Nucleosides Part XI (1): *O*- → *N*-Glycosyl Rearrangement of Theobromine-*O*-Glucoside to Theobromine-1-Glucoside

J. A. Elvidge, G. T. Rogers and T. L. V. Ulbricht (2)

Department of Chemistry, University of Surrey

and

Twyford Laboratories Ltd., Twyford Abbey Road,
London N. W. 10, England

Received February 25, 1971

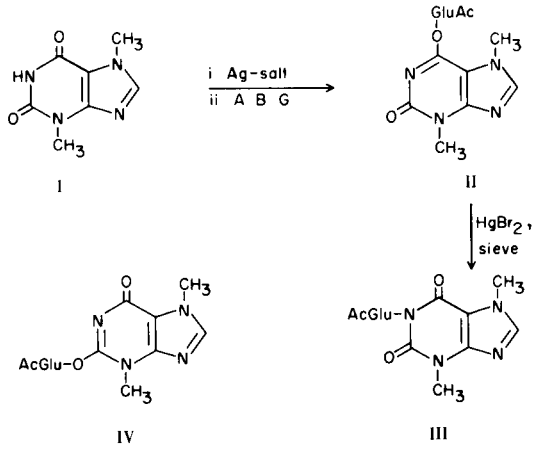
O- → *N*-Glycosyl rearrangements in heterocyclic derivatives have been studied extensively in recent years (3-10) and evidence presented for the existence of two alternative mechanisms (5,6): 1) an acid-catalysed reaction and 2) a mercuric bromide-catalysed inter-molecular reaction (favoured by non-polar solvents). An *O*- → *N*-glycosyl rearrangement in a purine derivative has not previously been reported and we now describe as the first example the rearrangement of the labile theobromine-*O*-glucoside (II).

Theobromine-*O*-glucoside was first prepared by Fischer (11) and later by Ballou and Link (12) by treating theobromine silver salt with acetobromoglucose in boiling toluene. The glycosidic bond in this product was found to be very labile to cleavage by acid or alkali, showing it to be an *O*-glucoside. The exact position of the glycosidic bond, however, was not established (enolisation is possible at either C-2 or C-6). Treatment of theobromine-*O*-glucoside with barium methoxide in methanol (12) gives a monomethyltheobromine which contains a methoxy group and, as expected, imidoester rearrangement (13) of this methoxypurine gives caffeine.

We prepared theobromine-*O*-glucoside as described by Ballou and Link (12) and obtained a crystalline product from ethyl acetate-*n*-hexane. The product was resolved into two spots by prolonged thin layer chromatography (tlc) (ethyl acetate) but the components could not be isolated by preparative tlc.

There are two possible structures for the *O*-glucoside (II and IV). That structure (II) is correct was shown by ^{1}H nmr spectroscopy, which showed the *O*-glucoside to be a mixture of the α and β forms of O_6-(2′,3′,4′,6′-tetra-*O*-acetylglucopyranosyl) theobromine, with the β anomer predominating. The results are presented in Table I.

Theobromine gives a sharp spectrum and the assignments of the 3-N-Me, 7-N-Me and 8-H are supported by the spectrum of caffeine (Varian Catalog 1, No. 204) in which there are two high field methyl signals and one at lower field. The spectrum for theobromine-*O*-glucoside (II) shows signals at τ 6.23 and 6.04 for the 7-N-Me groups



in the β and α derivatives respectively. The fact that the 3-N-Me group is not affected by the glycoside centre and shows up as a single signal at τ 6.57 definitely establishes the structure of the *O*-glucoside as the O_6-glucoside derivative (II) and not the O_2-glucoside (IV). The existence of α and β forms (β form predominating 5:3) was established by the signals for the anomeric protons which are centred at τ 3.57 (1′ -Hβ, $J_{1'2'}$ = 7.7 Hz) and at 3.32 (1′ -Hα, $J_{1'2'}$ = 3.1 Hz) (14). Moreover the signals for the 8-H proton were also resolved for the α and β forms: see Table I.

