Diehl for a supply of 2,3-*O*-benzylidene-β-D-ribofuranose and of 2,3,5-tri-*O*-benzoyl-D-ribose. Analyses were performed in the Institutes' Microanalytical Laboratory under the supervision of Dr. W. C. Alford.

BETHESDA 14, MD.

[CONTRIBUTION FROM THE ORGANIC CHEMICAL RESEARCH SECTION AND THE VIRAL AND RICKETTSIAL RESEARCH SECTION, RESEARCH DIVISION, LEDERLE LABORATORIES, AMERICAN CYANAMID COMPANY]

Synthesis and Biological Properties of Certain 5,6-Dichlorobenzimidazole Ribosides

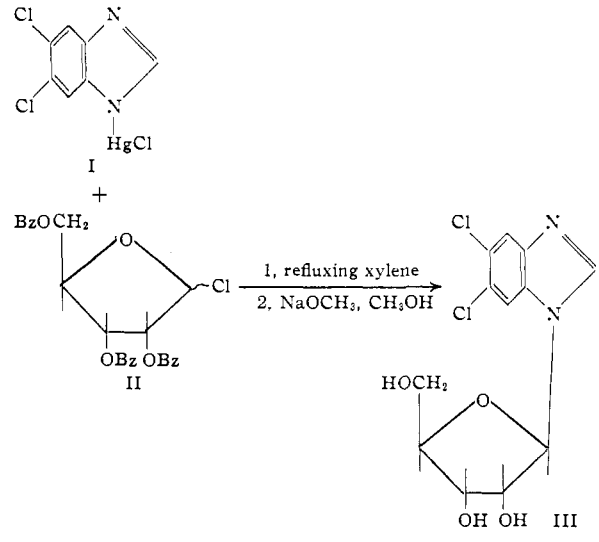
BY HENRY M. KISSMAN, RALPH G. CHILD AND MARTIN J. WEISS

RECEIVED SEPTEMBER 21, 1956

The syntheses of 1-β-D-ribofuranosyl-5,6-dichlorobenzimidazole (DRB, III) a known[1] but hitherto undescribed compound and of the β- and α-anomers of 1-(5-deoxy-D-ribofuranosyl)-,5,6-dichlorobenzimidazole (VI and VII, respectively) are reported. Evidence is presented showing that DRB does not inhibit the multiplication of the PR8 strain of influenza virus n mice. DRB and compounds VI and VII are also inactive against a variety of other viruses *in ovo*, in tissue culture and in mice.

The report by Tamm, *et al.*,[1] that 1-β-D-ribofuranosyl-5,6-dichlorobenzimidazole (III, DRB) inhibits influenza virus multiplication *in ovo* and in mice prompted us to test this and related compounds in our antiviral testing program.[2]

Compound II was unavailable to us and since experimental directions for its preparation have never been published, it was necessary to devise a synthesis. The only preparation of an N-glycoside of 5,6-dichlorobenzimidazole described in the literature is that of Weygand, Wacker and Wirth,[3] who condensed acetobromoglucose with the silver salt of 5,6-dichlorobenzimidazole to obtain 1-β-D-glucopyranosyl-5,6-dichlorobenzimidazole in 34% yield. A similar experiment by Antaki and Petrow[4] with the silver salt of 2-methyl-5,6-dichlorobenzimidazole failed. In our work we applied the "mercuric chloride method" which Davoll and Brown[5] had developed for the synthesis of glycosides of benzimidazole and 5,6-dimethylbenzimidazole. The chloromercuri derivative I of 5,6-dichlorobenzimidazole[3] was prepared according to the general procedure of these authors[5] and was allowed to react in xylene suspension with an equivalent of the 1-chloro sugar (II) obtained[6] from 1-O-acetyl-2,3,5-tri-O-benzoyl-D-ribofuranose (IV).[6,7] The blocked condensation product was not isolated *per se* but was de-O-benzoylated with sodium methoxide in methanol[8] to afford III as a crystalline solid, m.p. 215–216°,[9] in 68% over-all yield from IV. While it is not possible to furnish rigorous proof for the β-configuration of our product, this can be assumed from the high negative rotation value ($[\alpha]^{25}D$ −63° in pyridine[10]) and from analogy to results observed in the purine series by Baker and co-workers.[11] These authors found that in a condensation of a 1-chloro sugar with a chloromercuripurine, the entering purine moiety assumes a configuration *trans* to the 2-acyloxy group of the sugar ("C_1–C_2 *trans* rule").



