouracil diacetate " [39] (1·0 g.) was heated under reflux in dry dimethylformamide (20 c.c.) with sodium hydrogen carbonate (1·0 g.). Benzyl chloride (1·3 c.c.) was added dropwise and, after 2 hr., the solution was cooled and evaporated to dryness *in vacuo*. The residue was triturated with water (10 c.c.) and extracted with ether (2 × 30 c.c.). The extracts were washed with aqueous sodium hydrogen carbonate solution and water and then dried (Na_2CO_3). After evaporation, the residual *product* crystallised from ethanol as needles (0·2 g.), m. p. 237° (Found: C, 64·9; H, 5·1; N, 13·6. $C_{22}H_{22}O_4N_4$ requires C, 65·0; H, 5·45; N, 13·8%), $\nu_{max.}$ (KBr disc) 1656s and 1704s cm.$^{-1}$.

1,3-*Dibenzylxanthine* (XVII).—A solution of the above dibenzyl derivative (200 mg.) in formamide (10 c.c.) was heated under reflux during 15 min. and poured into water (15 c.c.). The *xanthine* was collected and crystallised from methanol and from benzene–light petroleum (b. p. 60—80°) as rods (130 mg.), m. p. 225° (corr.) (Found: C, 69·1; H, 5·05; N, 16·9. $C_{19}H_{16}O_2N_4$ requires C, 68·7; H, 4·85; N, 16·85%), $\lambda_{max.}$ 204 and 274 mμ (log ε 4·58 and 4·06).

2,8-*Dichloro*-6-*dibenzylaminopurine* (VIII; R = R″ = Cl, R′ = H).—2,6,8-Trichloropurine [21] (2 g.) and dibenzylamine (3·5 c.c.) were heated under reflux in dry butan-1-ol during 3 hr. and the solution was cooled. The precipitate was dissolved in 0·1N-sodium hydroxide (50 c.c.) and the solution extracted with ether (2 × 50 c.c.) and acidified with dilute acetic acid to pH 5. The *purine* separated and crystallised from ethanol in needles (2·5 g.), m. p. 257° (corr.) (Found: C, 56·9; H, 4·4; N, 17·1; Cl, 17·6. $C_{19}H_{15}N_5Cl_2,H_2O$ requires C, 56·8; H, 4·25; N, 17·35; Cl, 17·7%).

2,8-*Dichloro*-6-*dibenzylamino*-9-*methylpurine* (VIII; R = R″ = Cl, R′ = Me).—2,6,8-Trichloro-9-methylpurine [22] (0·5 g.) and dibenzylamine (1 c.c.) were heated under reflux in dry butan-1-ol (12 c.c.) during 3 hr. and then cooled. The crystalline precipitate was extracted with hot ether (2 × 30 c.c.), and the extracts were evaporated *in vacuo*. The residual *methylpurine* recrystallised from ethanol as needles (0·55 g.), m. p. 151·5° (corr.) (Found: C, 60·3; H, 4·4; N, 17·4; Cl, 18·7. $C_{20}H_{17}N_5Cl_2$ requires C, 60·3; H, 4·3; N, 17·6; Cl, 17·9%), $\lambda_{max.}$ 283 mμ (log ε 4·28) with inflexions at 210 and 225 mμ.

6-*Dibenzylaminopurine* (VIII; R = R′ = R″ = H).—6-Chloropurine [23] (0·2 g.) and dibenzylamine (2·1 c.c.) were heated under reflux in dry butan-1-ol (10 c.c.) during 4 hr. The

[39] Bredereck, Hennig, Pfleiderer, and Weber, *Chem. Ber.*, 1953, **86**, 333.

yellow solution was cooled to 40°, diluted with ethanol (5 c.c.), and filtered to remove dibenzylamine hydrochloride (0·6 g.; m. p. 256°). On cooling of the filtrate at 0°, 6-*dibenzylaminopurine* separated; it crystallised from ethanol as prisms (1·1 g.), m. p. 184° (Found: C, 72·2; H, 5·25; N, 22·6. $C_{19}H_{17}N_5$ requires C, 72·35; H, 5·45; N, 22·2%). The *picrate* crystallised from ethanol as plates, m. p. 203° (Found: C, 55·1; H, 3·8; N, 20·3. $C_{25}H_{20}O_7N_8$ requires C, 55·15; H, 3·75; N, 20·6%).

6-(N-*Benzyl*-N-*methylamino*)*purine* (IX; R = H) was prepared by the method of Bullock *et al.*[27] and found to have m. p. 212·5° (corr.) (Found: N, 29·3. Calc. for $C_{13}H_{13}N_5$: N, 29·3%).