The formation of both anomers of theobromine-*O*-glucoside in this reaction is another exception to Baker's rule (15,16) which states that both 1,2-*cis* and *trans* glycosyl halides give a 1,2-*trans* nucleoside by a single or double S_N2 reaction respectively, when condensed with a heavy metal salt of a purine. According to this rule only the β-anomer of theobromine-*O*-glucoside should be formed. The formation of an α-anomer in the condensation of the heavy metal salt of a purine with a crystalline 1,2-*trans* glycosyl halide was reported for the first time recently (16,17).

On treatment of the *O*-glucoside (II) in toluene with an equivalent of mercuric bromide, cleavage of the glycosidic

TABLE I

^{1}H nmr Data for Theobromine Derivatives Measured in D_6-Dimethyl Sulphoxide

	3 Me (τ)	7 Me	8-H	1′-H	$J_{1'2'}$ (Hz)
Theobromine	6.68	6.17	2.04		
O-Glucoside II	6.57	6.04 (α)	1.85 (α)	3.32 (α)	3.1 (α)
		6.23 (β)	1.89 (β)	3.57 (β)	7.7 (β)
N-Glucoside III	6.70	6.17	2.03	3.28 (α)	
				3.58 (β)	

TABLE II

Ultraviolet Spectral Data of Theobromine Derivatives (95% Ethanol)

	λ max (mμ)	ϵ max x 10^{-4}	λ min (mμ)
Theobromine	275	0.93	245
Caffeine (8)	275	0.87	245
Theobromine N_1-glucoside (III)	274	0.78	247
Theobromine-*O*-glucoside (II)	247	0.41	237
	296	0.70	260
Monomethyl theobromine (8)	245	0.36	235
(methoxy derivative)	295	0.85	260

bond occurs and theobromine can be recovered quantitatively. When the amount of mercuric bromide is restricted to 0.1 equivalents, theobromine-*O*-glucoside (II) partly rearranges to the N_1-isomer, but extensive cleavage still occurs.

It is likely that this cleavage takes place by an intermolecular reaction with mercuric bromide (mechanism 2), since when the reaction mixture is diluted 4-fold unchanged *O*-glucoside can be recovered quantitatively (10). Such a mechanism would involve the initial formation of acetobromoglucose (8,9) which in the presence of traces of water would give rise to acidic conditions and cleavage of the sensitive *O*-glucoside. Extensive cleavage of the corresponding *O*-glycosidic bond is also observed in O_2,O_4-bis-(2′,3′,4′,6′-tetra-*O*-acetyl-β-D-glucopyranosyl) uracil when it is treated with mercuric bromide (10), although the simultaneous O_2- $\rightarrow$ N_1-glycosyl rearrangement in this compound appears to be quantitative.

Further experiments with theobromine-*O*-glucoside showed that cleavage of the glycosidic bond can be largely prevented if traces of water and acid are rigorously removed from the reaction mixture with molecular sieve. When this is done the *O*-glucoside (II) can be converted to an N_1-isomer in boiling toluene containing an equivalent of mercuric bromide, in 17% yield.

The ultra-violet spectra of theobromine and its glucosides are listed in Table II. The spectra of the *N*-glucoside and caffeine are virtually identical, indicating that the product is the N_1-glycosyl derivative (III).

The ^{1}H nmr spectrum of the product is consistent with this structure. The methyl and ring proton shifts are very like those in theobromine (see Table I), in accord with structure (III) where the *N*-tetraacetylglucosidyl group is well removed from those groups. The signals for the anomeric protons centred at τ = 3.28 and 3.58 indicate the N_1-glucosidyl derivative also to be a mixture of the α and β forms.

When the mercury salt of theobromine was heated with acetobromoglucose in a mixture of toluene and dimethylacetamide, it gradually went in solution, and then a new precipitate appeared. This was shown to be theobromine, which was quantitatively recovered, suggesting that the mercury salt reacts initially to give the *O*-glucoside, which is cleaved, since mercuric bromide is also formed; no theobromine N_1-glucoside (III) could be detected in this experiment. Attempts to prepare the N_1-glucoside by direct reaction of theobromine with acetobromoglucose in the presence of mercuric cyanide (6,19) gave only traces (< 2%) of the desired product.