Since 1,2,3-tri-O-acetyl-5-deoxy-D-ribofuranose (V)[12] was readily available in our laboratory, it was of interest to prepare 1-(5-deoxy-β-D-ribofuranosyl)-5,6-dichlorobenzimidazole (VI) as an analog of III. The synthesis was effected by condensing I with 2,3-di-O-acetyl-5-deoxy-D-ribofuranosyl chloride in refluxing xylene. The gummy reaction product was deblocked with methanolic sodium methoxide[8] to give a mixture of solids. This mixture was resolved into two crystalline compounds

(1) (a) I. Tamm, K. Folkers, C. Shunk and F. L. Horsfall, Jr., *J. Exptl. Med.*, **99**, 227 (1954); (b) I. Tamm and D. A. Tyrrell, *ibid.*, **100**, 541 (1954).

(2) It may be noted that other benzimidazole derivatives have been reported to exhibit similar properties:[1] *cf.* I. Tamm, *et al.*, *ibid.*, **98**, 245 (1953); *J. Bact.*, **72**, 42, 54, 59 (1956). Furthermore, DRB and a trichlorobenzimidazole riboside have been reported to inhibit mumps virus multiplication *in ovo*. I. Tamm, *Science*, **120**, 847 (1954); *cf.* F. L. Horsfall, Jr., *Bull. N. Y. Acad. Med.*, **31**, 783 (1955).

(3) F. Weygand, A. Wacker and F. Wirth, *Z. Naturforsch.*, **6b**, 25 (1951).

(4) H. Antaki and V. Petrow, *J. Chem. Soc.*, 2873 (1951).

(5) J. Davoll and G. B. Brown, THIS JOURNAL, **73**, 5781 (1951).

(6) H. M. Kissman, C. Pidacks and B. R. Baker, *ibid.*, **77**, 18 (1955).

(7) R. K. Ness, H. W. Diehl and H. G. Fletcher, Jr., *ibid.*, **76**, 763 (1954).

(8) G. Zemplen and E. Pacsu, *Ber.*, **62**, 1613 (1929).

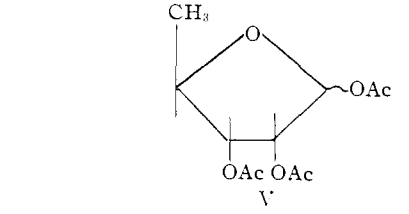
(9) As far as could be ascertained, physical characteristics of III have never been published.

(10) The rotation of β-ribazole, which is the corresponding 5,6-dimethylbenzimidazole derivative, is $[\alpha]^{20}D$ −45° in pyridine; F. Weygand and F. Wirth, *Chem. Ber.*, **85**, 1000 (1952). The rotation of α-ribazole is $[\alpha]^{23}D$ +9.9° in pyridine; N. G. Brink and K. Folkers, THIS JOURNAL, **74**, 2856 (1952).

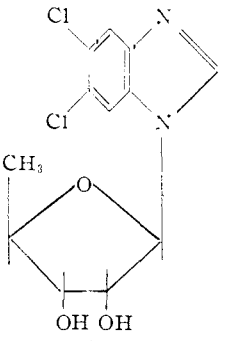
(11) B. R. Baker, J. P. Joseph, R. E. Schaub and J. H. Williams, *J. Org. Chem.*, **19**, 1786 (1954).

(12) H. M. Kissman and B. R. Baker, to be published.

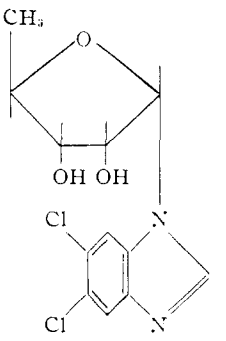
by three partition chromatograms on Celite from an ethyl acetate–heptane–methanol–water system (see Experimental section). Combustion analyses established that the two substances were isomeric and polarimetric data showed that the lower melting compound (m.p. 131–132°; $[\alpha]^{25}D$ −20.3° in ethanol) was the β-anomer VI while the higher melting substance (m.p. 182–183°; $[\alpha]^{25}D$ +12.1° in ethanol) was the α-anomer VII. There was obtained a 24% yield of VI, a 5% yield of VII and a 7% yield of mixed anomers. The formation of this relatively small amount of α-anomer as a by-product of the condensation reaction is an apparent exception to the "C_1–C_2 *trans* rule"[11] cited above. However, it should be realized that this rule is only a generalization and that reaction pathways[13] leading to α-anomers are also possible but less favored.



