Carbohydrate Research, 27 (1973) 353–362

BRANCHED-CHAIN GLYCOSYL α-AMINO ACIDS

PART II SYNTHESIS OF 2-D- AND 2-L-(3-DEOXY-1,2 5,6-DI-*O*-ISOPROPYLIDENE-α-D-GLUCOFURANOS-3-YL)GLYCINE ANALOGS OF THE SUGAR MOIETY OF THE POLYOXINS

ALEX ROSENTHAL, COLIN M RICHARDS, AND KOICHI SHUDO
Department of Chemistry, The University of British Columbia, Vancouver 8, B C (Canada)
(Received October 13th, 1972, accepted November 7th, 1972)

ABSTRACT

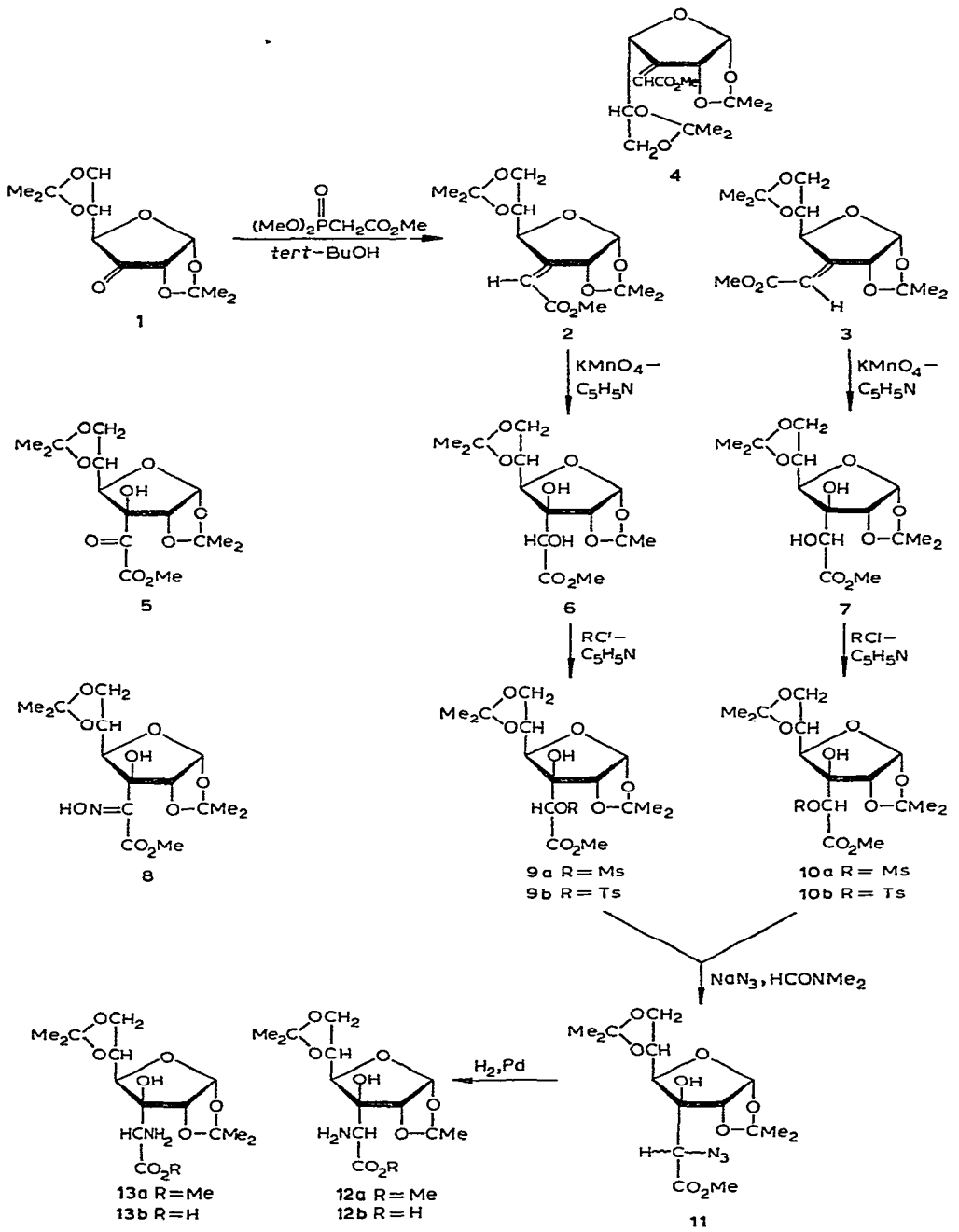
Stereospecific hydroxylation of 3-deoxy-1,2 5,6-di-*O*-isopropylidene-3-*C*-*trans*- and 3-*C*-*cis*-(methoxycarbonylmethylene)-α-D-*ribo*-hexofuranose (**2** and **3**, respectively), with potassium permanganate in pyridine afforded 3-*C*-[*S*- and *R*-hydroxy-(methoxycarbonyl)methyl]-1,2 5,6-di-*O*-isopropylidene-α-D-glucofuranose, (**6** and **7**, respectively), in a combined yield, after chromatography, of 43% Selective formation of monomethanesulfonates (**9a** and **10a**) and *p*-toluenesulfonates (**9b** and **10b**), followed by treatment with sodium azide and reduction of the azide, afforded the methyl 2-D-(and 2-L-)(3-deoxy-1,2 5,6-di-*O*-isopropylidene-α-D-glucofuranos-3-yl)-glycinates (**12a** and **13a**, respectively) Basic hydrolysis of the latter compounds yielded 2-D- and 2-L-(3-deoxy-1,2 5,6-di-*O*-isopropylidene-α-D-glucofuranos-3-yl)glycine (**12b** and **13b**, respectively) The structures of the glycosyl amino acids were correlated with that of L-alanine by circular dichroism