EXPERIMENTAL

Ultraviolet Spectra, in 95% ethanol, were recorded on a Hilger Watts 'Ultrascan' spectrophotometer, and ir spectra in methylene chloride, were obtained with a Perkin Elmer 237 grating spectro-

photometer. ^{1}H nmr spectra were obtained using a Perkin Elmer R.10 spectrometer operating at 60 m Hz and with tetramethyl silane as internal standard. Thin layer chromatography (tlc) was carried out on Merck silica gel 254. Toluene was dried by azeotropic distillation before use and mercuric bromide crystallised from dry toluene. Molecular sieve refers to BDH powder type 3A. Preparation of the silver salt of theobromine and its reaction with acetobromoglucose were carried out in the dark.

O_6-(2′,3′,4′,6′-Tetra-*O*-acetyl-D-glucopyranosyl)theobromine (II).

The silver salt of theobromine (8.0 g.) was condensed with acetobromoglucose (11.5 g.) in toluene (130) ml.) as described by Fischer (11). The toluene solution of the product was added to light petroleum (b.p. 60-80°) (800 ml.), and the pale cream solid obtained (7.0 g., 50%) washed with light petroleum, dissolved in boiling ethyl acetate and treated with *n*-hexane until a precipitate started to appear. Sufficient ethyl acetate was added to redissolve the precipitate and the solution left to crystallise (3.4 g., m.p. 176-178°). Recrystallisation from ethyl acetate gave a product m.p. 186-187°; ν max 1750 cm^{-1} (CO ester); λ max 247 mμ (ϵ = 0.41 x 10^{-4}): λ min. 237, 260 mμ, tlc in ethyl acetate, R_F 0.06 (bright blue in U.V. light). This product was shown to consist of a mixture of the α and β anomers (3:5) by ^{1}H nmr spectroscopy (see Table I).

Anal. Calcd. for $C_{21}H_{26}N_4O_{11}$: C, 49.3; H, 5.1; N, 11.0. Found: C, 49.1; H, 5.0; N, 10.8.

Attempted Rearrangement of Compound (II).

a)

In the Absence of Molecular Sieve.

i) The *O*-glucoside (II) (200 mg.) was added to a dried solution of mercuric bromide (144 mg., 1 Eq.) in toluene (8 ml.) and the mixture heated under reflux for 60 minutes after which time a white precipitate appeared. This was filtered, washed with chloroform and shown to be theobromine (50 mg.). Starting material could not be detected in the filtrate by tlc. The filtrate was evaporated *in vacuo*, chloroform added and the suspension shaken with potassium iodide solution (2 x 5 ml., 30%) when a further precipitate of theobromine (20 mg.) separated. (Quantitative recovery.)

ii) A similar result was obtained when the above reaction was carried out for 30 minutes at 80°.

iii) The *O*-glucoside (II) (200 mg.) was added to a dried solution of mercuric bromide (15 mg., 0.1 Eq.) in toluene (8 ml.) and the mixture heated under reflux for 3 hours. No theobromine-*O*-glucoside could be detected after this time. Theobromine was precipitated from the reaction mixture and also from the filtrate after evaporation, dissolution of the residue in chloroform and washing with potassium iodide solution (total recovery 55 mg., 80%). Tlc of the chloroform soluble components showed the presence of a trace of the N_1-glucoside (III) (R_F 0.03 in ethyl acetate.)

b) In the Presence of Molecular Sieve.

i) The *O*-glucoside (II) (200 mg.) was added to a solution of mercuric bromide (144 mg., 1 Eq.) in toluene (8 ml.) containing molecular sieve (1 g.). The suspension was heated under reflux for three hours, filtered and evaporated *in vacuo*. Tlc (ethyl acetate) on the solid obtained showed the presence of the N_1-glucoside, unchanged *O*-glucoside and a trace of theobromine. The residue was treated with chloroform (25 ml.) washed with potassium iodide solution, dried and evaporated. The residue obtained was treated with aqueous ethanol, filtered and left at 0°. Theobromine N_1-glucoside tetraacetate (III) was obtained as a white amorphous solid m.p. 300° (34 mg., 17%). Tlc (ethyl acetate) showed a main spot R_F 0.03 accompanied by a very weak spot corresponding to theobromine R_F 0.1. Recrystallisation from aqueous ethanol gave N_1-(2′,3′,4′,6′-tetra-O-acetylglucopyranosyl) theobromine, m.p. 300°; λ max 274 mμ (ϵ = 0.78 x 10^{-4}), λ min. 247 mμ.