V



VI



VII

Biological Results.—DRB (III) was tested for potential antiviral activity in mice infected with the PR8 strain of influenza virus. In this test an increase in mean survival days of treated mice as compared to that of controls is used as the most significant criterion of virus multiplication inhibition. The results, which are summarized in Table I, clearly indicate that DRB has no effect on the multiplication of influenza virus in mice. Likewise, in tissue culture of HeLa cells infected with poliomyelitis virus, DRB and the two deoxyribosides, VI and VII, show no ability to prevent the multiplication of virus at the maximum tolerated dose (see Table II).

Tests with Columbia S.K. virus in mice demonstrated that DRB and compounds VI and VII were inactive at 1 mg./mouse twice daily for two days.[14a] Furthermore, DRB was shown to be inactive against Western equine encephalomyelitis, laryngotracheitis and pigeon pox *in ovo*.[14b]

(13) One possibility is a direct S_N2 displacement with a β-chloro sugar.

(14) Private communications from (a) Mr. E. Ewald and (b) Mr. F. Popken of these laboratories.

TABLE I

THE LACK OF EFFECT OF DRB (III) ON THE LONGEVITY OF INFLUENZA INFECTED MICE

Test no.	Compd. dose, mg./ mouse/ day	Virus dose, no. of LD_{50}s	Mortality ratios Treated	Mortality ratios Controls	Mean survival, days Treated	Mean survival, days Controls
1	6	52.7	5/5	10/10	1.0	6.1
2	4	20.6	4/5	9/10	6.8	7.2
3	4	2.1	3/5	6/10	7.8	10.0
4	4	2.1	3/4	6/10	8.6	10.0
5	4	1.5	3/4	5/10	8.3	10.6

TABLE II

THE LACK OF EFFECT OF DRB (III), VI AND VII ON POLIOMYELITIS VIRUS IN *HeLa* CELL CULTURES

Compound	Test no.	Compd. dose, mg./ml. of media	Virus dose, no. of ID_{50}s	Result
DRB	1	1.0	2.8	Toxic
DRB	1	0.1	2.8	Toxic
DRB	1	.01	2.8	Negative
DRB	2	.1	17.8	Toxic
DRB	2	.075	17.8	Toxic
DRB	2	.050	17.8	Toxic
DRB	2	.025	17.8	Negative and slightly toxic
DRB	3	.06	31.6	Toxic
DRB	3	.04	31.6	Negative and slightly toxic
DRB	3	.03	31.6	Negative and slightly toxic
DRB	3	.02	31.6	Negative
VI	1	1.0	28.0	Toxic
VI	1	0.01	28.0	Negative
VI	2	.5	16.0	Toxic
VI	2	.1	16.0	Toxic
VI	2	.05	16.0	Negative
VII	1	1.0	28.0	Toxic
VII	1	0.01	28.0	Negative
VII	2	.5	16.0	Toxic
VII	2	.1	16.0	Negative
VII	2	.05	16.0	Negative

Additional testing in other systems showed that DRB and compound VI were inactive against *Trypanosoma equiperdum* in mice at 50 and 100 mg./kg., respectively.[15a] DRB was inactive against a transplanted mammary adenocarcinoma in the C3H mouse, sarcoma 180 in mice and against the lymphosarcoma 6C3HED in mice all at 50 mg./kg. (slight toxicity at this level). Compound VI was inactive against a transplanted mammary adenocarcinoma in the C3H mouse and the 755 mammary adenocarcinoma in the C57/BL6 mouse at 75 mg./kg.[15b]

Tamm, *et al.*,[1] have reported that DRB inhibits the multiplication of influenza virus in chick embryo and in mice as evidenced by a drop in hemagglutination titer of the virus. With respect to the difference between this interpretation of the effect of DRB and our results, we would suggest that it is possible that hemagglutination inhibition is not a valid measure of the inhibition of virus multiplica-

(15) Private communications from (a) Dr. R. Hewitt and (b) Dr. J. Oleson and Miss S. Halliday of these laboratories.