DISCUSSION

Polyoxins A-L, found in 1965, are a group of antifungal agents produced by *Streptoamyces cacavi* var *asoensis* and are useful as an agricultural fungicide[1]

The structures of the polyoxins, established by degradative chemical studies[1,2], was confirmed by chemical synthesis[3–5] The principal structural features of all of the polyoxins include the following (*i*) possession of 5-amino-5-deoxy-D-allofurano-syluronic acid sugar moiety[2], and (*ii*) a unique L-amino acid moiety attached to C-4 of the ribofuranosyl ring

In view of the fact that the introduction of branching at C-3 on the sugar moiety of naturally occurring nucleosides has been found to result in interesting changes in the biological activity of the nucleosides[6], it seemed of interest to synthesize structural analogs of the sugar moiety of the polyoxins in which the amino acid moiety would be attached to C-3, rather than C-4, of the sugar In this paper, we report the synthesis of two analogs of the sugar moiety of the polyoxins, one having an α-L and the other possessing the α-D-amino acid moiety attached by a carbon–carbon bond to C-3 of the sugar moiety This paper is a follow-up to a previous paper[7] in which we



reported the stereospecific synthesis of 2-L-(3-deoxy-1,2-*O*-isopropylidene-α-D-allofuranos-3-yl)glycine

The first step in the synthesis involved utilization of the key intermediates

3-deoxy-1,2 5,6-di-*O*-isopropylidene-3-*C*-*trans*-(methoxycarbonylmethylene)-α-D-*ribo*-hexofuranose (**2**), a compound previously described[8], and in addition, the *cis* isomer of **2** (compound **3**) Both of these unsaturated sugars were obtained in a 3 1 mixture by condensing 1,2 5,6-di-*O*-isopropylidene-α-D-*ribo*-hexofuranos-3-ulose[9] (**1**) with phosphonoacetic acid trimethyl ester in the presence of potassium *tert*-butoxide at room temperature Hydroxylation of the *trans*-unsaturated sugar (**2**) with osmium tetraoxide, osmium tetraoxide–hydrogen peroxide, and potassium permanganate in pyridine, proceeded stereospecifically[7] to afford 3-*C*-[*S*-hydroxy(methoxycarbonyl)-methyl]-1,2 5,6-di-*O*-isopropylidene-α-D-glucofuranose (**6**) *A priori*, it could therefore be assumed that a similar reaction applied to the unsaturated sugar **3** would afford the diastereoisomeric diol **7** Because of the great difficulty of obtaining pure **3**, it was decided to use the mixture of unsaturated sugars **2** and **3** in the hydroxylation step in the hope that the resultant diols could be separated This indeed proved feasible, and the mixture of **2** and **3** afforded the mixture of diols **6** and **7** in the same ratio as that of the unsaturated sugars The diols were readily separated by column chromatography on t l c -grade silica gel with benzene–ethyl acetate as developer under pressure, to afford pure **6** and **7** in a combined yield (after chromatography) of 44% or greater, depending on control of the reaction conditions The structures of the diols were assigned on the basis of their optical rotatory dispersion (o r d) and circular dichroism (c d) spectra (see Fig 1) compared with that of lactic acid Based on mechanistic considerations[10], it was assumed that the permanganate ion would attack

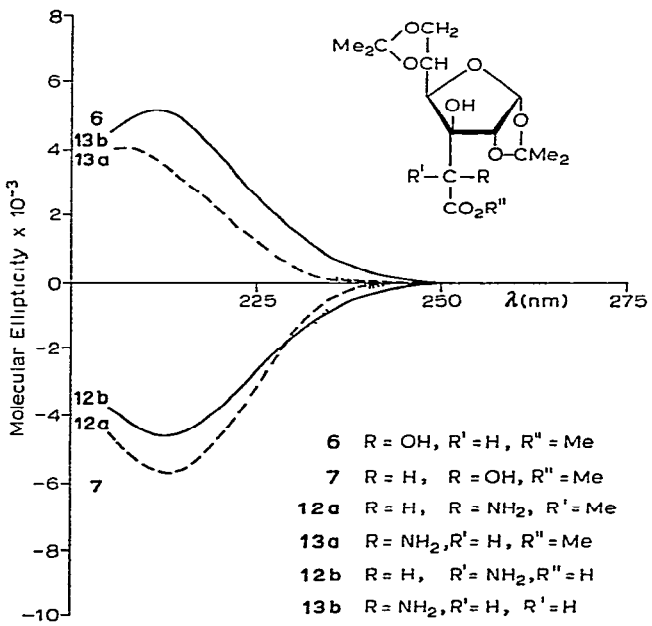


Fig 1 Circular-dichroism curves of branched-chain α-hydroxy ester sugars **6** and **7**, D- and L-(3-deoxy-glucos-3-yl)amino acid esters **12a** and **13a**, and D- and L-amino acids **12b** and **13b**