6-*Benzylaminopurine* (X; R = H).—2,8-Dichloro-6-dibenzylaminopurine (500 mg.) was stirred in liquid ammonia (30 c.c.), and a solution of sodium (300 mg.) in liquid ammonia (20 c.c.) added dropwise until the blue colour persisted. The solution was then stirred during 1 hr. Ammonium chloride (1 g.) was added and the ammonia allowed to evaporate. The residue crystallised from water, to give the purine (230 mg.), m. p. 230° (corr.) (Found: C, 63·9; H, 5·1; N, 31·1. Calc. for $C_{12}H_{11}N_5$: C, 64·0; H, 4·9; N, 31·1%). A sample prepared by the method of Skinner and Shive [26] had m. p. 229—230° and did not depress the m. p. of the above product.

4-*Amino*-6-*dibenzylamino*-2-*methylthiopyrimidine*.—4-Amino-6-chloro-2-methylthiopyrimidine [32] (2 g.) and dibenzylamine (5 c.c.) were heated at 180° during 4 hr. in a sealed tube which was then cooled and opened. The solid mass was extracted with hot benzene (2 × 15 c.c.), and the combined extracts were washed with water (2 × 20 c.c.) and dried (Na_2CO_3). Addition of light petroleum (100 c.c.; b. p. 60—80°) precipitated a yellow *amine* which crystallised from ethyl acetate and from ethanol as colourless prisms (2·2 g.), m. p. 138° (Found: C, 67·6; H, 5·6. $C_{19}H_{20}N_4S$ requires C, 67·4; H, 5·95%).

6-*Dibenzylamino*-2-*methylthiopurine*.—The above pyrimidine (5 g.) in 50% aqueous acetic acid (100 c.c.) containing concentrated hydrochloric acid (1 c.c.) was stirred at −5° while a solution of sodium nitrite (1·25 g.) in water (10 c.c.) was added dropwise. After 2 hr. the solution was diluted with water (200 c.c.) and extracted with chloroform (2 × 30 c.c.). After drying ($MgSO_4$), the extracts were evaporated and the nitroso-compound was extracted with light petroleum (2 × 50 c.c.; b. p. 60—80°) to give a green oil (2·4 g.) on evaporation. This was dissolved in warm acetone (30 c.c.) and reduced with sodium dithionite (2 g.) in water (25 c.c.). After 3 min., the colourless solution was diluted with water (50 c.c.) and extracted with chloroform (3 × 10 c.c.). The extracts were dried and evaporated, to give the crude amine (1·0 g.). This was heated in formic acid (10 c.c.) at 100° during 1 hr. and poured into water. The formamido-compound was separated and obtained from methanol–light petroleum as a white solid (120 mg.), m. p. 204° (decomp.), $\lambda_{max.}$ 245 and 269 mμ (log ε 4·56 and 4·05). Further dilution of the mother-liquor with light petroleum gave a white solid, m. p. 130—140°, which was not investigated further. The formamido-compound was fused at 250° during 15 min.; the *product* (40 mg.), crystallised from ethanol (charcoal), had m. p. 206·5° (corr.) (Found: C, 67·0; H, 5·7; S, 8·6. $C_{20}H_{19}N_5S$ requires C, 66·5; H, 5·3; S, 8·85%).

8-*Mercapto*-2-*methylthioadenine*.—4,5,6-Triaminopyrimidine [32] (1·7 g.) in pyridine (15 c.c.) was warmed with carbon disulphide (2 c.c.) at 60° during 30 min. The excess of carbon disulphide was then allowed to evaporate, the solution heated under reflux at 115° during 10 min., and most of the pyridine then evaporated *in vacuo*. The yellow *mercaptopurine* was triturated with hot ethanol and, crystallised from pyridine, had m. p. 305° (decomp.) (1·9 g.) (Found: C, 33·8; H, 2·95; S, 30·6. $C_6H_7N_5S_2$ requires C, 33·8; H, 3·3; S, 30·0%).

2,8-*Dimethylthioadenine* (XI; R = H).—The above mercaptopurine (1·9 g.) was dissolved in an equivalent amount of N-sodium hydroxide solution (9 c.c.) and stirred while dimethyl sulphate (0·85 c.c.) was added dropwise. After 10 min., the solid was separated, washed with aqueous ethanol, and dried. The *product* (1·75 g.) crystallised either from ethanol as needles or from water as prisms, m. p. 254° (corr.) (Found: C, 37·3; H, 4·15; S, 27·8. $C_7H_9N_5S_2$ requires C, 37·0; H, 3·95; S, 28·2%).

