771. *Ribose and its Derivatives. Part IX.* The Use of Carbonyl Esters in Synthesis of α-Derivatives.*

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Chloroformic esters condense with methyl ribofuranoside and with ribose, yielding products containing both cyclic and acyclic carbonyl residues, the uses of which in preparing α-derivatives of the sugar are discussed. Methyl and benzyl ribopyranosides, on condensation with chloroformic esters, yield acyclic or mixed cyclic and acyclic carbonyl derivatives according to the conditions under which the condensation is carried out.

We have reported briefly [1] attempts to use carbonate esters of ribose as intermediates in synthesis. While these investigations were in progress, similar experiments were communicated to us privately by Khorana and have since been reported in full.[2,3,4] We have used methods for the synthesis of carbonyl esters of ribose which differ from those employed by Khorana and his co-workers, and we now report the potential uses of both furanose and pyranose carbonyl derivatives of ribose.

Zemplén and Laszlo [5] first prepared alkoxycarbonyl derivatives of carbohydrates by condensation with chloroformic esters; then Allpress and Haworth [6] carried out similar condensations in presence of pyridine, but they obtained both acyclic and cyclic carbonate esters if the reaction was carried out in presence of aqueous alkali. It seemed to us that the latter method might yield useful derivatives of ribose. Chloroformic esters are to be preferred to carbonyl chloride for the preparation of carbonyl esters, since, in the latter case, isolated hydroxyl residues must be protected (*e.g.*, with a benzyl residue [2]) to avoid the formation of polymeric products and we found this to be unnecessary if a chloroformic ester is used.

* Part VIII, *J.*, 1959, 584.

[1] Barker, Gillam, and Spoors, *Chem. and Ind.*, 1956, 1312.
[2] Tener, Wright, and Khorana, *J. Amer. Chem. Soc.*, 1956, **78**, 506.
[3] Tener and Khorana, *J. Amer. Chem. Soc.*, 1957, **79**, 437.
[4] Tener, Wright, and Khorana, *J. Amer. Chem. Soc.*, 1957, **79**, 441.
[5] Zemplén and Laszlo, *Ber.*, 1915, **48**, 915.
[6] Allpress and Haworth, *J.*, 1924, 1223.

Condensing methyl αβ-D-ribofuranoside with methyl chloroformate in presence of either pyridine or aqueous sodium hydroxide yielded what we presume to be methyl 5-*O*-methoxycarbonyl-αβ-D-ribofuranoside 2,3-carbonate. D-Ribose with methyl chloroformate in presence of aqueous sodium hydroxide was then found to yield what are assumed to be 1,5-di-*O*-methoxycarbonyl-α- and -β-D-ribofuranose 2,3-carbonates. In an attempt to establish the general course of this type of reaction, ribose was condensed with benzyl chloroformate, but no crystalline product could be obtained. Methyl chloroformate with methyl αβ-D-ribopyranoside in pyridine gave crystalline methyl 2,3,4-tri-*O*-methoxycarbonyl-αβ-D-ribopyranoside. On the other hand, methyl or benzyl αβ-D-ribopyranoside, in presence of aqueous sodium hydroxide, yielded what, for reasons outlined below, we believe to be methyl and benzyl 2-*O*-methoxycarbonyl-αβ-D-ribopyranoside 3,4-carbonate respectively. Benzyl β-D-ribopyranoside with benzyl chloroformate in presence of aqueous sodium hydroxide yielded benzyl 2-*O*-benzyloxycarbonyl-β-D-ribopyranoside 3,4-carbonate. Hydrogenation of this material in dioxan with palladised charcoal yielded crystalline benzyl β-D-ribopyranoside 3,4-carbonate, and this was hydrogenated in ethanol with the same catalyst to D-ribose 3,4-carbonate, an oil which was converted into the crystalline toluene-*p*-sulphonylhydrazone. The structure of the ribose carbonate was proved when it consumed one mol. of sodium metaperiodate and released one mol. of titratable acid. It follows that the structures of the two intermediates from which the ribose carbonate was obtained are as specified above. It is also considered likely that condensation of methyl and benzyl ribopyranoside with methyl chloroformate introduces a cyclic carbonyl residue at positions 3 and 4. We believe that the formation of a cyclic carbonate from methyl ribofuranoside in presence of aqueous alkali or pyridine is due to the favourable disposition of the 2- and the 3-hydroxyl group and find it highly likely that the cyclic carbonyl group is at these positions in the ribofuranosyl residue.