the *trans*-unsaturated sugar **2** from the less-hindered face of the molecule and therefore, the configuration of the carbon atom of the exocyclic branched chain would be the same as that of L-lactic acid[11] This indeed proved to be the case, and **6** is therefore suggested to be 3-*C*-[*S*-hydroxy(methoxycarbonyl)methyl]-1,2 5,6-di-*O*-isopropylidene-α-D-glucofuranose The diastereoisomeric diol **7** displayed the opposite c d curve to that of **6** and it therefore must be 3-*C*-[*R*-hydroxy(methoxycarbonyl)-methyl]-1,2 5,6-di-*O*-isopropylidene-α-D-glucofuranose

From the products arising from the hydroxylation of the unsaturated sugars there were isolated two additional products **1** and **5** in 19 and 3% yields, respectively The first proved to be identical with the ketose **1**, evidently arising by over-oxidation of the diols Prolonged reaction of the unsaturated sugars with permanganate increased the yield of the ketose. The minor component **5** exhibited two carbonyl peaks at 1720 and 1740 cm^{-1} in its i r spectrum, thus indicating an α-keto ester grouping The presence of the ketone group was confirmed by the fact that sodium borohydride reduction of **5** yielded the diols **6** and **7** In addition, treatment of **5** with hydroxylamine afforded a crystalline oxime, namely 1,2 5,6-di-*O*-isopropylidene-3-*C*-(methoxydi-carbonyl)-α-D-glucofuranose oxime (**8**)

Mesylation of the diols **6** and **7** afforded the monosulfonates **9a** and **10a** in 74% and 80% yields, respectively, after chromatography Similarly, tosylation of **6** and **7** afforded **9b** and **10b** in almost quantitative yields in both cases

Displacement of the tosyl, or preferably the mesyl, group with sodium azide in *N*,*N*-dimethylformamide under anhydrous conditions at 55–60° in the dark, and subsequent reduction to the α-amino ester, did not proceed stereospecifically as expected, but instead gave the same apparent equilibrium mixture of **12a** and **13a** independent of which sulfonic ester (**9a** or **10a**) was used The ratio of **12a** to **13a** was generally of the order 1 3 and was approximately the same for reaction times of 18 to 82 h In a typical reaction of 40-h duration, **12a** and **13a** were obtained in 17 and 51% yields, respectively, based on sulfonate consumed In addition, the reaction did not proceed to completion even after 120 h under the foregoing conditions The unreacted sulfonate was readily separated from the two α-amino esters by column chromatography on t l c -grade silica gel under pressure

The displacement reaction had to be conducted under strictly anhydrous conditions to prevent reversion of the pure sulfonic ester to a mixture of the diols **6** and **7** Hydrolysis of the methyl ester also occurred when the crude azide was hydrogenated in ethanol, methanol or ethyl acetate, and hence the hydrogenation was effected in ahydrous benzene over 5% palladium on charcoal, when reduction was complete in 1 25 h, with no apparent hydrolysis of the ester

The apparent lack of specificity in the formation of the α-amino ester **12a** from the sulfonate **9a**, and similarly **13a** from **10a**, is noteworthy This phenomenon might probably be due to the fact that each sulfonic ester is undergoing conversion into the α-azido esters by two competing reactions (*i*) firstly, in which the adjacent tertiary *C*-3 hydroxyl group participates, and (*ii*) secondly a reaction involving direct displacement of sulfonic ester by the azide ion In the first reaction, the hydroxyl group at

C-3 displaces, via an intramolecular mechanism, the sulfonate group, to afford an epoxide This epoxide may subsequently be attacked at the less-hindered, exocyclic position to afford the α-azido ester having the same stereochemistry as the starting diol Thus, the α-amino ester **13a** or **12a** produced on reduction of the azides via the first mechanism would have the same stereochemistry as the starting diol **6** or **7** In the second reaction, the sulfonate is displaced by the azide by an SN2 mechanism and thus the amino ester might be expected to have the opposite configuration from that of the starting diol As a consequence, each sulfonate might be expected to afford a diastereoisomeric mixture of products Support for this postulation is provided by the fact that the corresponding 3-deoxy analog *p*-toluenesulfonate of **10b** was converted exclusively into an α-amino ester having a configuration opposite from that of the starting compound[7]

The α-amino ester **13a**, assigned as methyl 2-L-(3-deoxy-1,2 5,6-di-*O*-isopropylidene-α-D-glucofuranos-3-yl)glycinate on the basis of its intensely positive Cotton effect[12] (see Fig 1), was carefully hydrolyzed in 1 25% aqueous methanolic sodium hydroxide (1 1 solution of methanol and 2 5% aqueous sodium hydroxide solution), at the completion of the reaction, as shown by t l c , the product was passed through a short column of Rexyn RG-51 (H^+) resin Elution with water afforded the unblocked amino acid 2-L-(3-deoxy-1,2 5,6-di-*O*-isopropylidene-α-D-glucofuranos-3-yl)glycine (**13b**), which crystallized in 85% yield on removal of the solvent under vacuum The c d spectrum of **13b** also exhibited a positive peak at 212 nm in 0 5M HCl in 95% ethanol, in agreement with the observed positive Cotton effects of other L-amino acids[12]