9-*Methyl*-2,8-*dimethylthioadenine* (XI; R = Me).—(i) 5,6-Diamino-4-methylamino-2-methylthiopyrimidine was prepared by the method of Baddiley *et al.*[32] and crystallised from water as yellow prisms, m. p. 151°. The *picrate*, crystallised from ethanol, had m. p. 208° (decomp.) (rapid heating) or m. p. 195° (slow heating) (Found: C, 35·3; H, 3·4; S, 8·05. $C_{12}H_{14}O_7N_8S$ requires C, 34·7; H, 3·4; S, 7·7%). The pyrimidine was cyclised with carbon disulphide in pyridine in the above manner and the product methylated as above. The *methylpurine* crystallised from ethanol as rods, m. p. 235·5° (Found: C, 40·3; H, 4·75; N, 29·1; S, 26·6. $C_8H_{11}N_5S_2$ requires C, 39·85; H, 4·6; N, 29·05; S, 26·55%).

(ii) 2,8-Dimethylthioadenine (0·9 g.) and methyl iodide (0·25 c.c.) in 0·5N-ethanolic sodium hydroxide (8 c.c.) were warmed at 70° during 30 min. and the solution cooled. The product was filtered off, washed, and recrystallised from ethanol as rods (0·8 g.), m. p. 236°, undepressed on admixture with the product obtained as above (Found: C, 39·8; H, 4·5; S, 25·8%). Both preparations had the same ultraviolet spectrum, *viz.*, in EtOH $\lambda_{max.}$ 204, 230, and 290 mμ (log ε 4·20, 4·41, and 4·32 respectively) or in 0·01N-hydrochloric acid $\lambda_{max.}$ 228 and 290 mμ (log ε 4·36 and 4·31).

9-*Benzyl*-2,8-*dimethylthioadenine* (XI; R = CH_2Ph).—This *compound* was prepared by the action of benzyl bromide (0·53 c.c.) on the purine (1·0 g.) as for the 9-methyl derivative in method (ii) above and, crystallised from ethanol–benzene, had m. p. 205—206° (corr.) (Found: C, 52·8; H, 4·9; S, 20·2. $C_{14}H_{15}N_5S_2$ requires C, 53·0; H, 4·8; S, 20·2%), $\lambda_{max.}$ 208, 230, and 291 mμ (log ε 4·43, 4·41, and 4·33 respectively).

4,6-*Diamino*-5-(N′-*ethylthioureido*)-2-*methylthiopyrimidine* (XII).—A solution of 4,5,6-triamino-2-methylthiopyrimidine (2·5 g.) and ethyl isothiocyanate (1·4 c.c.) in ethanol (15 c.c.) was heated under reflux during 30 min. and cooled. The solid was separated, washed with water and ethanol, and crystallised from water to give the *product* (2·8 g.) m. p. >315° (Found: C, 37·0; H, 5·3; N, 33·0; S, 24·4. $C_8H_{14}N_6S_2$ requires C, 37·2; H, 5·45; N, 32·55; S, 24·8%).

2-*Chloro*-9-*methyl*-8-*methylthioadenine*.—2,8-Dichloro-9-methyladenine [34] (250 mg.), methanethiol (0·2 c.c.), and sodium methoxide (from 60 mg. of sodium) were heated under reflux in absolute methanol (5 c.c.) during 5 min. and then shaken at room temperature during 12 hr. The precipitate was filtered off and crystallised from ethanol as plates (150 mg.), m. p. 283° (Found: C, 36·4; H, 3·25; N, 29·6; S, 13·9; Cl, 15·6. $C_7H_8ClN_5S$ requires C, 36·55; H, 3·5; N, 30·4; S, 13·9; Cl, 15·4%), $\lambda_{max.}$ 223 and 281 mμ (log ε 4·44 and 4·34).

7-*Methyl*-2,8-*dimethylthioadenine*.—2,8-Dichloro-7-methyladenine [33] (1 g.) and a solution of potassium sulphide (1 g.) in water saturated with hydrogen sulphide were heated at 120° in a sealed tube during 6 hr. After cooling, the solution was filtered and acidified with dilute acetic acid. The white precipitate was washed, dissolved in N-sodium hydroxide (10 c.c.), and shaken with dimethyl sulphate (0·7 c.c.). After 15 min. the precipitated *sulphide* was filtered off, washed with aqueous methanol, and crystallised from methanol and then aqueous methanol as needles (0·25 g.), m. p. 261—263° (corr.) (Found: C, 39·6; H, 4·35; N, 28·9; S, 26·9. $C_8H_{11}N_5S_2$ requires C, 39·85; H, 4·6; N, 29·05; S, 26·55%), $\lambda_{max.}$ 241 and 298 mμ (log ε 4·39 and 4·04).