In the above compounds the presence of cyclic or acyclic carbonyl residues can readily be detected by infrared spectroscopy. The C=O stretching frequency of the cyclic carbonyl residue lay between 1810 and 1820 cm.$^{-1}$, whereas that of the acyclic residue occurred at 1750 cm.$^{-1}$. This distinction has been confirmed in a large number of cases.[7]

The object of the present investigations was to facilitate the production of α-ribosyl derivatives by introducing a 2-acyl group that cannot participate in reactions at position 1. The above results do not do this in the ribopyranose series. To test the possibility in the ribofuranose series, the α- and the β-form of 1,5-di-*O*-methoxycarbonyl-D-ribofuranose 2,3-carbonate were separately converted into the 1-chloro-derivatives. These, on treatment with methanol and silver carbonate followed by removal of ester residues, yielded methyl ribofuranosides; chromatography of the crude products showed that the β-ester gives rise entirely to methyl α-D-ribofuranoside, whereas the α-ester yields largely the α-glycoside. Syrupy 5-*O*-methoxycarbonyl-αβ-D-ribofuranosyl bromide 2,3-carbonate prepared from the anomeric mixture of esters also yielded a mixture of α- and β-glycosides, approximately in the proportion 3 : 1. Experiments were then carried out by Wright and Khorana's method [8] to test the potentialities of carbonate esters in preparing α-ribofuranose 1-phosphate. Speed of operation is essential in this method of phosphorylation and it was found that the 1-chloro-derivatives described above did not react rapidly enough to make their use profitable. Interaction of 5-*O*-methoxycarbonyl-αβ-D-ribofuranosyl bromide 2,3-carbonate with diethylammonium dibenzyl phosphate gave, after removal of protecting groups, D-ribofuranose 1-phosphate (isolated as its dicyclohexylammonium salt). This material was shown by the method of Khorana, Tener, Wright, and Moffatt [9] to contain some β-isomer, the proportion of which was determined by using the enzyme nucleoside phosphorylase. Unreliable results were obtained with the enzyme

[7] Hough, Priddle, Theobald, Barker, Douglas, and Spoors, *Chem. and Ind.*, 1960, 148.
[8] Wright and Khorana, *J. Amer. Chem. Soc.*, 1955, **77**, 3423.
[9] Khorana, Tener, Wright, and Moffatt, *J. Amer. Chem. Soc.*, 1957, **79**, 430.

from pig liver [10] owing to the presence in the preparation of xanthine oxidase, and it has been pointed out [11] that falsely high estimates of the proportion of α-D-ribofuranose 1-phosphate may be obtained with enzyme preparations which have not been thoroughly tested for the presence of xanthine oxidase. It was found that the nucleoside phosphorylase of calf spleen [12] is free from xanthine oxidase and, using this enzyme, we estimated that the crude ribose 1-phosphate contained approximately 25% of the α-isomer. A similar conclusion was reached by measurement of the specific rotation of the dicyclohexylammonium salt of the product.