Similarly, hydrolysis of the α-amino ester (**12a**), yielded the crystalline α-amino acid (**12b**) Because both **12a** and **12b** exhibited intensely negative Cotton effects (see Fig 1), both possess the same stereochemistry at the chiral exocyclic carbon atom, and **12b** must be 2-D-(3-deoxy-1,2,5,6-di-*O*-isopropylidene-α-D-glucofuranos-3-yl)-glycine

The α-keto ester **5**, briefly mentioned as a component of the oxidation mixture of either the oxidation of pure *trans*-unsaturated sugar **2** or of the oxidation of a mixture of *trans*- and *cis*-unsaturated sugars, is a potentially useful compound as an intermediate in further amino acid synthesis[13,14] Attempts to obtain the α-keto ester by oxidation of either diol **6** or **7** (or in admixture) by the potassium permanganate–pyridine method resulted in total oxidative cleavage of the diol to afford the ketose **1** Similarly, treatment of **6** or **7** with ruthenium tetraoxide or Sarett oxidation failed to improve the yield of **5**

Treatment of **5** with hydroxylamine hydrochloride in pyridine resulted in the formation of the expected oxime **8** in 81% yield However, attempts to reduce the oxime **8** to an α-amino ester by using platinum oxide in methanol, at atmospheric pressure or at 40 lb in^{-2}, for periods of up to 3 days at room temperature, failed

EXPERIMENTAL

General — P m r. spectra were determined in deuteriochloroform solution with Me_4Si as the internal standard by using a Varian XL-100 spectrometer Optical rotations were measured at ambient temperature with a Perkin–Elmer model 141 automatic polarimeter. The c d measurements were performed with a Jasco J-20 Automatic Recording spectropolarimeter at room temperature, and i r spectra were recorded on a Perkin–Elmer 337 spectrometer. Column chromatography was performed on t l c -grade Silica Gel G, without binder, (Mondray) under a pressure of 4–8 lb in^{-2} and flow-rates of 70–140 ml/h T l c with Silica Gel G (Mondray) was used to monitor all reactions All melting points were determined on a Leitz microscope heating-stage, model 350, and are corrected Chemical analyses were performed by Mr P Borda, Microanalytical Laboratory, University of British Columbia, Vancouver.

Oxidation with potassium permanganate of the unsaturated esters (**2**) *and* (**3**) *to yield* 3-C-[S- *and* R-*hydroxy(methoxycarbonyl)methyl*]-*1,2 5,6-di*-O-*isopropylidene-α*-D-*glucofuranose* (**6** *and* **7**, *respectively*), *and 1,2 5,6-di*-O-*isopropylidene-3*-C-(*methoxydicarbonyl*)-*α*-D-*glucofuranose* (**5**) — According to the previously published procedure[1], a mixture of the unsaturated esters **2** and **3** (2 6 g) in water (20 ml) and pyridine (40 ml), maintained internally at −5°, was treated dropwise, with vigorous stirring, with a solution of potassium permanganate (1 4 g) in water (40 ml), added during 20 min The reaction mixture was extracted with chloroform (5 × 200 ml) The combined organic extracts were washed with water, dried over sodium sulfate, and evaporated to yield a yellow syrup (2 2 g) Column chromatography on t l c grade silica gel (120 g, column dimensions 4 × 25 cm) packed and eluted with 3.1 benzene–ethyl acetate, under a pressure of 8 lb in^{-2}., afforded the α-keto ester **5** (75 mg, 3%), ketose **1** (0 41 g, 19%), diol **7** (0 34 g, 12%), and diol **6** (0 89 g, 31%) An analytical sample of **5** was prepared by molecular distillation $[\alpha]_D^{22}$ +78 2° (*c* 0 7, chloroform), $\lambda_{max}^{CHCl_3}$ 3400 (OH), 1720 (C=O), 1740 cm^{-1} (CO_2Me), τ^{CDCl_3} 3 99 (d, 1, $J_{1,2}$ 4 Hz, H-1), 5 06 (d, 1, $J_{4,5}$ 7 Hz, H-4), 5 45 (d, 1, H-2), 6 01 (s, 1, OH)

