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Preliminary communication

A convenient synthesis of 2-deoxy-2-(D- and -(L-3-hydroxytetradecanoylamino)-D-glucose: diastereoisomers of the monomeric, lipid A component of the bacterial lipopolysaccharide

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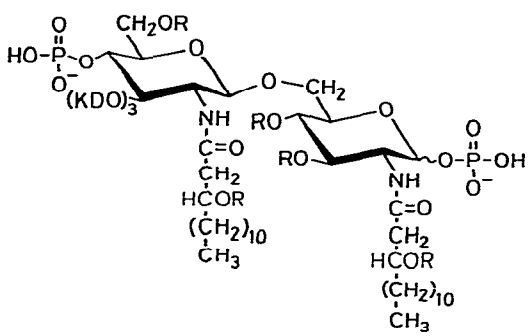
Lipid A is a unique, hydrophobic component of the endotoxic, bacterial lipopolysaccharide (LPS), and, for *Salmonella*[1–3] and *Escherichia coli*[1,4], it has been concluded that its fundamental structure is a monomer of a β-D-(1→6)-linked disaccharide (1) of 2-deoxy-2-(D-3-hydroxytetradecanoylamino)-D-glucopyranose esterified with fatty acyl and phosphate groups. Amide-linked D-3-hydroxytetradecanoic acid[5] is a common and prominent constituent that constitutes a characteristic marker of lipid A. On the other hand, it has been suggested that the lipid A component plays an important role in a variety of biological activities of LPS, but chemical investigations of the structure and activity have not so far been satisfactory.



1

R = fatty acyl residue or H

KDO = 3-deoxy-D-*manno*-octulosonate

As outlined in a previous paper[6], we have recently developed a new synthetic route to analogs of the lipid A component, and have found that ferric chloride-catalyzed glycosylation[7,8] is useful for the preparation of *N*-fatty acylated 2-amino-2-deoxy-β-D-glucopyranosides. We now describe a convenient preparation of optically active 2-deoxy-2

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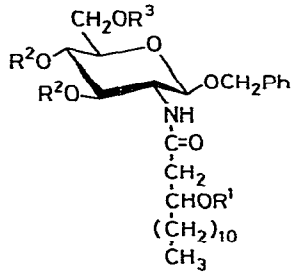
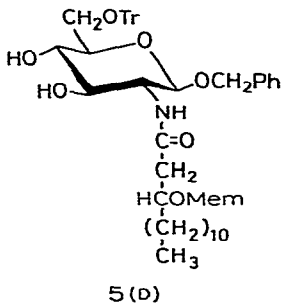
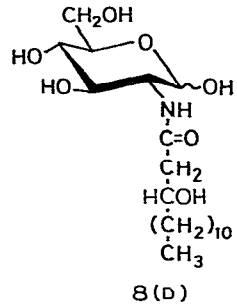


2 R^1 = H, R^2 = R^3 = Ac
3 R^1 = Mem, R^2 = R^3 = Ac
4 R^1 = Mem, R^2 = R^3 = H
5 R^1 = Mem, R^2 = H, R^3 = Tr
6 R^1 = R^2 = R^3 = Ac
7 R^1 = R^2 = R^3 = H

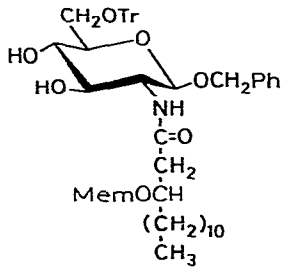
Mem = $MeO(CH_2)_2OCH_2$
Tr = Ph_3C



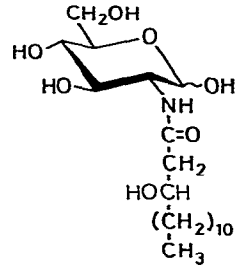
5(D)



8(D)



5(L)



8(L)

(D- and -(L-3-hydroxytetradecanoylamino)-D-glucose [8(D) and 8(L)] starting from diastereoisomeric benzyl 3,4,6-tri-*O*-acetyl-2-deoxy-2-(DL-3-hydroxytetradecanoylamino)-β-D-glucopyranoside[6] (2).