7-*Methyl*-2-*methylthioadenine* (XIV).—4,6-Diamino-5-formamido-2-methylthiopyrimidine [32] (0·9 g.) was introduced into refluxing anhydrous dioxan (100 c.c.) containing lithium aluminium hydride (600 mg.) by hot extraction from a thimble. After extraction was complete, the solution was heated under reflux during 1 hr. and left to cool overnight. The excess of hydride was destroyed by wet ethyl acetate (15 c.c.). Water (10 c.c.) was added and the suspension warmed to 80° and filtered from the inorganic precipitate. The brown solution was acidified with dilute sulphuric acid and evaporated to dryness *in vacuo*. The residue was dissolved in hot water (10 c.c.), filtered, and neutralised to pH 9 with alkali to precipitate 4,6-diamino-5-methylamino-2-methylthiopyrimidine, which crystallised from ethanol as brown needles (100 mg.), m. p. 170° raised to m. p. 172° after recrystallisation from hot water, $\lambda_{max.}$ in EtOH 218 and 277 mμ (log ε 4·52 and 4·00), in 0·1N-hydrochloric acid 215, 239, and 279 mμ (log ε 4·27, 4·28, and 4·03 respectively).

The methylaminopyrimidine (60 mg.) was heated under reflux in dry formamide (2 c.c.) during 1 hr. and the solution poured into water (10 c.c.) and evaporated to dryness *in vacuo*. The residue crystallised from ethanol as prisms, and then sublimed at 230°/10^{-1} mm. as prisms (30 mg), m. p. 282—283° (corr.) (Found: C, 43·3; H, 4·4; N, 34·8; S, 16·3. $C_7H_9N_5S$ requires C, 43·05; H, 4·65; N, 35·9; S, 16·4%), $\lambda_{max.}$ in H_2O 212, 237, and 286 mμ (log ε 4·19, 4·33, and 3·84 respectively), in 0·1N-HCl 246 and 289 mμ (log ε 4·31 and 4·03).

An attempt to cyclise the 5-methylaminopyrimidine with carbon disulphide in pyridine under the usual conditions resulted in 95% recovery of unchanged starting material.

2,8-*Dimercaptoadenine*.—An intimate mixture of dry thiourea (1 g.) and 4,5,6-triamino-2-mercaptopyrimidine [40] (1 g.) was fused during 1 hr. at 180°. The melt was cooled and disolved in 10% aqueous ammonia (100 c.c.), and the solution was filtered through charcoal. The *product* was precipitated by dilute acetic acid, collected, and washed. Repeated precipitation from dilute ammonia solution gave yellow crystals (0·8 g.), m. p. 320° (Found: C, 30·0; H,

[40] Traube, *Annalen*, 1904, **331** 64.

2·65; S, 31·9. $C_5H_5N_5S_2$ requires C, 30·1; H, 2·5; S, 32·2%), $\lambda_{max.}$ 311 and 363 mμ (log ε 4·34 and 4·17) in dimethylformamide.

Partial Desulphurisation of 9-Methyl-2,8-dimethylthioadenine.—The dimethylthio-compound (190 mg.) was heated with Raney nickel (matured at 100° in preparation) in boiling absolute ethanol (12 c.c.) during 1 hr. The hot solution was filtered and the nickel residue twice extracted with hot ethanol (2 × 5 c.c.). The combined extracts were evaporated to dryness with the bulk of the solution and the residue crystallised from alcohol. It had m. p. 259° undepressed on mixing with an authentic sample of 9-methyl-2-methylthioadenine,[32] $\lambda_{max.}$ 277 mμ (log ε 4·11) in 0·05N-NaOH, 270 mμ (log ε 4·09) in 0·05N-HCl.

Chloromercuripurines.—These were prepared by the general method of Davoll and Lowy,[14] by slow addition of an equivalent amount of ethanolic mercuric chloride to a stirred aqueous, aqueous-ethanolic, or ethanolic solution of the sodium derivative of the purine. The precipitated salt was collected, washed, and dried (P_2O_5 *in vacuo*), ground, and dried again. The yields of the glycosides obtained on reaction of these salts with sugar derivatives were not consistently improved by precipitation of the salt in the presence of " Diatomite " but were more dependent on the thoroughness of grinding of the dry chloromercuripurine.

Some of these salts appeared to be light-sensitive, becoming blue or green-blue after a few hours' exposure to daylight. They were not characterised but were brought into reaction immediately with the acylhalogeno-sugar in dry xylene in the usual way.[14]

The finely divided chloromercuripurine was stirred vigorously in xylene which was heated under a Dean and Stark separator until the condensate was water-free. The dry acylhalogeno-sugar (small excess) was rapidly added and stirring continued under reflux for 2 or more hours after which the mixture was filtered while hot through " Diatomite " and the residue extracted with chloroform. The xylene solution was diluted with light petroleum, and the crude precipitated product taken up in the chloroform extracts. This solution was then extracted with 10% potassium iodide solution (to remove mercury salts), then with water, dried, and evaporated to dryness under reduced pressure. The crude gummy product was then purified.

