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The Synthesis of 2-Bromoadenosine (1)

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Adenosine diphosphate causes the aggregation of human blood platelets suspended in plasma containing citrate (2). Adenosine was found to prevent this aggregation if added to the plasma before the addition of adenosine diphosphate (3). In an extension of this work, a number of purine nucleosides were evaluated as blood platelet aggregation inhibitors. None of these compounds approached the effectiveness of adenosine itself (4). More recently, however, 2-chloroadenosine (5) was found to be more inhibitory than adenosine (5). From results obtained thus far it appears that a high degree of structural specificity is associated with this inhibitory activity (4-6). Only two 2-substituted adenosines have shown significant activity. Changes in the sugar moiety or the 6-amino group result in complete loss of activity (6).

These results led us to improve the synthesis of 2-chloroadenosine (8) and to use our improved procedure for the synthesis of 2-bromoadenosine for evaluation as a blood platelet aggregation inhibitor. Fusion of 2,6-dichloropurine *in vacuo* with tetra-*O*-acetyl-β-D-ribofuranose (9) gave crystalline 9-(tri-*O*-acetyl-β-D-ribofuranosyl)-2,6-dichloropurine, which was converted to 2-chloroadenosine by treatment with methanolic ammonia at room temperature. The same general procedure applied to 2,6-dibromopurine (10) gave 2-bromoadenosine in satisfactory yield. The spectral and chromatographic characteristics of 2-bromoadenosine are very similar to those of 2-chloroadenosine (see Experimental). The ultraviolet spectra of both 2,6-dichloro- and 2,6-dibromo-9-(tri-*O*-acetyl-β-D-ribofuranosyl)purine in 0.1 *N* sodium hydroxide indicate that both these nucleosides are readily converted to the corresponding 2-substituted inosines at room temperature in this media.

EXPERIMENTAL

The melting points reported were determined on a Kofler Heizbank and are corrected. The ultraviolet spectra were determined in aqueous solution with a Cary Model 14 spectrophotometer, whereas the infrared spectra were determined in pressed potassium bromide discs with a Perkin Elmer Model 221 spectrophotometer. The p.m.r. spectra were determined in 10% (w/v) DMSO-d_6 with a Varian A-60 spectrometer.

9-(Tri-*O*-acetyl-β-D-ribofuranosyl)-2,6-dichloropurine.

A mixture of tetra-*O*-acetyl-β-D-ribofuranose (10.0 g., 32 mmoles) and 2,6-dichloropurine (6.0 g., 32 mmoles) was heated, with continuous stirring, *in vacuo* at 130° until an opaque melt was obtained and vigorous gas evolution had ceased (3 to 5 min.). After the reaction flask had been cooled at room temperature for 5 min., the vacuum was broken and *p*-toluenesulfonic acid (150 mg.) was stirred into the thick melt. Vacuum and heat were reapplied and the reaction mixture was heated with continuous stirring at 130-135° for 25 min. The resulting clear glass was cooled to room temperature before it was dissolved in chloroform (25 ml.). The chloroform solution was filtered through dry Celite to remove unreacted purine and the filtrate was washed first with an equal volume of saturated aqueous sodium bicarbonate and then with water before it was dried over magnesium sulfate and evaporated to dryness *in vacuo*. The resulting residue was triturated with ether (100-125 ml.) and the crystals that formed were collected by filtration, washed with fresh ether, and dried *in vacuo*: yield, 9.0 g. (63%), m.p. 159°. Thin-layer chromatography on silica gel H (Merck) using 99 chloroform:1 methanol as eluant indicated the isolated product was essentially homogenous. λ max in mμ (ε x 10^{-3}): *p*H 1, 7-252 (5.0), 273 (9.1); *p*H 13-258 (11.6). ν̄ max in cm^{-1}: 3145, 3010, 2975, 2950, 2925 (CH); 1775, 1765, 1740 (C=O); 1590, 1555 (C=C, C=N).

2-Chloroadenosine.