tion. It may be noted that although Aureomycin[16] reduced or completely inhibited the hemagglutinating activity of mumps virus in chick embryos, it apparently had no appreciable influence on the rate of multiplication and infective titer of the mumps virus as measured by infectivity tests carried out in embryonated eggs.[17,18]

Experimental[19]

Chloromercuri-5,6-dichlorobenzimidazole (I).—A hot solution of 18.7 g. (0.1 mole) of 5,6-dichlorobenzimidazole[3] in 100 ml. of ethanol was added to a hot solution of 4 g. (0.1 mole) of sodium hydroxide in 100 ml. of water. The homogeneous solution was further diluted with 250 ml. of hot water and the temperature of the solution was held at 70°. There was added with vigorous stirring, a solution of 27.1 g. (0.1 mole) of mercuric chloride in 60 ml. of hot ethanol and then quickly with stirring two 500-ml. portions of hot water. The precipitate was collected and washed with two 150-ml. portions of hot water. After drying, there was obtained 39.7 g. (94%) of white powder.

Anal. Calcd. for $C_7H_3N_2Cl_3Hg$: Cl, 25.2. Found: Cl, 24.5.

1-β-D-Ribofuranosyl-5,6-dichlorobenzimidazole (II, **DRB**).—To 350 ml. of an ethereal hydrogen chloride solution (saturated at 0°) was added 7.9 g. (15.6 mmoles) of dried 1-O-acetyl-2,3,5-tri-O-benzoyl-D-ribofuranose (IV)[6,7] and 1 ml. of acetyl chloride. The solution was allowed to stand at −3° for 3 days and was then evaporated at room temperature *in vacuo*. The sirupy chloro sugar II was evaporated with several portions of benzene and was dissolved in 25 ml. of xylene. This solution was added to an azeotropically dried supension of 6.58 g. (15.6 mmoles) of I in 200 ml. of xylene and the mixture was stirred at reflux for 3 hours. The cooled reaction mixture was filtered through Celite[20] and the filtrate was evaporated to dryness *in vacuo*. The residue was dissolved in a mixture of chloroform (150 ml.) and of 30% aqueous potassium iodide solution (30 ml.) and the layers were separated. The organic phase was washed with another 20 ml. of the potassium iodide solution and then with 20 ml. of water. After drying over magnesium sulfate, the chloroform solution was filtered and evaporated under reduced pressure. The viscous residue was taken up in 150 ml. of ether and was partially decolorized with charcoal (Darco). The solution was again evaporated *in vacuo* and the residue was dissolved in 40 ml. of absolute methanol. To the solution was added 1.5 ml. of a 1 *N* methanolic sodium methoxide. To the solution was added 1.5 ml. of a 1 *N* methanolic sodium methoxide solution[8] and the mixture was allowed to reflux for 40 minutes. The solution remained at *p*H 8 throughout this period. The reaction mixture was concentrated to a small volume *in vacuo* and the solid which crystallized on standing was collected and washed with a little methanol to afford, after drying, 2.94 g.; m.p. 214–217°. Total evaporation of the filtrate and crystallization of the residue from a little methanol yielded another 0.73 g. of this material. The combined fractions were recrystallized from methanol with Darco and there was obtained in several portions 3.38 g. (68%) of white crystalline material; m.p. 214–217°. For analysis, a small amount of this was crystallized once more from ethanol and dried at 74° for 3 hours *in vacuo;* m.p. 215–216°; $[\alpha]^{25}D$ −63.3° (*c* 2.05 in pyridine). The compound showed the following absorption maxima in the ultraviolet[21]: λ_{max} 252 mμ (ε 4220), 284 mμ (ε 6520) and 293 mμ (ε 6380) in acid; 255 mμ (ε 5620), 287 mμ (ε 4600) and 296 mμ (ε 4730) in neutral solution; 255 mμ (ε 6000), 287 mμ (ε 4600) and 296 mμ (ε 4470) in base.

Anal. Calcd. for $C_{12}H_{12}N_2O_4Cl_2$: C, 45.16; H, 3.79; N, 8.78; Cl, 22.22. Found: C, 44.94; H, 3.98; N, 8.94; Cl, 22.23.