These results were unexpected for two reasons. First, it had been found that 5-*O*-methoxycarbonyl-αβ-D-ribofuranosyl bromide 2,3-carbonate gave predominantly the α-glycoside on reaction with methanol. Secondly, Khorana and his co-workers had reported obtaining predominantly the α-phosphate from their 5-*O*-acetyl-D-ribofuranosyl bromide 2,3-carbonate. As has been previously emphasised,[3,13] the formation of the β-isomer in reactions involving 2-*O*-acetylpentafuranosyl halides is attributable to shielding of the α-side of $C_{(1)}$ by the neighbouring acetoxy-residue, resulting in Walden inversion when an α-halogeno-derivative is concerned. Khorana and his co-workers believe that their halogeno-sugar derivative has the β-configuration because the product consists mainly of the α-phosphate. However, it is considered inadmissible to assume an inversion of configuration during a reaction in which the shielding effect of a 2-acetoxy-residue is absent. The difference between our results and those of Khorana and his co-workers may be due to the fact that, whereas in our experiments the bromine atom was introduced at position 1 under mild conditions, the replacement of the glycosidic methoxyl group by bromine, in Khorana's experiments, required much more vigorous conditions and the composition of the anomeric mixture of bromides may have been different. It is concluded that, under certain conditions, cyclic carbonates of ribofuranose, but not ribopyranose, are useful intermediates in the synthesis of α-derivatives of the sugar. It is believed that the absence of a participating group at position 2 results in a less uniform steric course in reactions at position 1 and that the proportions of α- and β-isomers produced by replacement of halogen at this position depend to a large extent on such factors as the configuration at position 1 in the starting material, the nature of the incoming residue, and the conditions under which the reaction is carried out.

Experimental

Methyl 5-O-*Methoxycarbonyl*-αβ-D-*ribofuranoside* 2,3-*Carbonate*.—(*a*) Methyl αβ-D-ribofuranoside (3·6 g.), water (10 c.c.), and methyl chloroformate (7 c.c.) were vigorously stirred together at 0° and 2N-sodium hydroxide was added at such a rate that the temperature did not rise above 3°. Addition of sodium hydroxide was stopped when the solution became permanently alkaline to litmus. The gum which was formed was washed twice with water at 0° (100 c.c.) and distilled under reduced pressure to yield a *product* having b. p. 140°/0·05 mm., n_D^{20} 1·4590 (Found: C, 43·3; H, 4·7; OMe, 24·8. $C_9H_{12}O_8$ requires C, 43·5; H, 4·9; OMe, 25·0%), $\nu_{max.}$ (C=O) 1750, 1810 cm.$^{-1}$.

(*b*) A mixture of methyl αβ-D-ribofuranoside (2 g.), methyl chloroformate (7 c.c.), and pyridine (15 c.c.) was left at room temperature with the exclusion of moisture for 4 hr. The solvent was removed under reduced pressure and the residue was shaken with water (40 c.c.) and extracted three times with chloroform (25 c.c.). The combined extracts were washed with water, then dried ($MgSO_4$), and the solvent was removed under reduced pressure. The residue had b. p. 140°/0·05 mm., n_D^{20} 1·4586 (Found: OMe, 24·9%).

1,5-*Di*-O-*methoxycarbonyl*-D-*ribose* 2,3-*Carbonate*.—To a cooled and stirred mixture of D-ribose (3 g.), water (25 c.c.) and methyl chloroformate (10 c.c.), 2N-sodium hydroxide (approx.

[10] Kalckar, *J. Biol. Chem.*, 1947, **167**, 477.
[11] Barker and Gillam, *Biochim. Biophys. Acta*, 1960, **40**, 163.
[12] Price, Otey, and Plesner in Colowick and Kaplan's "Methods in Enzymology," Academic Press Inc., New York, 1957, Vol. II, p. 448.
[13] Haynes and Newth, *Adv. Carbohydrate Chem.*, 1955, **10**, 207.