Anal Calc for $C_{15}H_{22}O_9$ C, 52 02, H, 6 40 Found C, 52 05, H, 6 50

Analytical samples of **6** and **7** were prepared by molecular distillation at 105°/0 1 torr Diol **7** had R_F 0 25 (silica gel, 3 1 benzene–ethyl acetate, $[\alpha]_D^{22}$ +19° (*c* 1 6, chloroform), λ_{max}^{film} 3480 (OH), 1740 cm^{-1} (CO_2CH_3), o r d (*c* 0 07, ethanol $[\Phi]_{210}$ +2140°, $[\Phi]_{216}$ 0°, $[\Phi]_{220}$ −1960°, $[\Phi]_{228}$ −3510° (trough), $[\Phi]_{230}$ −3400°, $[\Phi]_{250}$ −1080°, $[\Phi]_{300}$ −258°, c d (*c* 0 13 ethanol) $[\theta]_{205}$ −6410°, $[\theta]_{210}$ −7640°, $[\theta]_{212}$ −7760° (trough), $[\theta]_{220}$ −5730°, $[\theta]_{230}$ −2180°, τ^{CDCl_3} 4 13 (d, 1, $J_{1,2}$ 4 Hz, H-1), 5.40–6 05 (m, 5), 6 20 (s, 3, CO_2Me), 6.30 (s, 1, OH, exchanges in D_2O), 6 36 (s, 1, OH, exchanged by D_2O)

Anal Calc for $C_{15}H_{24}O_9$· C, 51 72, H, 6 94 Found· C, 51 59, H, 6 99

Diol **6** had R_F 0 15, $[\alpha]_D^{22}$ +54° (*c* 1 5, chloroform), o r d (*c* 0 07, ethanol) $[\Phi]_{210}$ +950°, $[\Phi]_{220}$ +3530°, $[\Phi]_{224}$ +3770° (peak), $[\Phi]_{230}$ +3530°, $[\Phi]_{250}$ +1720°,

$[\Phi]_{300}$ +620°; c d (*c* 0 10, ethanol), $[\theta]_{205}$ +2800°, $[\theta]_{208}$ +5280° (peak), $[\theta]_{220}$ +3135°, $[\theta]_{230}$ +660°, τ^{CDCl_3} 4 15 (*d*, 1, $J_{1,2}$ 3 8 Hz, H-1), 5 56 (d, 1, H-2), 5 60–6 10 (overlapping peaks), 6 18 (s, 3, CO_2Me), 6 54 (s, 2, two OH, exchanged by in D_2O)

Anal Calc for $C_{15}H_{24}O_9$· C, 51 72, H, 6 94 Found C, 51 50, H, 6 93

1,2 5,6-Di-O-isopropylidene-3-C-[R-methylsulfonyloxy(methoxycarbonyl)methyl]-α-D-glucofuranose (**10a**) — Methanesulfonyl chloride (0 960 g) was added dropwise to a solution of **7** (0 965 g) in pyridine (15 ml) at 0°. After the solution had been stirred overnight at ~25°, dichloromethane (50 ml) and ice–water (50 ml) were added, and the resultant aqueous layer extracted with dichloromethane (2 × 25 ml) The combined extracts were washed with saturated sodium hydrogen carbonate solution (25 ml), water (25 ml), dried over calcium sulfate, and evaporated under diminished pressure to a brown oil that was then chromatographed on a column of t l c grade silica gel (50 g, column dimensions 4 × 15 cm) under a pressure of 8 lb in^{-2}, packed and eluted with 4 1 benzene–ethyl acetate, to afford **10a** (0 971 g, 80%) An analytical sample was prepared by recrystallization from ether–hexane and sublimation at 130°/0 1 torr, m p 126 5–127 0°, $[\alpha]_D^{33}$ +54 8° (*c* 1, chloroform), τ^{CDCl_3} 4 03 (d, 1, $J_{1\ 2}$ 3 5 Hz, H-1), 4 74 (s, H-1′), 5 56 (d, 1, H-2), 5 65–6 05 (overlapping peaks, 4), 6 14 (s, 3, CO_2Me), 6 22 (s, OH, exchanges in D_2O), 6 85 (s, 3, SO_3Me), 8 47-8 68 (4s, 12, 4Me)

Anal Calc for $C_{16}H_{26}O_{11}S$· C, 45 07; H, 6 15 Found C, 44 94; H, 6 23

1,2 5,6-Di-O-isopropylidene-3-C-[S-methylsulfonyloxy(methoxycarbonyl)methyl]-α-D-glucofuranose (**9a**) — To the diol **6** (0 700 g) in pyridine (8 ml) at 0°, was added methanesulfonyl chloride (0 700 g) After the solution had been stirred for 18 h, dichloromethane (25 ml) and ice–water (25 ml) were added The resulting aqueous phase was then extracted with dichloromethane (2 × 25 ml) The combined organic extracts were subsequently washed with a saturated sodium hydrogen carbonate solution (25 ml) followed by water (25 ml), dried over calcium sulfate, filtered, and evaporated under diminished pressure to yield an orange solid (0 806 g) Pressure chromatography on t l c -grade silica gel (40 g) packed and eluted with benzene–ethyl acetate (3 1) afforded **9a** as a clear syrup that crystallized on standing (0 641 g, 74%) An analytical sample was prepared by recrystallization from ether–hexane and sublimation at 145° and 0 1 torr, m p 160 0–160 5°, $[\alpha]_D^{25}$ +39 2° (*c* 0 4, chloroform), λ_{max}^{film} 3400 (OH), 1740 cm^{-1} (CO_2Me). τ^{CDCl_3} 4 10 (d, 1, $J_{1\ 2}$ 4 Hz, H-1), 4 44 (s, H-1′), 5 36 (d, 1, $J_{4\ 5}$ 6 Hz, H-4), 5 56 (d, 1, H-2), 5 55 (q 1, $J_{5,6}$ 6 Hz, H-5), 5 86 (m, 2, H-6), 6 19 (s, 3, CO_2Me), 6 54 (s, 1, OH, exchanged by with D_2O), 6 77 (s, 3, SO_3Me), 8 53-8 70 (4s, 12, 4Me)

