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Note

Synthesis of 9-α-D-rhamnofuranosyladenine

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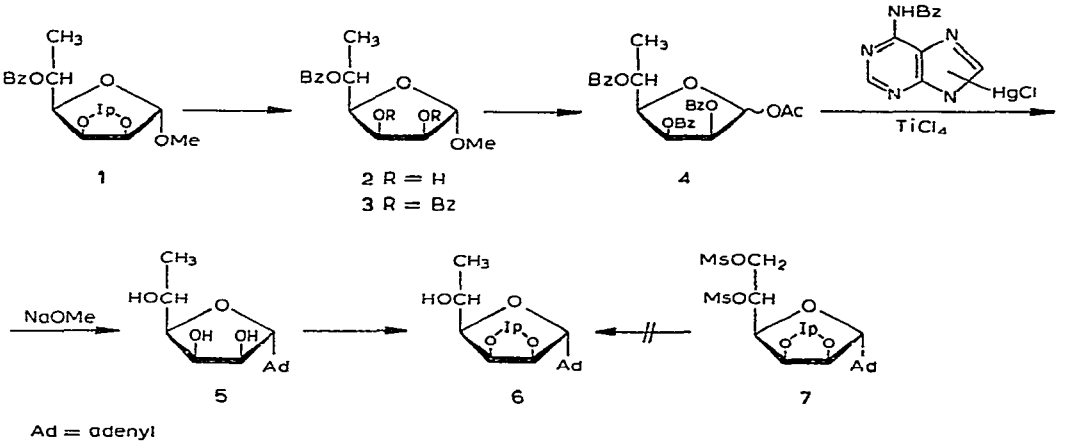
A number of 6-deoxyhexofuranosyl nucleosides were prepared by Baker and coworkers[1] as potential antitumor agents in a program of synthesis of "fraudulent" nucleosides. Although none of these compounds were useful as chemotherapeutic agents, it has become evident in recent years that some of them can bind to enzymes of nucleic acid metabolism. For example, 9-(6-deoxy-β-D-allofuranosyl)adenine was found to be an inhibitor of adenine phosphoribosyl transferase (EC 2.4.2.7)[2], and a substrate for adenosine deaminase (EC 3.5.4.4)[3]. The 5'-epimer, 9-(6-deoxy-α-L-talofuranosyl)adenine was also a substrate for the latter enzyme[3]. In a recent study[4], 9-α-D-mannofuranosyladenine[5] was found to be inhibitory to adenine phosphoribosyl transferase extracted from Ehrlich ascites tumor cells. This observation is of interest because it suggests that an "adenosine-like" configuration of the anomeric carbon atom and of the ring hydroxyl groups is not a requirement for binding sites to be located on nucleic acid enzymes. On the other hand, what may be more important is the *trans* relationship between the pair of *cis* hydroxyl groups of the furanose ring and the adenine ring, which is a structural feature common to both adenosine and 9-α-D-mannofuranosyladenine. In view of the evidence that such compounds can, indeed, bind to enzymes of nucleic acid metabolism, it is quite possible that suitable structural alterations could yield chemotherapeutically useful nucleosides. With these considerations in mind, the preparation of 9-(6-deoxy-α-D-mannofuranosyl)adenine was undertaken.

An earlier paper[6] reporting the synthesis and complete structure proof of 9-(6-deoxy-α-L-mannofuranosyl)adenine suggested that the structure proposed by Baker and Hewson[7] could not be correct. The preparation of the D enantiomer has now been accomplished by a route different from that published earlier[6]. Advantage was taken of the convenient preparation of methyl 5-*O*-benzoyl-2,3-*O*-isopropylidene-α-D-rhamnofuranoside (**1**) from D-mannose[8]. The isopropylidene group of **1** was removed with trifluoroacetic acid and the hydroxyl groups were benzoylated to give **3** in a crude form. Acetolysis was performed, under the conditions described by Baker and coworkers[1], to replace the methoxyl group with an acetoxyl group (**4**). It is important to note that these conditions cause an epimerization at C-2 when the *cis*

hydroxyls of the furanose ring are blocked as acetates[6,8], but the present work further demonstrates that such conversions will not occur with benzoyl blocking groups. It is fortuitous that earlier experiments utilized benzoate esters[1], since this epimerization had not yet been demonstrated.