6-*Chloro*-9-(*tetra*-O-*acetyl*-β-D-*glucopyranosyl*)*purine.*—6-Chloropurinylmercuric chloride [20] (1 g.) in xylene (100 c.c.) was allowed to react with acetobromoglucose (0·9 g.) during 4 hr. as described above. The crude product was dissolved in hot ethanol (10 c.c.) from which the glycoside separated on cooling. This was chromatographed on acid-washed alumina, elution with benzene–10% chloroform, and crystallisation from ethanol giving the *product* (100 mg.), m. p. 163° (Found: C, 46·1; H, 4·1; N, 11·6; Cl, 7·1. $C_{19}H_{21}ClN_4O_9$ requires C, 47·0; H, 4·35; N, 11·6; Cl, 7·3%).

9-β-D-*Glucopyranosyladenine.*[13]—The above glycoside (25 mg.) was heated in a sealed tube with ethanolic ammonia (6 c.c.) at 140° during 2 hr. The solution was evaporated to dryness and the *glucoside* crystallised twice from aqueous acetone, to give colourless crystals (7 mg.), m. p. 207° undepressed on admixture with an authentic sample of m. p. 209°, $\lambda_{max.}$ 260 mμ (log ε 4·08) in 0·1N-HCl.

2,8-*Dichloro*-6-*dibenzylamino*-9-(*tetra*-O-*acetyl*-β-D-*glucopyranosyl*)*purine* [VIII; R = R″ = Cl, R′ = $C_6H_7O(OAc)_4$].—2,8-Dichloro-chloromercuri-6-dibenzylaminopurine (1·2 g.) and acetobromoglucose (1·2 g.) reacted in refluxing xylene (50 c.c.) during 3 hr. The *glycoside* crystallised from ethanol as needles (200 mg.), m. p. 208°, $[\alpha]_D^{22}$ −51° (*c* 1·5 in $CHCl_3$) (Found: C, 55·1; H, 4·8; N, 9·75; Cl, 10·7. $C_{37}H_{33}Cl_2N_5O_9$ requires C, 55·4; H, 4·65; N, 9·8; Cl, 10·0%), $\lambda_{max.}$ 284 mμ (log ε 4·40), infl. 210 and 225 mμ (log ε 4·47 and 4·37 respectively).

6-*Dibenzylamino*-9-(*tetra*-O-*acetyl*-β-D-*glucopyranosyl*)*purine* [VIII; R = R″ = H, R′ = $C_6H_7O(OAc)_4$].—Chloromercuri-6-dibenzylaminopurine (2·6 g.) reacted with acetobromoglucose (3 g.) in dry boiling xylene (80 c.c.) during 3½ hr. The crude product crystallised from ethanol to give the *glycoside* (1·25 g.). A portion (100 mg.) was chromatographed on acid-washed alumina, and eluted with benzene–10% chloroform as a single band, fluorescent in ultraviolet light. On evaporation and crystallisation from ethanol this fraction gave the product (40 mg.), m. p. 162° (corr.), $[\alpha]_D^{21}$ −66° (*c* 1 in $CHCl_3$) (Found: C, 61·4; H, 5·6. $C_{33}H_{35}N_5O_9$ requires C, 61·4; H, 5·45%), $\lambda_{max.}$ 278 mμ (log ε 4·36).

6-*Benzylamino*-9-β-D-*glucopyranosylpurine* [VIII; R = R″ = H, R′ = $C_6H_7O(OH)_4$].—(i) 6-Dibenzylamino-9-(tetra-*O*-acetyl)-β-D-glucopyranosylpurine (520 mg.) was stirred in solution in liquid ammonia (40 c.c.) while a solution of sodium (300 mg.) in liquid ammonia (30 c.c.) was added dropwise until the blue colour persisted, then the mixture was stirred for a

further hour. Ammonium chloride (500 mg.) was added and the solvent allowed to evaporate. The residue was washed with ether (2 × 10 c.c.) and extracted with hot ethanol (2 × 15 c.c.). Evaporation of the extract gave the *glucoside*, which crystallised from ethanol as needles (300 mg.), m. p. 188—190° (corr.), solidifying and remelting at 211° (Found: C, 55·8; H, 5·2; N, 17·8. $C_{18}H_{21}N_5O_5$ requires C, 55·8; H, 5·45; N, 18·1%).

(ii) By a similar method, 2,8-dichloro-6-dibenzylamino-9-(tetra-*O*-acetyl)-β-D-glucopyranosylpurine (120 mg.) was reduced to the same glucoside (50 mg.), m. p. 188—190° (corr.).