A suspension of 9-(tri-*O*-acetyl-β-D-ribofuranosyl)-2,6-dichloropurine (8.6 g., 193 mmoles) in absolute methyl alcohol (200 ml.) was saturated at 5° with dry ammonia. Continuous magnetic stirring was carried out during the saturation and complete solution was effected after 1 hr. After saturation was complete, the reaction solution was sealed in a glass pressure bottle and allowed to stand at room temperature for one week. The reaction solution was evaporated to dryness *in vacuo* (after the removal of excess ammonia in a stream of dry nitrogen) and the resulting residue was triturated with chloroform (125 ml.) (using a magnetic stirrer) until a filterable solid was obtained. The solid was collected by filtration and this crude product was recrystallized from water (25 ml.) to give the pure material: yield, 3.3 g. (56%), m.p. 143-145°. Thin-layer chromatography on silica gel H (Merck) using 3 chloroform:1 methanol as the eluant indicated the sample was homogenous. λ max in mμ (ε x 10^{-3}): *p*H 1-265 (14.6); *p*H 7-264 (15.2); *p*H 13-265 (15.3). ν̄ max in cm^{-1}: 3400, 3340, 3160 broad (NH, OH); 2920 (CH); 1655 (NH); 1600 and 1575 (C=C, C=N); 1130, 1110, 1080, and 1040 (COC). P.m.r. (τ): 6.31 ($C_4'H$), 5.99 and 5.85 overlaping triplets ($C_4'H$ and $C_3'H$ or $C_2'H$), 5.40t ($C_3'H$ or $C_2'H$), 4.89t ($C_3'OH$ and $C_2'OH$), 4.57d ($C_5'OH$), 4.13d ($C_1'H$), 2.72 (NH_2), 1.62 (C_8H).

In a second, larger run 10 g. of 2,6-dichloropurine gave 7.8 g. (49%) of 2-chloroadenosine.

9-(Tri-*O*-acetyl-β-D-ribofuranosyl)-2,6-dibromopurine.

A mixture of 2,6-dibromopurine (1.6 g., 5.8 mmoles) and tetra-*O*-acetyl-β-D-ribofuranose (1.8 g., 5.8 mmoles) was treated as described above. After cooling to room temperature, the clear reaction glass was dissolved in chloroform (5 ml.), filtered to remove unreacted purine and the filtrate was washed first with an equal volume of saturated aqueous sodium bicarbonate and then with water before it was dried over magnesium sulfate and evaporated to dryness. Trituration of the resulting residue with ether gave the product as a crystalline solid; yield, 1.5 g. (50%), m.p. 147°. Thin-layer chromatography on silica gel H (Merck) using 99 chloroform:1 methanol as the eluant indicated the sample was essentially homogenous. λ max in mμ (ε x 10^{-3}): *p*H 1, 7-254 (5.1), 277 (9.4); *p*H 13-260 (11.2). ν̄ max in cm^{-1}: 3460-3400 (OH, H_2O); 3110, 2955 (CH); 1750, 1740 (C=O); 1580, 1550 (C=C, C=N).

2-Bromoadenosine.

A suspension of 9-(tri-*O*-acetyl-β-D-ribofuranosyl)-2,6-dibromopurine (1.4 g., 2.8 mmoles) in absolute methanol (40 ml.) was treated as described above. Evaporation of the reaction solution to dryness *in vacuo* gave an oily residue which solidified on trituration with chloroform (10 ml.). The crude solid was collected by filtration and recrystallized from ethanol (20 ml.) to give a 60% yield of purified product. Additional pigmented and inorganic impurities were removed from an ethyl acetate solution of this material by filtration through

dry Celite. Evaporation of the ethyl acetate to dryness *in vacuo* and recrystallization of the residue from acetone gave the pure 2-bromoadenosine; yield, 260 mg. (26%); m.p. indefinite. $[\alpha]_D^{22}$ -48.5° ± 0.8 (conc. 1.03 g./100 ml. methanol). Thin-layer chromatography on silica gel H (Merck) using 3 chloroform:1 methanol as eluant showed a single spot. λ max in mμ (ε x 10^{-3}): *p*H 1-266 (14.3); *p*H 7, 13-265 (14.9). ν max in cm^{-1}: 3400, 3320, 3200 broad (NH, OH); 2920 (CH); 1700 (C=O of acetone); 1645 (NH); 1590 and 1565 (C=C, C=N); 1110, 1080, and 1040 (COC). P.m.r. (τ): 7.91 (acetone), 6.33 unsym. multiplet ($C_5'H$), 5.99 and 5.84 overlaping triplets ($C_4'H$ and $C_3'H$ or $C_2'H$), 5.39t ($C_3'H$ or $C_2'H$), 4.89t ($C_3'OH$ and $C_2'OH$), 4.57 unsym. d ($C_5'OH$), 4.15d ($C_1'H$), 2.22 (NH_2), 1.63 (C_8H).

Anal. Calcd. for $C_{10}H_{12}BrN_5O_4 \cdot {}^1/_3C_3H_6O$: C, 36.14; H, 3.87; N, 19.16. Found: C, 35.98; H, 4.20; N, 18.85.

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