The nucleoside analog, 9-α-D-rhamnofuranosyladenine (**5**), was prepared by reaction of 6-benzamidochloromercuripurine with **4** by the titanium tetrachloride method[9]. Removal of the blocking groups with hot methanolic sodium methoxide afforded the crude nucleoside, which was purified on an ion-exchange column[10]. The 39% yield of **5** was unexpectedly high and was considerably higher than that of similar coupling reactions for the L analog that used acetate blocking groups [6].It has been generally recognized that benzoyl blocking groups result in higher yields of nucleosides obtained from coupling reactions involving heavy metal salts of purines[11]. Treatment of **5** with acetone afforded the isopropylidene derivative **6**, which was identical to the L enantiomer with respect to melting point and i.r. spectrum[6].

The original intent of this work was to prepare **5** by direct reduction of 9-(2,3-*O*-isopropylidene-5,6-di-*O*-mesyl-α-D-mannofuranosyl)adenine[12] (**7**) with lithium aluminum hydride in one step to give **6**, or with the sodium borohydride–dimethyl sulfoxide reagent[13] in two steps, but these two methods were unsuccessful. In fact, **7** was usually recovered unchanged whereas a similar treatment of the corresponding methyl glycoside gave nearly stoichiometric yields[8].

EXPERIMENTAL

General. — Melting points were determined on a Kofler micro hot-stage and correspond to corrected values. I.r. spectra were recorded with a Perkin–Elmer Model 21 spectrophotometer and n.m.r. spectra with a Varian T-60A spectrometer. Moist organic solutions were dried with anhydrous magnesium sulfate. Evaporations

were performed under reduced pressure in a rotary evaporator at a bath temperature of 40°. Elemental analyses were performed by the Baron Consulting Co., Orange, Conn.

Methyl 2,3,5-tri-O-benzoyl-α-D-rhamnofuranoside (3). — Methyl 5-*O*-benzoyl-2,3-*O*-isopropylidene-α-D-rhamnofuranoside[8] (1, 9.53 g, 29.6 mmoles) was dissolved in 9:1 (v/v) trifluoroacetic acid–water[14] (100 ml) and the solution was kept for 0.5 h at room temperature. The mixture was evaporated, and the syrup was dissolved in chloroform (100 ml) and washed with saturated sodium hydrogencarbonate solution (100 ml), water (100 ml), and dried. Evaporation and drying in a high vacuum at 55° gave a gum weighing 8.35 g (100%); i.r. data: ν_{max}^{film} 3430 (strong, OH), 1722 (carbonyl), and 712 cm^{-1} (monosubstituted phenyl); n.m.r. data: τ 3.30 (sharp peak, OCH_3).

A solution of the gum in dry pyridine (40 ml) was stirred and chilled in an ice-bath, and treated with benzoyl chloride (8.5 ml). The reaction mixture was stirred for 1 h at 0°, and then for 24 h at room temperature. The dark solution was poured into a mixture of ice and saturated sodium hydrogencarbonate solution and stirred for 2 h. Chloroform (100 ml) was added to extract the product, and after 0.5 h the layers were separated. After an additional extraction of the aqueous layer with chloroform (50 ml), the extracts were combined, washed with saturated sodium hydrogencarbonate (100 ml), water (100 ml), and dried. Evaporation and three coevaporations with toluene to remove traces of pyridine gave an orange syrup, which was triturated with hot petroleum ether (b.p. 60–110°), and the extract decanted. This procedure was repeated and evaporation of traces of solvents yielded a hard glass (8.91 g); t.l.c. (Merck Silica Gel HF, 0.25 mm) indicated mainly one component, R_F 0.74 (9:1, v/v, ethyl acetate–methanol); the i.r. spectrum showed no hydroxyl peak.