Anal Calc for $C_{16}H_{26}O_{11}S$ C, 45 07, H, 6 15 Found C, 44 95, H, 6 09

1,2 5,6-Di-O-isopropylidene-3-C-[R-p-tolylsulfonyloxy(methoxycarbonyl)methyl]-α-D-glucofuranose (**10b**) — The diol **7** (0 110 g) and *p*-toluenesulfonyl chloride (0 300 g) in pyridine (2 ml) were stirred for 18 h at room temperature Water (10 ml) was added and the solution was extracted with chloroform The chloroform phase was dried and evaporated under diminished pressure to yield a crystalline product (0 155 g, 97%) that was recrystallized from chloroform–hexane (or methanol) to afford fine

needles, m p 185–186°, $[\alpha]_D^{24}$ +40 0° (*c* 2 2, chloroform), τ^{CDCl_3} 4 17 (d, 1, $J_{1\,2}$ 3 5 Hz, H-1), 4 52 (s, 1, H-1'), 5 4-6 1 (m, 5), 6 95 (s, 1, OH)

Anal Calc for $C_{22}H_{30}O_{11}S$· C, 52 55, H, 6 12 Found C, 52 60, H, 6 01

1,2 5,6-Di-O-*isopropylidene-3*-C-[S-p-*tolylsulfonyloxy(methoxycarbonyl)methyl*]-α-D-*glucofuranose* (**9b**) — Diol **6** (0 030 g) in pyridine (1 ml) and *p*-toluenesulfonyl chloride (0 040 g) were maintained for 18 h at room temperature Water (5 ml) was added, and the solution was extracted with chloroform to give a crystalline sulfonate in quantitative yield An analytical sample was recrystallized from ethanol–hexane, m p 132–133°, $[\alpha]_D^{25}$ +72° (*c* 1 2, chloroform), τ^{CDCl_3} 4 19 (d, 1, $J_{1\,2}$ 3 5 Hz, H-1), 4 93 (s, 1, H-1'), 5 75 (d, 1, H-1), 6 25 (s, 1, OH)

Anal Calc for $C_{22}H_{30}O_{11}S$ C, 52 55, H, 6 12 Found C, 52 32, H, 5 95

Methyl 2-D-(*3-deoxy-1,2 5,6-di*-O-*isopropylidene*-α-D-*glucofuranos-3-yl)glycinate* (**12a**) *and methyl 2*-L-(*3-deoxy-1,2 5,6-di*-O-*isopropylidene*-α-D-*glucofuranos-3-yl)glycinate* (**13a**) — Methanesulfonate **9a** (300 mg) and sodium azide (300 mg) in *N,N*-dimethylformamide (20 ml were warmed for 40 h at 55–60° (not higher) in the dark, after which time the reaction mixture was evaporated to dryness The residue was suspended in dichloromethane and filtered Evaporation of the solvent afforded a clear syrup (295 mg) that revealed two spots on t l c , R_F 0 66 and R_F 0 30 (silica gel, 4 1 benzene–ethyl acetate), the faster-moving spot corresponding to the azido sugar **11**, and the slower component to unreacted sulfonate Benzene (15 ml) and 5% palladium on charcoal (150 mg) were added immediately and the mixture was hydrogenated for 1 25 h at room temperature and atmospheric pressure Filtration and evaporation of the solution afforded a clear syrup (293 mg) that showed that the faster-moving α-azido-ester had been reduced to give two ninhydrin-positive components R_F 0 58 (major) and R_F 0 47 (minor) (silica gel, ethyl acetate) whereas the methanesulfonate remained unchanged Preparative t l c (6 plates of 20 × 20 cm) afforded the α-amino esters (96 mg, 70% based on sulfonate consumed) and unreacted sulfonate (126 mg) Column chromatography of the partly purified α-amino esters on t l c -grade silica gel (60 g) packed and eluted with 1 9 benzene–ethyl acetate under 8 lb in^{-2} pressure, afforded two pure compounds

Compound **12a** (18 mg, 17%) twice distilled at 105°/0 1 torr had $[\alpha]_D^{28}$ +33 5° (*c* 2 3, chloroform), λ_{max}^{film} 3400 (OH, NH_2), 1740 cm^{-1} (CO_2Me), c d (*c* 0 19, 95% ethanol) $[\theta]_{203}$ −4070°, $[\theta]_{210}$ −5430°, $[\theta]_{213}$ −5700° (trough), $[\theta]_{220}$ −4710°, $[\theta]_{230}$ −1550°, τ^{CDCl_3} 4 13 (d, 1, $J_{1\,2}$ 4 Hz, H-1), 5 57 (d, 1, H-2), 5 50–6 15 (overlapping multiplets), 6 21 (s, 3, CO_2Me), 7 45 (broad singlet, 3, OH and NH_2 exchanges in D_2O), 8 48 (s, Me), 8 56 (s, Me), 8 67 (s, Me), 8 69 (s, Me) Irradiation at τ 4 13 collapsed the doublet at τ 5 57 to a singlet