Anomeric Mixture of 1-(5-Deoxy-D-ribofuranosyl)-5,6-dichlorobenzimidazoles (VI and VII).—A solution of 5.2 g. (20 mmoles) of 1,2,3-tri-O-acetyl-5-deoxy-D-ribofuranose (V)[12] in 150 ml. of ethereal hydrogen chloride (saturated at 0°) and 2 ml. of acetyl chloride was allowed to stand at −3° for 3 days and was then worked up as described for the preparation of II (see above). The solution of the 1-chloro sugar in 40 ml. of anhydrous xylene was added to an azeotropically dried suspension of 8.44 g. (20 mmoles) of I in 200 ml. of xylene. The resulting mixture was allowed to reflux with stirring for 3 hours and was then filtered through a layer of Celite. The filtrate was evaporated *in vacuo* and the residue was dissolved in 100 ml. of chloroform and 20 ml. of a 30% potassium iodide solution. The layers were separated and the organic phase was washed with 20 ml. of the potassium iodide solution and with 20 ml. of water. The dried chloroform solution (magnesium sulfate) was evaporated under reduced pressure to afford 6.8 g. of yellow gum. This was redissolved in 30 ml. of absolute methanol containing 0.8 ml. of a 1 *N* methanolic sodium methoxide solution. The mixture was allowed to reflux for 30 minutes and was evaporated *in vacuo*. The brown residue was dissolved in 80 ml. of methylene chloride and the solution was partially decolorized with Darco. Evaporation of the solvent left 4.43 g. of tan glass. This was redissolved in methylene chloride and the solution was diluted with ether at the boiling point until crystallization started. There was obtained a total of 1.68 g. of solid in several fractions which melted from 122–125° to 129–133°. The combined fractions were used for *partition chromatogram A*. Total evaporation of the filtrates left 2.0 g. of fluffed glass which was used in *partition chromatograms B and C*.

1-(5-Deoxy-β-D-ribofuranosyl)-5,6-dichlorobenzimidazole (VI). Partition Chromatogram A.—The crystalline material (1.68 g.) isolated above was dissolved in 8 ml. of the lower and 8 ml. of the upper phase of the system heptane:ethyl acetate:methanol:water (3:2:3:2) and the liquid was mixed thoroughly with 16 g. of Celite.[22] This mixture was packed on top of a column which had been prepared from 150 g. of Celite and 75 ml. of the lower phase of the solvent system just described. The column (65.5 cm. × 3 cm.) was eluted with the upper phase and the effluent was allowed to pass through a recording ultraviolet spectrophotometer which had been set at 288 mμ. A small amount of material having absorption at that wave length was eluted from the column in the first 270 ml. of effluent, while the following 170 ml. did not contain such material. These fractions were discarded. A final fraction of 1155 ml. which contained much ultraviolet absorbing material was evaporated to a small volume to afford 1.24 g. of solid with m.p. 131–132°. Further evaporation of the filtrate give another 0.22 g. of material with m.p. 125–128°. The combined weight of 1.46 g. represents a 24% yield over-all from V. For analysis the substance was recrystallized from ether–methylene chloride; m.p. 131–132°; $[\alpha]^{25}D$ −20.3° (*c*, 0.98 in ethanol). The compound showed the following maxima in the ultraviolet: λ_{max} 254 mμ (ε 4500), 286 mμ (ε 6670) and 294 mμ (ε 6430) in acid; 255 mμ (ε 6060), 287 mμ (ε 4730) and 296 mμ (ε 4730) in

(16) Aureomycin is the Lederle Laboratories Division, American Cyanamid Company, trade mark for the antibiotic chlorotetracycline.

(17) S. C. Wong and H. R. Cox, *Ann. N. Y. Acad. Sci.*, **51**, 290 (1948).

(18) V. Cabasso, unpublished data.

NOTE ADDED IN PROOF.—Results obtained by Dr. V. Cabasso of These Laboratories since the writing of this paper support the contention that DRB has no effect on the infectivity titer of certain viruses although it may reduce their hemagglutination titer. Repetition and extension of the Tamm[2] experiments with DRB and mumps virus *in ovo* gave the following results. (1) DRB did not reduce the hemagglutination titer to the same extent reported by Tamm. The greatest decrease observed was 3- to 5-fold that of controls. (2) The reduction of hemagglutination titer was observed only after incubation periods of 120 to 144 hours. (3) A decrease in infectivity titer for the DRB treated chick embryos as compared to controls could not be demonstrated after incubation periods of 120 to 144 hours, despite the drop in the hemagglutination titer. Details of this work will be reported elsewhere. We are grateful to Dr. Cabasso for making these results available to us prior to publication.