6-(N-*Benzyl*-N-*methylamino*)-7-(*tetra*-O-*acetyl*-β-D-*glucopyranosyl*)*purine* [IX; R = $C_6H_7O(OAc)_4$].—In this case, the chloromercuripurine was prepared by the slow addition of the sodium hydroxide solution to a stirred suspension of 6-(*N*-benzyl-*N*-methylamino)purine in a warm ethanolic solution of the equivalent amount of mercuric chloride. The mercuri-compound (630 mg.) and acetobromoglucose (750 mg.) were heated during 4 hr. in dry xylene (50 c.c.). The crude product was chromatographed on alumina (Spence grade " H ") and developed with chloroform, the eluted fractions (of 10 c.c.) being examined for ultraviolet absorption.

The first eluted fractions had $\lambda_{max.}$ 305 mμ and were combined and evaporated. Crystallisation from ethanol gave the 7-*glycoside* (50 mg.), m. p. 127°, $[\alpha]_D^{21}$ −15° (*c* 0·55 in $CHCl_3$) (Found: C, 56·5; H, 5·45; N, 12·0. $C_{27}H_{31}N_5O_9$ requires C, 56·9; H, 5·5; N, 12·3%), $\lambda_{max.}$ 223 and 305 mμ (log ε 4·20 and 4·21 respectively). The next fractions showed a gradual change of maximum absorption to 280 mμ. Evaporation of these fractions and crystallisation of the residue from ethanol gave the 9-isomer contaminated with the 7-isomer as shown by the shoulder at the longer wavelength. It had m. p. 105°; $\lambda_{max.}$ 277 mμ (log ε 4·11), infl. 302 mμ. When this material was rechromatographed, the shoulder persisted through all fractions.

6-*Benzylamino*-9-(*tri*-O-*acetyl*-β-D-*ribopyranosyl*)*purine* [X; R = $C_5H_6O(OAc)_3$].—6-Benzylaminochloromercuripurine (1·5 g.) and tri-*O*-acetylribopyranosyl chloride (from 1·5 g. of tetra-*O*-acetyl-D-ribopyranose [29]) in dry xylene (100 c.c.) were heated during 3 hr. and the crude product isolated in the usual way. This was chromatographed on acid-washed alumina and developed with chloroform. All the ultraviolet-fluorescent fractions eluted had maximum absorption at 271 mμ. These were combined and evaporated to dryness. The *glycoside* (0·82 g.) thus obtained formed an amorphous solid from methanol–ether. A sample was twice rechromatographed for analysis (Found: C, 55·5; H, 5·2; N, 13·7. $C_{23}H_{25}N_5O_7,CH_3{\cdot}OH$ requires C, 55·9; H, 5·65; N, 13·6%), $\lambda_{max.}$ 208 and 271 mμ (log ε 4·36 and 4·19 respectively).

A sample was deacetylated with sodium ethoxide in boiling ethanol to give 6-*benzylamino*-9-β-D-*ribopyranosylpurine*, which crystallised from water as prisms, m. p. 123—124° (corr.) (Found: C, 55·9; H, 5·75; N, 19·0. $C_{17}H_{19}N_5O_4,\frac{1}{2}H_2O$ requires C, 55·75; H, 5·5; N, 19·15%), $\lambda_{max.}$ 209 and 268 mμ (log ε 4·36 and 4·25) in MeOH.

Reaction of 6-(N-*Benzyl*-N-*methylamino*)*purine and Tri*-O-*benzoylribofuranosyl Bromide.*—The chloromercuripurine (above) (0·75 g.) reacted with tri-*O*-benzoylribofuranosyl bromide (from 1·4 g. of 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-D-ribofuranose [41]) in boiling xylene (70 c.c.) during 3 hr. and the crude product was isolated in the usual way. This was chromatographed on alumina (Spence grade " H "), with elution by chloroform–benzene; the ultraviolet-fluorescent eluates were examined for light absorption. All such fractions had $\lambda_{max.}$ 277 mμ. These were combined and evaporated to a colourless glass (0·8 g.).

An attempt to reduce the glycoside with sodium in liquid ammonia appeared to decompose the compound.

2,8-*Dimethylthio*-9-β-D-*glucopyranosyladenine* (XI; R = $C_6H_{11}O_5$).—Chloromercuri-2,8-dimethylthioadenine (1·4 g.) was treated with acetobromoglucose (1·5 g.) in refluxing dry xylene (40 c.c.) during 1 hr. The crude glycoside, obtained as a gum, was deacetylated in methanolic ammonia during 2 days at 10°. The product was obtained as a pale yellow powder (from ethanol) (0·5 g.), m. p. 184°, $\lambda_{max.}$ 231 and 290 mμ (log ε 4·31 and 4·28 respectively). The picrate was obtained as plates (from ethanol), m. p. 188° (decomp.) (Found: C, 36·9; H, 3·75; N, 18·1. $C_{19}H_{22}N_8O_{12}S_5$ requires C, 36·65; H, 3·55; N, 18·0%).