45 c.c.) was added at such a rate that the temperature did not rise above 1°. Addition was stopped when the solution became permanently alkaline to litmus. The precipitate was dissolved in hot ethyl acetate (25 c.c.) and the solution, after filtration, was kept at room temperature overnight. Fractional crystallisation from ethyl acetate of the anomeric mixture obtained (5 g.) yielded the less soluble 1,5-*di*-O-*methoxycarbonyl*-β-D-*ribofuranose* 2,3-*carbonate* as prisms, m. p. 180°, $[\alpha]_D^{16}$ −152° (*c* 1·03 in $CHCl_3$) (Found: C, 41·3; H, 4·3; OMe, 21·5. $C_{10}H_{12}O_{10}$ requires C, 41·1; H, 4·1; OMe, 21·2%), $\nu_{max.}$ (C=O) 1750, 1810 cm.$^{-1}$, and 1,5-*di*-O-*methoxycarbonyl*-α-D-*ribofuranose* 2,3-*carbonate* as needles, m. p. 162°, $[\alpha]_D^{16}$ −76° (*c* 1·0 in $CHCl_3$) (Found: C, 41·3; H, 4·4; OMe, 21·5%), $\nu_{max.}$ (C=O) 1750, 1810 cm.$^{-1}$.

Conversion of 1,5-*Di*-O-*methoxycarbonyl*-D-*ribofuranose* 2,3-*Carbonate into Methyl Ribofuranosides.*—(*a*) The material (0·6 g.) was boiled in a mixture of chloroform (15 c.c.) and titanium tetrachloride (1·5 c.c.) with exclusion of moisture for 1 hr., then poured into water at 0°. The chloroform layer was washed with water (6 × 50 c.c.), dried ($MgSO_4$), and evaporated under reduced pressure. The residual crystals (0·3 g.) were dissolved in dioxan (6 c.c.) and shaken for 3 hr. at room temperature with methanol (60 c.c.), freshly prepared silver carbonate (3·0 g.), and anhydrous calcium sulphate (1·5 g.). Solids were removed and the filtrate was evaporated to a syrup under reduced pressure. The syrup was warmed in methanol (30 c.c.) on the steam-bath for 0·5 hr. with 5% (w/v) aqueous barium hydroxide (30 c.c.). The solution was cooled, saturated with carbon dioxide, filtered, and evaporated under reduced pressure, yielding syrupy methyl D-ribofuranoside (0·15 g.). A portion of the material obtained from 1,5-di-*O*-methoxycarbonyl-α-D-ribofuranose 2,3-carbonate, when oxidised by sodium metaperiodate, consumed 1·05 mol. Another portion of this syrup was chromatographed on paper in butan-1-ol–water and gave a strong spot (R_F 0·33) corresponding to methyl α-D-ribofuranoside and a faint spot (R_F 0·42) corresponding to methyl β-D-ribofuranoside.[14] A portion of the syrup obtained from 1,5-di-*O*-methoxycarbonyl-β-D-ribofuranose 2,3-carbonate was oxidised in the same way and consumed 1·03 mol. of sodium metaperiodate. On chromatography in the solvent system named above it gave a single spot (R_F 0·33).

(*b*) 1,5-Di-*O*-methoxycarbonyl-αβ-D-ribofuranose 2,3-carbonate was converted into the anomeric syrupy mixture of bromides [15] and was shaken for 2 hr. at room temperature with benzene (10 c.c.), methanol (40 c.c.), silver carbonate (8 g.), and anhydrous calcium sulphate (3 g.) in presence of a few glass beads. Solids were removed and the filtrate was evaporated under reduced pressure. The remaining syrup was dissolved in methanol (5 c.c.) and warmed on the steam-bath for 30 min. with barium hydroxide octahydrate (2·7 g.) in water (20 c.c.). Carbon dioxide was passed through the solution and, after filtration, the filtrate was evaporated under reduced pressure to a syrup. Chromatography of a portion of this material in butan-1-ol–water gave two spots corresponding to methyl α-D-ribofuranoside and methyl β-D-ribofuranoside. A portion of the crude syrup was sublimed at 0·01 mm. (bath-temp. 100°). The sublimed syrup had $[\alpha]_D^{14}$ +95° (*c* 1·04 in MeOH), corresponding to 75% of the α-isomer in the mixture.