Anal Calc for $C_{15}H_{25}N_8O$ C, 51 88, H, 7 25, N, 4 03 Found C, 51 91, H, 7 29, N, 3 95

Compound **13a** (59 mg, 51%) twice distilled at 105°/0 1 torr had, $[\alpha]_D^{27}$ +53° (*c* 1, chloroform), λ_{max}^{film} 3400° (OH, NH_2), 1740 cm^{-1} (CO_2Me), c d (*c* 0 19, 95% ethanol) $[\theta]_{203}$ +3620°, $[\theta]_{207}$ +4070° (peak), $[\theta]_{220}$ +2260°, $[\theta]_{230}$ +450°, τ^{CDCl_3} 4 16 (d, 1, $J_{1\,2}$ 3 5 Hz, H-1), 5 71 (d, 1, H-2), 5 45–6 15 (overlapping multiplets),

6 25 (s, 3, CO_2Me), 7 16 (broad s, 3, OH, NH_2, exchanges in D_2O), 8 56 (s, two Me), 8 64 (s, Me), 8 74 (s, Me) Irradiation at τ 4 16 collapsed the doublet at τ 5 71 to a singlet

Anal Calc for $C_{15}H_{25}NO_8$ C, 51 88, H, 7 25, N, 4 03 Found C, 51 77, H, 7 26, N, 3 93

2-L-(3-Deoxy-1,2 5,6-di-O-isopropylidene-α-D-glucofuranos-3-yl)glycine (**13b**) — A solution of the α-amino ester **13a** (48 mg) in 1 25% aqueous methanolic sodium hydroxide (2 ml of 1 1 solution) was stirred for 25 min (reaction completed as indicated by t l c on silica gel with ethyl acetate as solvent, the free acid remained at the origin) and then passed through 15 ml of Rexyn RG-51 (H^+) (polystyrenecarboxylic acid type resin), that had been prewashed with 1% acetic acid and then water until the effluent was neutral The column was eluted with water, fractions giving a positive ninhydrin test were combined and evaporated under diminished pressure to yield the crystalline amino acid **13b** (39 mg, 85%) An analytical sample was recrystallized from ethanol, m p 185 5–186 5° (dec), $[\alpha]_D^{29}$ +51 1° (*c* 1, water), c d (*c* 0 15, 0 5M HCl in 95% ethanol), $[\theta]_{200}$ +3860°, $[\theta]_{212}$ +5110° (peak), $[\theta]_{220}$ +3750°, $[\theta]_{230}$ +1480°, $[\theta]_{240}$ +340° The c d spectrum was taken within 10 min, τ^{D_2O} (external Me_4Si) 3 99 (d, 1, $J_{1,2}$ 3 8 Hz, H-1), 5 23 (d, 1, H-2), 5 59–6 00 (m, 4), 5 92 (s, 1, H-1′), 8 43 (s, Me), 8 49 (s, Me), 8 55 (s, Me), 8 61 (s, Me)

Anal Calc for $C_{14}H_{23}NO_8$ 10 $5H_2O$ C, 49 12, H, 7 06, N, 4 09 Found C, 49 23, H, 6 84, N, 4 14

2-D-(3-Deoxy-1,2 5,6-di-O-isopropylidene-α-D-glucofuranos-3-yl)glycine (**12b**) — A solution of **12a** (17 mg) in 1 25% aqueous methanolic sodium hydroxide (0 5 ml of 1 1 solution) was stirred at room temperature T l c (silica gel, ethyl acetate) indicated complete reaction after 15 min Elution with water through 10 ml of Rexyn RG-51 (H^+) (prewashed with 1% acetic acid and then with water) and evaporation of the fractions that afforded a ninhydrin-positive test gave the crystalline amino acid **12b** (12 mg, 75%) An analytical sample was recrystallized from ethanol, m p 193 5–195 0°, $[\alpha]_D^{27}$ +35° (*c* 0 9, water), c d (*c* 0 14, 0 5M hydrochloric acid in 95% ethanol) $[\theta]_{200}$ −2810°, $[\alpha]_{212}$ −4650° (trough), $[\theta]_{220}$ −3790°, $[\theta]_{230}$ −1590°, $[\theta]_{240}$ −245° The c d spectrum was determined within 10 min, τ^{D_2O} (external Me_4Si) 3 92 (d, 1, $J_{1,2}$ 4 Hz, H-1), 5 37 (d, 1, H-2), 5 40–6 05 (overlapping peaks), 8 40 (s, Me), 8 51 (s, Me), 8 60 (s, two Me)

Anal Calc for $C_{14}H_{23}NO_8$ $2H_2O$ C, 45 51, H, 7 37, N, 3 80 Found C, 45 72, H, 7 27, N, 3 60

