

Communications to the Editor

[Chem. Pharm. Bull.]
33(9)4098-4101(1985)

EFFICIENT SYNTHESIS OF NOVEL MONOSACCHARIDE ANALOGS OF LIPIDS A¹⁾

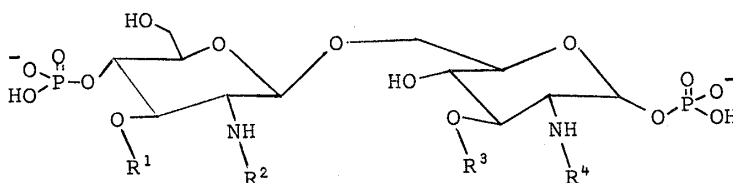
Shinichi Nakamoto, Toshio Takahashi, Kiyoshi Ikeda, and Kazuo Achiwa*
Shizuoka College of Pharmacy, 2-2-1 Oshika, Shizuoka 422, Japan

The efficient synthesis of the monosaccharide analogs of lipids A bearing two 3-acyloxytetradecanoyl and phosphoryl groups at the C-2,3 and C-4 positions of the glucosamine skeleton is described. Also a preliminary analysis of their biological activities is presented.

KEYWORDS — lipid A analog; glucosamine derivative; 4-phosphorylated monosaccharide; endotoxic lethal activity; antitumor activity

Although many attempts have been made to synthesize lipids A and related analogs according to their wrongly assigned structures,²⁾ few synthetic and biological works based on the reversed structure of lipids A³⁾ (1a-d) have been reported.^{1,4)}

We describe here a new synthesis of the monosaccharide analogs (2a-d) of lipids A according to the corrected structures, and the preliminary results of their biological activities.

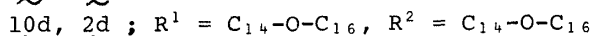
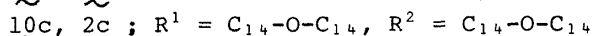
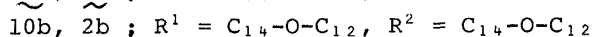
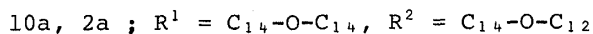
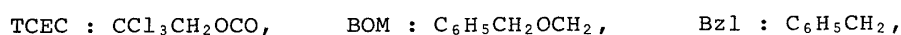
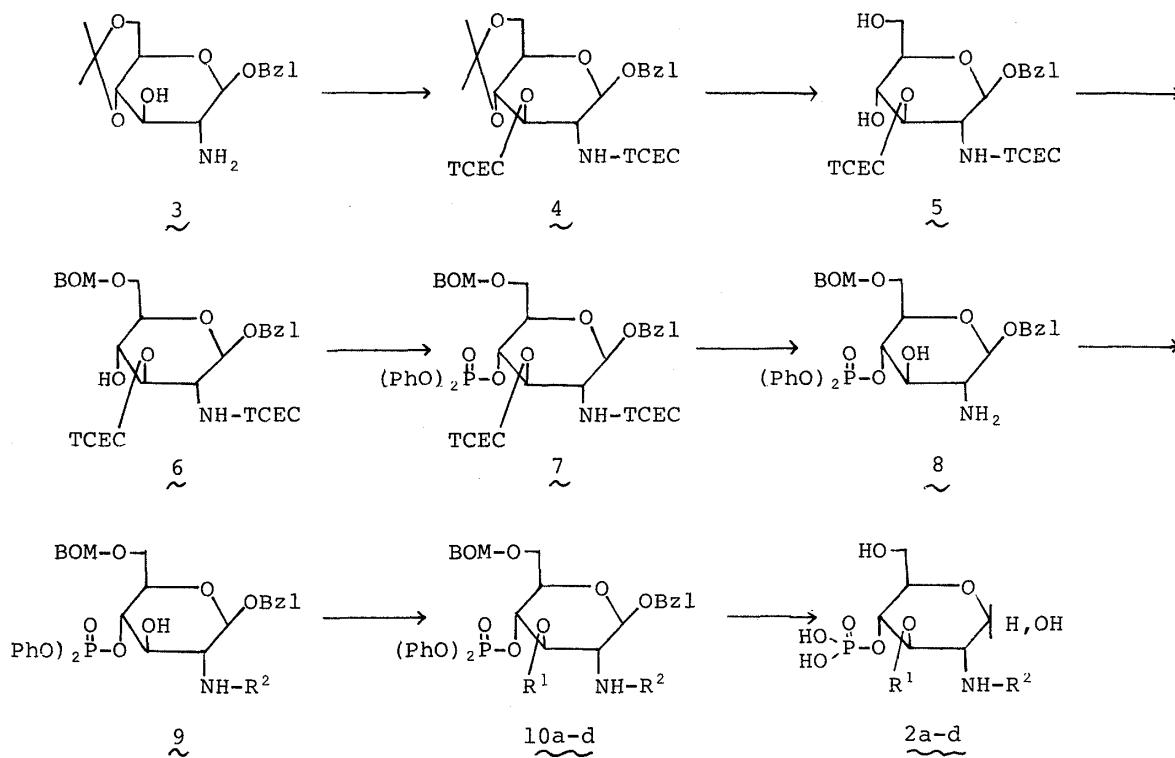