Treatment of compound 2 with (2-methoxyethoxy)methyl (Mem) chloride[9] in dichloromethane containing diisopropylethylamine gave 3 in almost quantitative yield. *O*-Deacetylation of 3, to give 4, and selective tritylation of the primary hydroxyl group on C-6 of 4 afforded 5(D) and 5(L) in the ratio of ~1 : 1. These showed different mobilities in t.l.c., and were readily separable by chromatography on a column of silica gel; 5(D) was eluted slightly faster than 5(L) with 70 : 1 (v/v) chloroform–methanol. Both compounds had elemental compositions agreeing with the theoretical values, and showed the same mass spectrum, but 5(D) was an amorphous material, whereas 5(L) was crystalline. In the 90-MHz, ^{1}H-n.m.r. spectra of 5(D) and 5(L) in chloroform-*d*, the anomeric protons appeared as doublets (*J* 8.0 Hz) at δ 4.64 and 4.45, respectively, but no other remarkable difference was observed, except that the methylenic protons on C-2 of the 3-hydroxytetradecanoyl group showed complex, but clearly different, signal patterns at δ ~2.4. The physical properties of the benzyl 2-deoxy-2-(D-, -(L-, and -(DL-3-hydroxytetradecanoylamino)-D-glucopyranoside derivatives are summarized in Table I.

The optically pure samples of 5(D) and 5(L) thus obtained were, by mild hydrolysis with 70% acetic acid, separately converted into 4(D) and 4(L), and these, by acetylation, respectively into 3(D) and 3(L) in quantitative yield. Removal of the Mem group was achieved by treatment of 3(D) and 3(L) with ferric chloride in dichloromethane.

TABLE I

PHYSICAL PROPERTIES OF BENZYL 2-DEOXY-2-(3-HYDROXYTETRADECANOYLAMINO)-β-D-GLUCOPYRANOSIDE DERIVATIVES

Compound	*M.p.* (°C)	$[\alpha]_D$ (degrees)[a] (c)	*Compound*	*M.p.* (°C)	$[\alpha]_D$ (degrees)[a] (c)	*Compound*	*M.p.* (°C)	$[\alpha]_D$ (degrees)[a] (c)
3(D)	103–104[b]	−23.8 (1.463)	3(L)	81–81.5[b]	−14.6 (1.446)	3(DL)	amorph.	−19.8 (0.479)
4(D)	amorph.	−20.3[c] (1.203)	4(L)	amorph.	−32.2[c] (1.189)			
5(D)	amorph.	−52.0 (0.911)	5(L)	107[d]	−35.2 (0.381)			
6(D)	148–149[b]	−25.1 (0.522)	6(L)	124–125[b]	−20.9 (0.435)	6(DL)	amorph.	−21.9 (0.530)
7(D)	amorph.	−22.7[c] (0.463)	7(L)	amorph.	−23.9[c] (0.368)			

[a] Specific rotations (concentration, *c*) determined in chloroform, except as noted. [b] Crystallized from ethanol. [c] Specific rotation determined in methanol. [d] Crystallized from ether–hexane.

to yield 2(D) and 2(L), quantitatively. For characterization, these were reacetylated to 6(D) and 6(L). The 90-MHz, ^{1}H-n.m.r. spectra of 6(D) and 6(L) in chloroform-*d* (see Fig. 1) were quite similar to each other, but could be clearly distinguished by comparing the chemical shift of the highest-field OAc group [6(D), s, δ 1.99 (arrow I); 6(L), s, δ 1.97 (arrow I′)], which corresponds to the one on the asymmetric carbon atom (C-3) of the 3-hydroxytetradecanoyl group. Moreover, the signal patterns of the methylenic protons at C-2 of the same moiety were remarkably different [6(D), ~dd, δ 2.37 (arrow II); 6(L), ~d, δ 2.38 (arrow II′)]. In fact, the racemate[6] 6(DL) showed both signals, the same as observed for 6(D) and 6(L), in the ratio of ~1 : 1.