Anal. Calcd. for $C_{21}H_{26}N_4O_{11}$: C, 49.4; H, 5.1; N, 11.0. Found: C, 49.4; H, 5.1; N, 11.0.

ii) When the above experiment was repeated using 0.1 equivlents of mercuric bromide, only a trace of the N_1-isomer could be isolated. Tlc on the reaction mixture showed that most of the glucoside was unchanged after three hours.

Mercury Salt of Theobromine.

Theobromine (720 mg.) was dissolved at 60° in water (150 ml.) containing *N* sodium hydroxide (4 ml.). Mercuric chloride (1.08 g., 1 Eq.) dissolved in ethanol (10 ml.) was slowly added and the white suspension heated to 70° for 30 minutes. The product was filtered, washed with water, ethanol, ether and dried (1.3 g.).

Reaction of Theobromine Mercury Salt with Acetobromoglucose.

The mercury salt of theobromine (650 mg.) was suspended in a mixture of toluene (15 ml.) and dimethylacetamide (15 ml.) and about 1 ml. of solvent was distilled. Acetobromoglucose (822 mg., 1 Eq.) was then added to the stirred suspension and the mixture heated under reflux. After 5 minutes the mercury salt went into solution and soon afterwards a white solid appeared. The heating was continued for two hours. The solid was collected and shown to be theobromine (340 mg.). The filtrate was evaporated *in vacuo* and treated with potassium iodide solution as described above. Tlc (ethyl acetate) of the residue obtained after evaporation of the organic layer only showed the presence of traces of theobromine; no N_1-glucoside could be detected.

REFERENCES

(1) G. T. Rogers and T. L. V. Ulbricht, Part X, *J. Chem. Soc.* (C), 508 (1970).

(2) To whom enquiries should be made at: Agricultural Research Council, 160 Great Portland Street, London W1N 6DT, England.

(3) G. Wagner, *Z. Chem.*, 6, 367 (1966).

(4) K. S. Kirby and T. L. V. Ulbricht, *Ann. Rep. Prog. Chem.*, 63, 537 (1966).

(5) D. Thacker and T. L. V. Ulbricht, *J. Chem. Soc.* (C), 333 (1968).

(6) G. T. Rogers and T. L. V. Ulbricht, *ibid.*, (C), 2450 (1969).

(7) T. L. V. Ulbricht and G. T. Rogers, *ibid.*, 6130 (1965).

(8) T. L. V. Ulbricht, *Proc. Chem. Soc.* (London), 298 (1962).

(9) T. L. V. Ulbricht and G. T. Rogers, *J. Chem. Soc.*, 6125 (1965).

(10) G. T. Rogers, R. S. Shadbolt and T. L. V. Ulbricht, *ibid.*, (C), 209 (1969).

(11) E. Fischer and B. Helferich, *Ber.*, 47, 210 (1914).

(12) C. E. Ballou and K. P. Link, *J. Am. Chem. Soc.*, 71, 3743 (1949).

(13) G. E. Hilbert and T. B. Johnson, *ibid.*, 52, 2001 (1930).

(14) R. V. Lemieux and J. D. Stevens, *Can. J. Chem.*, 43, 2059

(1965).

(15) B. R. Baker in "Ciba Foundation Symposium on the Chemistry and Biology of Purines", ed. G. E. W. Wolstenholme and C. M. O'Connor, Churchill, London 1957.

(16) T. L. V. Ulbricht, "Introduction to Nucleic Acids", Oldbourne Press, 1966, p. 32.

(17) G. T. Rogers and T. L. V. Ulbricht, *Tetrahedron Letters*, 1025 (1968).

(18) G. T. Rogers and T. L. V. Ulbricht, *J. Chem. Soc.* (C), 1929 (1968).

(19) G. T. Rogers and T. L. V. Ulbricht, *Chem. Commun.*, 508 (1969).