(19) Melting points were taken on a Kofler micro hot stage and are corrected. Ultraviolet absorption spectra were determined on a Cary recording spectrophotometer.

(20) Celite, a product of the Johns–Manville Corporation, is diatomaceous earth.

(21) For the acid and base spectra, a methanolic solution was diluted 1:1 with 0.1 *N* hydrochloric acid and 0.1 *N* sodium hydroxide, respectively. The neutral spectrum was determined in methanol solution.

(22) The material used in these partition columns was Celite[20] 545 which had been washed with 6 *N* hydrochloric acid and then with distilled water until neutral and finally with methanol. The substance was dried in air to give a fluffy powder.

neutral solution; 256 mμ (ε 5820), 287 mμ (ε 4490) and 296 mμ (ε 4490) in base.

Anal. Calcd. for $C_{12}H_{12}N_2O_3Cl_2$: C, 47.54; H, 3.98; N, 9.24; Cl, 23.39. Found: C, 47.81; H, 4.03; N, 9.34; Cl, 23.49.

1-(5-Deoxy-α-D-ribofuranosyl)-5,6-dichlorobenzimidazole (VII). Partition Chromatogram B.—The 2.0 g. of glass, which had been obtained above, was dissolved in 4 ml. of the lower and 4 ml. of the upper phase of the solvent system used in the previous chromatogram, and the solution was mixed with 8 g. of Celite. This mixture was packed on top of a column which had been prepared from 150 g. of Celite and 75 ml. of the lower phase of the solvent system. The column (61 cm. × 3 cm.) was washed with the upper phase and the ultraviolet absorption spectrum of the effluent was determined as before. The first 470 cc. of effluent was discarded. The following 895 ml. contained ultraviolet absorbing material which was isolated by evaporation of the solution *in vacuo*. The residual solid was triturated with a little ether and there was obtained 0.93 g. of mixed solids (m.p. 121–160°). Washing with hot ether left 0.84 g. of solids (m.p. 130–160°) and from the ether wash there was obtained 0.046 g. of material with m.p. 123–125° (*i.e.*, impure VI). The 0.84 g. of mixed solids was rechromatographed in *partition chromatogram C*. The material was dissolved in 5 ml. of the upper and 5 ml. of the lower phase of the solvent system heptane:ethyl acetate:methanol:water (3.5:2:3:2), and the liquid was mixed with 10 g. of Celite. This mixture was added to a column prepared as described above and the chromatogram was developed with the upper phase of the solvent system. Material with ultraviolet absorption was eluted after 840 ml. of the solvent had passed through the column and continued to come off throughout the subsequent 950 ml. The effluent containing this material was collected in 50-ml. fractions and each fraction was evaporated to dryness independently. Each residue was crystallized from a little ether and melting points were taken. This procedure showed that the first ten fractions contained mostly the β-anomer VI (m.p. 127–130°), the following three fractions contained mixed materials (m.p. 124–175°) and the last five fractions contained a substance with m.p. 173–183°. These last fractions were pooled and were recrystallized several times from chloroform to afford 0.3 g. (5% yield over-all from V) of a compound, m.p. 182–183°; $[\alpha]^{25}D$ +12.1° (*c* 0.99, in ethanol). In the ultraviolet, the material showed the following maxima: λ_{max} 253 mμ (ε 4000), 286 mμ (ε 6310) and 294 mμ (ε 6180) in acid; 257 mμ (ε 5340), 288 mμ (ε 4610) and 297 mμ (ε 4850) in neutral solution; 257 mμ ε 5460), 287 mμ (ε 4610) and 297 mμ (ε 4490) in base.

Anal. Calcd. for $C_{12}H_{12}N_2O_3Cl_2$: C, 47.54; H, 3.98; N, 9.24; Cl, 23.39. Found: C, 47.17; H, 4.00; N, 9.31; Cl, 23.81.

The combined mixed solids from fractions 11 through 13 weighed 0.45 g. (7% over-all yield from V).