Conversion of 1,5-*Di*-O-*methoxycarbonyl*-D-*ribose* 2,3-*Carbonates into* α- *and* β-D-*Ribofuranose* 1-*Phosphate.*—The anomeric mixture of the starting material (1 g.) was converted into the syrupy bromides which were treated in benzene (5 c.c.) with dibenzyl phosphate (1·37 g.) and triethylamine (0·5 g.) in benzene (7 c.c.). Dicyclohexylammonium αβ-D-ribofuranose 1-phosphate was isolated as described by Tener, Wright, and Khorana [4] and had m. p. 150—158°, $[\alpha]_D^{21}$ +18·0° (*c* 1·78 in H_2O) (Found: N, 6·3; P, 7·27. Calc. for $C_{17}H_{37}O_8N_2P$: N, 6·54; P, 7·23%). After conversion of a portion of this material into the pyridinium salt and treatment with dicyclohexylcarbodi-imide,[9] chromatography indicated that some of the phosphorus-containing material was converted into cyclic phosphate (R_F 0·49) and phosphorylurea derivative (R_F 0·9), but some of the the material remained unchanged and had R_F 0·1. A portion of the dicyclohexylammonium salt was incubated with calf-spleen nucleoside phosphorylase as described by Barker and Gillam.[11] The following values for ΔE_{290} were obtained: 0·46 (0·23) (5 min.), 0·40 (0·15) (10 min.), 0·40 (0·04) (20 min.): figures in parentheses are values obtained under comparable conditions with natural D-ribose 1-phosphate.

Methyl 2,3,4-*Tri*-O-*methoxycarbonyl*-αβ-D-*ribopyranoside.*—Methyl chloroformate (10 c.c.)

[14] Barker and Smith, *J.*, 1954, 2151.
[15] Howard, Lythgoe, and Todd, *J.*, 1947, 1052.

in chloroform (10 c.c.) was added slowly to methyl αβ-D-ribopyranoside (1·62 g.) in pyridine (10 c.c.) and chloroform (25 c.c.) at 0—3°. The mixture was left at room temperature with exclusion of moisture for 4 hr., then evaporated under reduced pressure. The residue was shaken with water (40 c.c.) and extracted with chloroform (3 × 25 c.c.), and the combined extracts were washed with water and dried ($MgSO_4$). Removal of solvent under reduced pressure gave *methyl* 2,3,4-*tri*-O-*methoxycarbonyl*-αβ-D-*ribopyranoside* which crystallised in needles (from methanol), m. p. 120—123°, $[\alpha]_D^{20}$ −74° (*c* 1·12 in $CHCl_3$) (Found: C, 42·3; H, 5·4; OMe, 36·4. $C_{12}H_{18}O_{11}$ requires C, 42·6; H, 5·3; OMe, 36·7%), $\nu_{max.}$ (C=O) 1750 cm.$^{-1}$.

Methyl 2-O-*Methoxycarbonyl*-αβ-D-*ribopyranoside* 3,4-*Carbonate*.—Methyl αβ-D-ribopyranoside (2·1 g.), water (10 c.c.) and methyl chloroformate (7 c.c.) were stirred at 0° and 2N-sodium hydroxide was added slowly, at <3°, until the solution became permanently alkaline to litmus. The solvent was decanted from the gum, which was washed twice with water (100 c.c.) and crystallised from ethanol, yielding the *product* as needles, m. p. 140—141°, $[\alpha]_D^{16}$ −158° (*c* 1·27 in $CHCl_3$) (Found: C, 43·75; H, 5·0; OMe, 25·2. $C_9H_{12}O_8$ requires C, 43·5; H, 4·85; OMe, 25·0%), $\nu_{max.}$ (C=O) 1750, 1810 cm.$^{-1}$.