2,8-*Dimethylthio*-9-(*tri*-O-*benzoyl*-β-D-*ribofuranosyl*)*adenine* [XI; R = $C_5H_6O(OBz)_3$].—The above chloromercuripurine (2·1 g.) was treated in dry toluene (150 c.c.) with 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-D-ribofuranose (1·85 g., converted into the 1-chloro-compound). The crude product was a yellowish amorphous solid (2·9 g.) after separation from ethanol. The product was washed with cold benzene and crystallised from ethanol–10% ethyl acetate to give the *riboside* as needles (1·1 g.), m. p. 210° (corr.), $[\alpha]_D^{23}$ +22·3° (*c* 1·04 in $CHCl_3$) (Found: C, 58·9;

[41] Kissman, Pidacks, and Baker, *J. Amer. Chem. Soc.*, 1955, **77**, 18.

H, 4·2; N, 10·4; S, 9·95. $C_{33}H_{29}O_7S_5$ requires C, 59·0; H, 4·35; N, 10·4; S, 9·55%), $\lambda_{max.}$ 234 and 292 mμ (log ε 4·77 and 4·35).

The benzene washings were chromatographed on acid-washed alumina and developed with benzene, to give, first, 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-D-ribofuranose (90 mg.), m. p. 125° undepressed on admixture with an authentic sample of m. p. 126°, and then the above riboside (400 mg.) as needles, m. p. 208° (from benzene).

2,8-*Dimethylthio*-9-(*tri*-O-*acetyl*-β-D-*ribofuranosyl*)*adenine* [XI; R = $C_5H_6O(OAc)_3$].—The chloromercuriadenine (3·2 g.) reacted in dry xylene (150 c.c.) with tri-*O*-acetyl-D-ribofuranosyl chloride (from 3·4 g. of the tetra-acetate,[29] m. p. 77—80°) during 2½ hr. Unchanged 2,8-dimethylthioadenine (270 mg.) was extracted with hot ethanol from the chloroform-insoluble reaction residue. The product, crystallised from methanol (yield 1·47 g.), had m. p. 125—130°. This had a broad ultraviolet absorption maximum from 280 to 300 mμ. The methanolic mother-liquors were evaporated to dryness and the residue combined with the crude product and chromatographed on an acid-washed alumina column (30 × 4 cm.) and developed with benzene–10% chloroform. Fractions of 15 c.c. were collected and examined for ultraviolet absorption in the region of 285—305 mμ. Fractions 1—12 showed practically no absorption in this region. Fractions 13—16 had $\lambda_{max.}$ 297 mμ and were combined (eluate A). Fractions 17—28 had $\lambda_{max.}$ 292 mμ and were combined (eluate B). Further fractions had negligible light absorption.

Eluate A was evaporated *in vacuo* and fractionally crystallised from propan-2-ol to give tetra-*O*-acetyl-D-ribofuranose (200 mg.), m. p. and mixed m. p. 81°. The mother-liquors were evaporated and the residue was chromatographed in benzene on an acid-washed alumina column (12 × 1·5 cm.). The eluate containing the ultraviolet-fluorescent band was evaporated to a pale yellow glass (80 mg.), $\lambda_{max.}$ 203, 232, and 297 mμ (log ε 4·17, 4·40, and 4·29 respectively, based on $C_{18}H_{23}N_5O_7S_2$). This will hereafter be referred to as the 7-isomer.

Eluate B was evaporated and recrystallised from methanol to give the 9-riboside (1·1 g.), m. p. 167° (corr.). A sample was rechromatographed and then crystallised from aqueous methanol, whereafter it had m. p. 178° (Found: C, 42·1; H, 5·5; N, 13·4; Loss, 5·75. $C_{18}H_{23}N_5O_7S_2,1\frac{1}{2}H_2O$ requires C, 42·2; H, 5·1; N, 13·65; Loss, 5·3. In a sample dried at 110° *in vacuo*: S, 13·3. $C_{18}H_{23}N_5O_7S_2$ requires S, 13·3%).

Distribution coefficients were measured spectroscopically for the system benzene–methanol–10% water: 9-isomer, D = 1·25; 7-isomer, D = 1·10.

2,8-*Dimethylthio*-9-β-D-*ribofuranosyladenine* [XI; R = $C_5H_6O(OH)_3$].—(i) The above 2,8-dimethylthio-9-(tri-*O*-benzoyl)-β-D-ribofuranosyladenine (0·85 g.) was debenzoylated in boiling methanol containing sodium methoxide (from 0·05 g. of sodium) during 45 min. The *product* crystallised from ethanol in rods (0·35 g.), m. p. 230° (corr.) (Found: C, 41·3; H, 5·25; N, 18·6; S, 16·4. $C_{12}H_{17}N_5O_4S_2,\frac{1}{2}C_2H_5{\cdot}OH$ requires C, 40·9; H, 5·25; N, 18·35; S, 16·7%), $\lambda_{max.}$ 289 mμ (log ε 4·31) in H_2O.