Acetolysis of 3. — The glass just described was dissolved in a mixture of acetic acid (65 ml) and acetic anhydride (7.3 ml). The solution was chilled in an ice-bath and sulfuric acid (3.9 ml) was slowly added, dropwise. The reaction mixture was kept for 21 h at room temperature, and the brown solution was poured on ice chips (350 ml) and stirred until the ice melted. A hard gum separated, which was extracted from the aqueous mixture with chloroform (150 ml). Two additional extracts with chloroform (75 ml each) were combined with the original, and the solution was washed with saturated sodium hydrogencarbonate solution (2 × 200 ml). Sodium chloride was added to break emulsions, and the chloroform solution was dried and evaporated to a syrup (8.83 g); n.m.r. data: the methoxyl group (τ 3.30) had been replaced by an acetyl group (τ 2.26).

9-α-D-Rhamnofuranosyladenine (5). — The syrup just described (8.83 g, 17 mmoles), 6-benzamidochloromercuripurine (9.7 g, 20.5 mmoles), Celite-545 (9.7 g), and 1,2-dichloroethane (600 ml) were stirred together, and 100 ml of solvent was distilled off to remove traces of water. Titanium tetrachloride (2.9 ml) in fresh, dry 1,2-dichloroethane (75 ml) was added and the mixture was heated for 22 h at reflux. The cooled reaction mixture was treated with saturated sodium hydrogencarbonate (325 ml), stirred for 1.5 h, and filtered by suction through a pad of Celite. The pad was washed with hot 1,2-dichloroethane (150 ml), and the organic layer was separated

and washed with 30% aqueous potassium iodide (2 × 200 ml). A final wash with saturated sodium chloride solution (200 ml), drying, and evaporation afforded a brown foam (10.02 g). The foam was dissolved in methanol (200 ml), and the solution was treated with M methanolic sodium methoxide (12 ml), and heated for 1.5 h at reflux. The cooled solution was neutralized with Bio-Rad AG-50 (H^+) ion-exchange resin, the resin was removed by filtration, and the filtrate was evaporated to a thick, dark syrup. Methyl benzoate was removed by several azeotropic evaporations with water. The dark residue was chromatographed on a column (29 × 2.3 cm) of Bio-Rad AG1-X2 (OH^-, 200–400 mesh) ion-exchange resin[10] with 30% aqueous methanol as the eluent. The product was eluted in tubes 8–454 (10 ml fractions), the dark-colored material remaining adsorbed on the resin. The product **5** was crystallized as large prisms from water in several crops (1.992 g, 39% yield). One recrystallization gave the analytical sample, m.p. 155–157°, 197–198°; $[\alpha]_D^{20}$ +73.8° (*c* 0.986, water); L enantiomer[6]: m.p. 155–156.5°, 195–196°; $[\alpha]_D^{23}$ −72.3° (*c* 0.65, water); the i.r. spectra of both enantiomers were identical.

Anal. Calc. for $C_{11}H_{15}N_5O_4 \cdot H_2O$: C, 44.14; H, 5.73; N, 23.40. Found: C, 43.92; H, 5.73; N, 23.11.

9-(2,3-O-Isopropylidene-α-D-rhamnofuranosyl)adenine (**6**). — The nucleoside **5** (130 mg) was suspended in a stirred mixture of acetone (39 ml), 2,2-dimethoxypropane (3.9 ml), and *p*-toluenesulfonic acid monohydrate (0.83 g). Complete dissolution occurred in a few min. After 3 h, the solution was poured into a mixture of sodium hydrogencarbonate (1.3 g) and water (13 ml). The precipitate was removed by filtration and washed with acetone. The combined filtrates were evaporated to dryness, and the residue was triturated with chloroform (50 ml). A small amount of insoluble material was filtered off and the chloroform was evaporated. The residue was crystallized from methanol–water to afford 113 mg in two crops. Slow recrystallization from 4 ml of methanol–water (1:3, v/v) in an open flask gave large prismatic crystals (85 mg), m.p. 228–230°; L enantiomer[6]: m.p. 226–229°; the i.r. spectra of **6** and of the L enantiomer were identical.

Anal. Calc. for $C_{14}H_{19}N_5O_4$: C, 52.32; H, 5.97; N, 21.80. Found: C, 52.19; H, 5.83; N, 22.00.

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