Benzyl 2-O-*Methoxycarbonyl*-β-D-*ribopyranoside* 3,4-*Carbonate*.—Benzyl β-D-ribopyranoside (2 g.) was treated with methyl chloroformate (7 c.c.) as described above. The *product* separated in needles from ethanol (1 g.), m. p. 157·5—158·5°, $[\alpha]_D^{23}$ −159° (*c* 5·75 in $CHCl_3$) (Found: C, 55·3; H, 5·0; OMe, 9·6. $C_{15}H_{16}O_8$ requires C, 55·5; H, 4·9; OMe, 9·6%), $\nu_{max.}$ (C=O) 1750, 1810 cm.$^{-1}$.

Benzyl 2-O-*Benzyloxycarbonyl*-β-D-*ribopyranoside* 3,4-*Carbonate*.—Benzyl β-D-ribopyranoside (2 g.) with benzyl chloroformate (10 c.c.), as described above, yielded a *product* (2 g.) as needles (from ethanol), m. p. 168·5—169·5°, $[\alpha]_D^{20}$ −151° (*c* 2·0 in $CHCl_3$) (Found: C, 62·5; H, 5·5. $C_{21}H_{20}O_8$ requires C, 62·8; H, 5·0%), $\nu_{max.}$ (C=O) 1750, 1810 cm.$^{-1}$.

Benzyl β-D-*Ribopyranoside* 3,4-*Carbonate*.—Benzyl 2-*O*-benzyloxycarbonyl-β-D-ribopyranoside 3,4-carbonate (0·59 g.) was hydrogenated under slight positive pressure in presence of dioxan (25 c.c.) and 10% palladium–charcoal (0·5 g.). Uptake of hydrogen was complete in 1 hr. and, after removal of solvent and catalyst, the *product* crystallised as needles and, recrystallised from water, had m. p. 119—120°, $[\alpha]_D^{20}$ −175° (*c* 2·5 in $CHCl_3$) (Found: C, 58·5; H, 5·2. $C_{13}H_{14}O_6$ requires C, 58·7; H, 5·26%), $\nu_{max.}$ (C=O) (evaporated film) 1810 cm.$^{-1}$.

Benzyl 2-O-*Toluene*-p-*sulphonyl*-β-D-*ribopyranoside* 3,4-*Carbonate*.—Benzyl β-D-ribopyranoside 3,4-carbonate (0·1 g.), pyridine (1 c.c.), and toluene-*p*-sulphonyl chloride (0·8 g.) were mixed at 0° and left overnight at room temperature with exclusion of moisture. Water (1 c.c.) was added and the *product* which separated was collected; crystallised from ethanol, it had m. p. 171° (Found: C, 57·3; H, 5·3. $C_{20}H_{20}O_8S$ requires C, 57·2; H, 4·8%). The material did not react with sodium iodide when heated in acetone at 100° for 2 hr.

D-*Ribose* 3,4-*Carbonate*.—Benzyl β-D-ribopyranoside 3,4-carbonate (0·5 g.) was hydrogenated under slight positive pressure in presence of ethanol (25 c.c.) and 10% palladium–charcoal (0·5 g.). Removal of catalyst and solvent gave D-ribose 3,4-carbonate as a hygroscopic gum. A portion of the gum (0·0245 g.) was diluted to 10 c.c. with 0·0638N-sodium metaperiodate. At various intervals samples (1 c.c.) were withdrawn and excess of periodate was determined by the arsenite method. The following mols. of periodate were consumed: 0·727 (10 min.), 0·770 (40 min.), 0·913 (100 min.), 0·955 (160 min.), 1·005 (240 min.), 1·079 (340 min.). The remaining syrupy D-ribose 3,4-carbonate was converted by the method of Easterby, Hough, and Jones [16] into D-*ribose* 3,4-*carbonate toluene*-p-*sulphonylhydrazone*, m. p. 197—198° (decomp.), $[\alpha]_D^{20}$ −129·5° (*c* 2·0 in pyridine) (Found: C, 45·5; H, 4·8; N, 8·3. $C_{13}H_{16}O_7N_2S$ requires C, 45·5; H, 4·7; N, 8·1%).

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[16] Easterby, Hough, and Jones, *J.*, 1951, 3416.