(ii) By the same method, the 2,8-dimethylthio-9-(tri-*O*-acetyl-β-D-ribofuranosyl)adenine (0·8 g.) gave the same riboside (0·2 g.), m. p. and mixed m. p. 230° (from ethanol–chloroform).

Adenosine.—2,8-Dimethylthio-9-(tri-*O*-benzoyl-β-D-ribofuranosyl)adenine was desulphurised by stirring it with Raney nickel (*ca.* 1·5 g.) in refluxing ethanol (30 c.c.)–ethyl acetate (10 c.c.). After ½ hr., more Raney nickel (0·5 g.) was added. After a further ½ hr. the hot solution was filtered and the nickel extracted with hot ethanol (2 × 20 c.c.). The combined filtrate and extracts were evaporated to dryness under reduced pressure and debenzoylated (as above) with sodium methoxide. The crude product crystallised from water (2 c.c.) to give adenosine as needles (49 mg.), m. p. 228°, mixed m. p. 227°. The picrate had m. p. and mixed m. p. 194° (Found: C, 39·1; H, 3·45. Calc. for $C_{16}H_{20}N_8O_{11}$: C, 39·2; H, 3·2%). The product had $\lambda_{max.}$ 207 and 260 mμ (log ε 4·27 and 4·13) in H_2O, 258 mμ (log ε 4·12) in 0·1N-HCl.

Desulphurisation and Hydrolysis of 2,8-*Dimethylthio*-7-(*tri*-O-*acetyl*-β-D-*ribofuranosyl*)*adenine.*—The 7-isomer (15 mg.) was heated under reflux in ethyl acetate (1 c.c.) and ethanol (1 c.c.), and Raney nickel (200 mg.) was added in portions during 15 min. The reaction was continued until the ultraviolet absorption maximum of the solution was constant at 273 mμ. The solution was cooled, filtered, and evaporated to dryness. The residue was dissolved in water (2 c.c.) (pH 8; B.D.H. indicator paper). It had $\lambda_{max.}$ 272 mμ, infl. 286 mμ. 2N-Hydrochloric acid (8 c.c.) was added and the solution heated under reflux during 1 hr. The solution then had $\lambda_{max.}$ 264 mμ. The solution was concentrated to 4 c.c. after neutralisation with dilute ammonia solution, "spotted" on Whatman No. 1 paper, and chromatographed in butanol–acetic acid–water

(5 : 1 : 4) against a control spot of D-ribose. When the dried paper was developed with aniline hydrogen phthalate,[42] both the test and the control gave red-brown spots at R_F 0·31.

9-*Benzyloxycarbonyl*-2-*methylthioadenine* (XVI).—2-Methylthioadenine (3·5 g.) was dissolved in warm water (20 c.c.) and N-sodium hydroxide (19 c.c.). Benzyl chloroformate (19 c.c. of a 20% dioxan solution) was added dropwise with stirring. The white precipitate was filtered off, washed, and rapidly crystallised from methanol–10% chloroform to give the *product* (1·4 g.), m. p. 172° (decomp.) (Found: C, 53·4; H, 4·25; N, 21·6. $C_{14}H_{13}N_5O_2S_2$ requires C, 53·3; H, 4·15; N, 22·2%), $\lambda_{max.}$ 235 and 277 mμ (log ε 4·21 and 4·11) in $CHCl_3$.

A suspension of this compound (1·4 g.) in dry acetone (15 c.c.) containing 2,3,5-tri-*O*-benzoylribosyl chloride (from 2 g. of the 1-*O*-acetylriboside [29]) was stirred under reflux during 24 hr. and filtered. The solid residue contained 2-methylthioadenine (0·53 g.), m. p. 287° (from ethanol). The acetone filtrate was evaporated under reduced pressure to 15 c.c. and cooled. A precipitate of 9-benzyloxycarbonyl-2-methylthioadenine (0·4 g.), m. p. 168°, was obtained. The acetone filtrate was evaporated to dryness *in vacuo* and the residue crystallised from dry benzene (10 c.c.) as needles (300 mg.), m. p. 139—140°, $[\alpha]_D^{23}$ +80·8° (*c* 1 in $CHCl_3$) (1,3,5-tri-*O*-benzoyl-D-ribose [38] has m. p. 142—143°, $[\alpha]_D^{22}$ +83·5°). It recrystallised from aqueous acetone as needles, m. p. 141·5—142·5° (corr.) (Found: C, 67·2; H, 4·65. Calc. for $C_{26}H_{22}O_8$: C, 67·5; H, 4·8%).

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[42] Partridge, *Nature*, 1949, **164**, 443.