Reduction by sodium borohydride of 1,2 5,6-di-O-isopropylidene-3-C-(methoxydicarbonyl)-α-D-glucofuranose (**5**) — To **5** (0 040 g) in anhydrous methanol (2 ml) at 0°, was added sodium borohydride (0 020 g) The solution was kept for 5 h at 0° and for a further 5 h, at room temperature and then evaporated to dryness under diminished pressure Water (10 ml) was added and the solution was extracted with chloroform (2 × 10 ml) The combined chloroform extracts were dried and evaporated under diminished pressure to an oil (0 035 g) T l c (silica gel, 3 1 benzene–ethyl acetate), revealed the presence of the diols **7** and **6** N m r spectroscopy indicated

the ratio of **7** to **6** to be 3 1 The mixture was subjected to preparative t l c (silica gel, 3 1 benzene–ethyl acetate, three times developed) to afford pure **7** (0 020 g) and pure **6** (0 007 g) Both were identified by n m r spectroscopy and by comparison with previously obtained samples

Preparation of 1,2 5,6-di-O-isopropylidene-3-C-(methoxydicarbonyl)-α-D-glucofuranose oxime (**8**) — A mixture of **5** (0 130 g) and hydroxylamine hydrochloride (0 200 g) in pyridine was heated for 3 h at 100° Pyridine was then removed under vacuum, and water (10 ml) added The resulting solution was extracted with chloroform (3 × 10 ml) After combination, drying, and evaporation of the organic extracts there was obtained **8** (0 110 g, 81%) as a compound homogeneous by t l c A portion of the product was sublimed at 110°/0 1 torr to give a white solid; m p 45°, $[\alpha]_D^{25}$ +73° (*c* 0 5, chloroform), τ^{CDCl_3} 4 10 (d, 1, $J_{1,2}$ 3 Hz, H-1), 5 35 (d, 1, $J_{4,5}$ 4 Hz, H-4), 5 56 (d, 1, H-2)

Anal Calc for $C_{15}H_{23}NO_9$· C, 49 86, H, 6 43, N, 3 88 Found C, 50 00, H, 6 64, N, 4 12

ACKNOWLEDGMENT

The authors thank the National Research Council of Canada for financial support.

REFERENCES

1 (a) S SUZUKI, K ISONO, J NAGATSU, T MIZUTANI, Y KAWASHIMA, AND T MIZUNO, *J Antibiotics*, A18 (1965) 131, (b) K ISONO, J. NAGATSU, Y KAWASHIMA, AND S SUZUKI, *Agr. Biol Chem*, 29 (1965) 848, (c) K ISONO, J NAGATSU, Y KAWASHIMA, H YAMAGATA, K SASAKI, AND S SUZUKI, *Agr Biol Chem*, 31 (1967) 190, (d) K ISONO, K ASAHI, S SUZUKI, *J Amer Chem Soc*, 91 (1969) 7490
2 (a) K ISONO AND S SUZUKI, *Tetrahedron Lett*, (1968) 203, (b) K ISONO AND S SUZUKI, *Tetrahedron Lett*, (1968) 1133
3 T NAKA, T HASHIZUME, AND M NISHIMURA, *Tetrahedron Lett*, (1971) 95
4 N P DAMODARAN, G H JONES, AND J G MOFFATT, *J Amer Chem Soc*, 93 (1971) 3812
5 H OHRUI, H KUZUHARA, AND S EMOTO, *Tetrahedron Lett*, (1971) 4267
6 (a) R F NUTT, M J DICKINSON, F W HOLLY, AND E WALTON, *J Org Chem*, 33 (1968) 2490, (b) E WALTON, S R JENKINS, R F NUTT, AND F W HOLLY, *J Med Chem*, 12 (1969) 308
7 A ROSENTHAL AND K SHUDO, *J Org Chem*, 37 (1972) 4391
8 A ROSENTHAL AND L NGUYEN, *J Org Chem*, 34 (1969) 1029
9 (a) P J BEYNON, P M COLLINS, AND W G OVEREND, *Proc Chem Soc*, (1964) 342, (b) K ONODERA, S HIRANO, AND N KASHIMURA, *J Amer Chem Soc*, 87 (1965) 4651, (c) K ONODERA, S HIRANO, AND N KASHIMURA, *Carbohyd Res*, 6 (1968) 276
10 (a) H O HOUSE, in "*Modern Synthetic Reactions*", W A BENJAMIN Inc, New York, N Y, 1965, p 142, (b) F D GUNSTONE, in R A RAPHAEL, E C TAYLOR, AND H WINBERG (Eds), *Advances in Org Chem, Methods and Results*, Vol 1, J Wiley, Interscience, New York, N Y (1960), 103–147, (c) M FIESER, A QUILICO, A NICKON, W E ROSEN, E J TARLTON, AND L F. FIESER, *J Amer Chem Soc*, 75 (1953) 4066
11 J C CRAIG AND S K ROY, *Tetrahedron*, 21 (1965) 1847
12 J C CRAIG AND S K ROY, *Tetrahedron*, 21 (1965) 391
13 E J COREY, RONALD J MCCAULLY, AND H S SACHDEV, *J Amer Chem Soc*, 92 (1970) 2476, and references therein
14 E J COREY, HARBANS S SACHDEV, J ZANOS GOUGOUTAS, AND W SAENGER, *J Amer Chem Soc*, 92 (1970) 2488