O-Deacetylation of 6(D) and 6(L), and hydrogenolytic removal of the benzyl group in the presence of 10% palladium–carbon catalyst, gave the desired, optically pure 2-deoxy-2-(D- and -(L-3-hydroxytetradecanoylamino)-D-glucoses: 8(D), $[\alpha]_D$ +23.7° (*c* 0.245, 2 : 1 (v/v) oxolane–water; equil.], +25.8° [*c* 0.240, 3 : 1 (v/v) methanol–chloroform; equil.]; and 8(L), $[\alpha]_D$ + 35.0° [*c* 0.240, 2 : 1 (v/v) oxolane–water; equil.], as amorphous materials. The two compounds showed different solubility in methanol–chloroform, in which 8(L) was essentially insoluble.

Very recently, Demary[10] synthesized 8(D), $[\alpha]_D$ + 26.9° [*c* 0.134, 2 : 1 (v/v) oxolane–water, equil.], by direct condensation of activated D-3-hydroxytetradecanoic acid, which had been resolved by means of dehydroabiethylamine, with 2-amino-2-deoxy-D-glucose. We next prepared 1,3,4,6-tetra-*O*-acetyl-2-deoxy-2-(D-3-hydroxytetradecanoylamino)-β-D-glucopyranose (**10**) by treatment of 1,3,4,6-tetra-*O*-acetyl-2-amino-2-deoxy-β-D-glucopyranose[11] (**9**) with activated D-3-hydroxytetradecanoic acid, and then derivatized it to 2-(D-3-acetoxytetradecanoylamino)-1,3,4,6-tetra-*O*-acetyl-2-deoxy-β-D-