Antiviral Tests

Influenza in Mice.—The PR8 strain of influenza virus was administered intranasally to 14 to 16 g. Swiss white albino mice under light ether anesthesia. DRB (III) was administered intraperitoneally (2.0 to 3.0 mg., suspended in 0.25 ml. of water with the aid of Tween-20[23]) 4 hours later and continued twice daily for 5 days. At the end of 13 days, the mortality rates of treated groups were compared to that of the control groups. Table I lists the results of five separate tests with decreasing doses of virus.

Poliomyelitis in Tissue Culture.—Washed test-tube cultures of a human epidermoid carcinoma, strain HeLa, were infected with Type I human poliomyelitis virus, Mahoney strain. One hour later, the media were replaced with maintenance media (Lactalbumen hydrolyzate plus 4% cow serum)[24] containing the compound to be tested in various dilutions. The cultures were incubated at 37° and were observed microscopically daily for 6 days. An identical set of cultures received the compound but no virus. A control group of non-treated and non-infected cells as well as a titration of the virus inoculum was carried along with each treated group. A negative result was manifested by a cytopathogenic degeneration in the treated group when no signs of toxicity were present in the group containing the compound and no virus. Table II lists the results of separate tests on the three compounds.

Acknowledgment.—We would like to thank Mr. Charles Pidacks for advice and help in carrying out the partition chromatograms and Mr. J. P. Joseph for the preparation of 5,6-dichlorobenzimidazole. We wish also to thank Dr. J. R. Vaughan for bringing this problem to our attention. Microanalyses were carried out by Mr. L. Brancone and staff and spectroscopic and polarimetric data were supplied by Mr. W. Fulmor and staff.

(23) Tween-20, a product of the Atlas Powder Company, is a wetting agent.

(24) J. L. Melnick, *Ann. N. Y. Acad. Sci.*, **61**, 754 (1955).

PEARL RIVER, N. Y.

[CONTRIBUTION FROM THE DEPARTMENT OF AGRICULTURAL BIOCHEMISTRY, UNIVERSITY OF MINNESOTA]

Synthesis of D-3,4-di-*O*-methyl-erythritol[1]

BY IRWIN J. GOLDSTEIN AND F. SMITH

RECEIVED SEPTEMBER 13, 1956

D-3,4-Di-*O*-methyl-erythritol has been prepared from methyl 5,6-di-*O*-methyl-D-glucofuranoside by oxidation with periodate followed by reduction and hydrolysis. It has been characterized as the bis-*p*-nitrobenzoate.

The successful use of periodic acid and its salts in synthetic and analytical organic chemistry for cleaving 1,2-glycols has been firmly established.[2–6]

This paper described its use for the synthesis of D-3,4-di-*O*-methyl-erythritol from methyl 5,6-di-*O*-methyl-D-glucofuranoside. The latter was obtained by the following series of reactions: D-glucose → 1,2;5,6-di-*O*-isopropylidene-D-glucofuranose → 3-*O*-benzyl-1,2;5,6-di-*O*-isopropylidene-glucofuranose → 3-*O*-benzyl-1,2-*O*-isopropylidene-D-glucofuranose → 3-*O*-benzyl-5,6-di-*O*-methyl-1,2-*O*-isopropylidene-D-glucose → 5,6-di-*O*-methyl-1,2-*O*-isopropylidene-D-glucose → methyl 5,6-di-*O*-methyl-D-glucofuranoside. Oxidation of the latter with periodic acid at room temperature in the usual manner furnished a dialdehyde by cleavage between the hydroxyl groups at C_2 and C_3. Reduction of the dialdehyde with sodium borohydride gave the corresponding alcohol from which D-3,4-

(1) Paper No. 3583, Scientific Journal Series, Minnesota Agricultural Experiment Station, University of Minnesota.

(2) E. L. Jackson, "Organic Reactions," Vol. II, John Wiley and Sons, Inc., New York, N. Y., 1944, p. 341.

(3) J. R. Dyer, "Methods of Biochemical Analysis," Vol. III, Ed. by D. Glick, Interscience Pub., Inc., New York, N. Y., 1956, p. 111.

(4) J. C. Sowden, THIS JOURNAL, **72**, 808 (1950).

(5) J. C. Sowden, *ibid.*, **73**, 5496 (1951).

(6) G. W. Huffman, Bertha A. Lewis, F. Smith and D. R. Spriestersbach. *ibid.*, **77**, 4346 (1955).