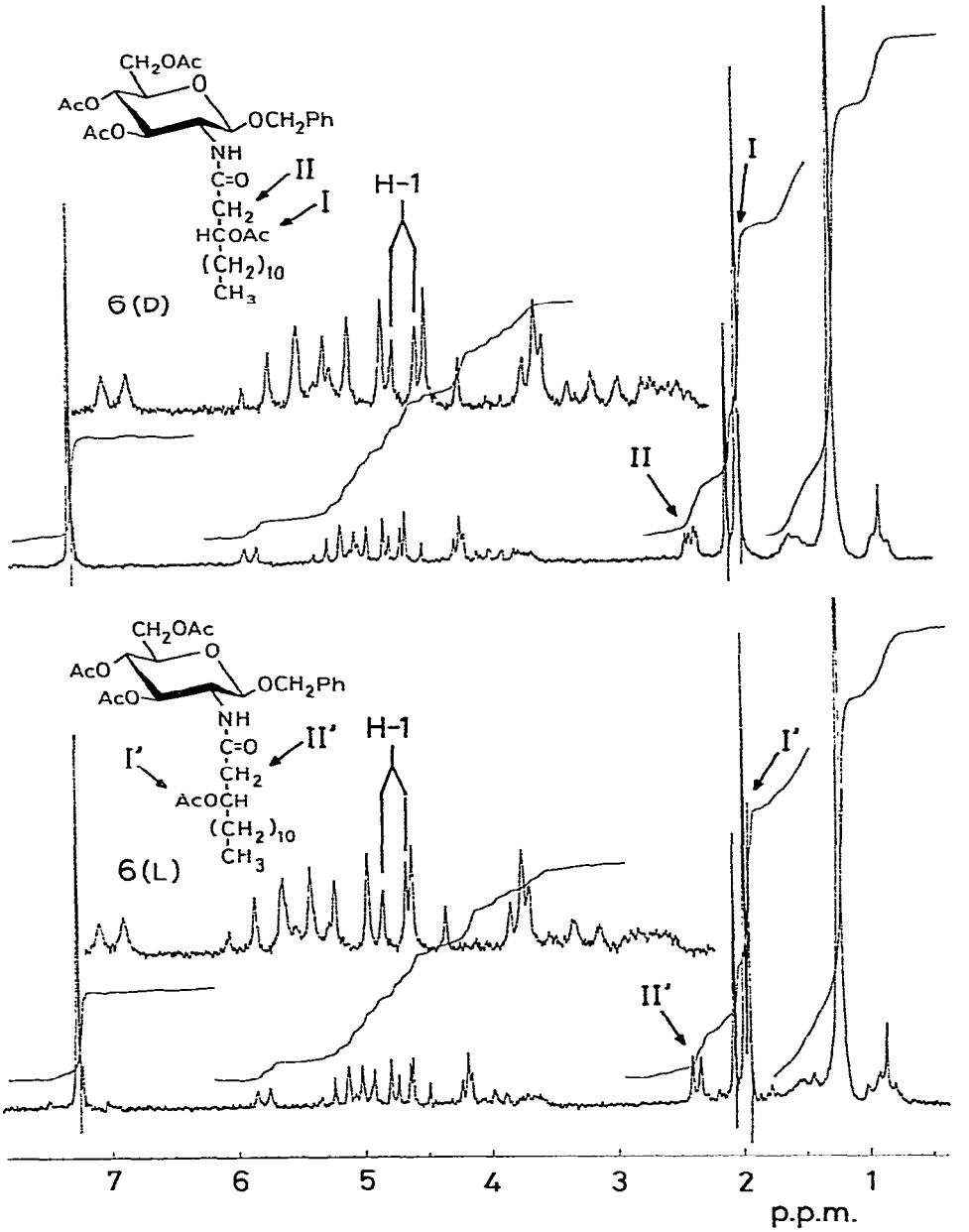
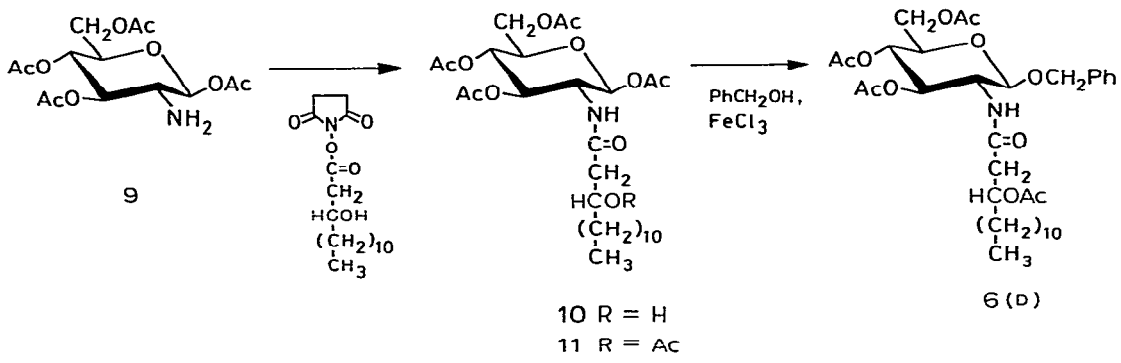


Fig. 1. ^{1}H-N.m.r. spectra (90 MHz) of 6(D) (D-3-hydroxytetradecanoyl derivative) and 6(L) (L-3-hydroxytetradecanoyl derivative) in chloroform-*d*.

glucopyranose (**11**). Compound **11** was readily converted into benzyl-2-(D-3-acetoxytetradecanoylamino)-3,4,6-tri-*O*-acetyl-2-deoxy-β-D-glucopyranoside [**6**(D)] by the ferric chloride-catalyzed glycosylation procedure[6—8]. The spectral data and physical properties of the benzyl glycoside were identical with those previously found for **6**(D).



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