- 1a; R¹ = C₁₄-O-C₁₄, R² = C₁₄-O-C₁₂, R³ = R⁴ = C₁₄-OH
 1b; R¹ = C₁₄-O-C₁₄, R² = C₁₄-O-C₁₂, R³ = C₁₄-OH, R⁴ = C₁₄-O-C₁₆
 1c; R¹ = C₁₄-O-C₁₄, R² = C₁₄-O-C₁₄, R³ = R⁴ = C₁₄-OH
 1d; R¹ = C₁₄-O-C₁₄, R² = C₁₄-O-C₁₄, R³ = C₁₄-OH, R⁴ = C₁₄-O-C₁₆

C₁₄-OH: (R)-3-Hydroxytetradecanoyl, C₁₄-O-C₁₂: (R)-3-dodecanoyloxytetradecanoyl, C₁₄-O-C₁₄: (R)-3-tetradecanoyloxytetradecanoyl, C₁₄-O-C₁₆: (R)-3-hexadecanoyloxytetradecanoyl.

In the previously reported synthesis of the monosaccharide analogs of lipids A, the only method developed was the introduction of the desired fatty acid moiety at the C-2 and C-3 positions of the glucosamine skeleton in the early stage of the synthesis process.⁵⁾

Our present strategy includes the introduction of optically active fatty acid moieties at the desired positions in the last stage. Thus, the monosaccharide 4-phosphate (8) bearing one amino and one hydroxyl group at the C-2 and C-3 positions of the glucosamine skeleton was exploited as the key common intermediate. Efficient conversion of 8 into several 4-phosphorylated monosaccharides (2a-d) substituted with suitable fatty acid groups proceeded as shown below.



The compound (4)⁶⁾ [96%, mp 65-68°C, $[\alpha]_D^{22} -32.5^\circ$ (c=1.00, CHCl_3)] was readily prepared by treating the free amino and hydroxyl groups of benzyl 2-amino-2-deoxy-4,6-isopropylidene- β -D-glucopyranoside (3)⁶⁾ with 2,2,2-trichloroethoxycarbonyl chloride in the presence of a catalytic amount of 4-dimethylaminopyridine at room temperature for 2 h.⁷⁾ Subsequent removal of the isopropylidene group of 4 was accomplished by hydrolysis with aqueous 90% acetic acid at 90°C for 15 min to yield 5⁶⁾ [98%, mp 102-103°C, $[\alpha]_D^{18} -26.2^\circ$ (c=1.20, CHCl_3)]. Treatment of the diol (5) with benzyloxymethyl chloride and tetramethylurea in CH_2Cl_2 at room temperature for 16 h followed by purification with silica gel column chromatography afforded the 6-benzyloxymethyl ether (6)⁶⁾ [85%, amorphous, $[\alpha]_D^{23} -24.2^\circ$ (c=0.28, CHCl_3)]. Phosphorization of 6 was carried out with diphenyl phosphorochloridate,

pyridine and 4-dimethylaminopyridine in benzene.^{4b)} The reaction was complete in 2 h at room temperature to give 7⁶⁾ [87%, mp 119-120°C, $[\alpha]_D^{23}$ -8.57° (c=0.98, CHCl₃)]. Deprotection of the 2,2,2-trichloroethoxycarbonyl group by treatment with zinc powder in acetic acid at room temperature for 5 h⁸⁾ afforded 8⁶⁾ [96%, amorphous, $[\alpha]_D^{21}$ -9.50° (c=1.14, CHCl₃)]. Acylation of this common intermediate with the desired acyl groups proceeded smoothly. Thus the amino-hydroxy-compound (8) was first acylated at the amino group with (R)-3-dodecanoyloxytetradecanoic acid in the presence of dicyclohexylcarbodiimide in CH₂Cl₂ at 0-5°C to yield 9a⁶⁾ [68%, mp 97-99°C, $[\alpha]_D^{23}$ -13.1° (c=0.61, CHCl₃)] and then at the hydroxyl group with (R)-3-tetradecanoyloxytetradecanoic acid, dicyclohexylcarbodiimide, and 4-dimethylaminopyridine in the same solvent⁹⁾ to give 10a⁶⁾ [70%, mp 71-73°C, $[\alpha]_D^{25}$ -8.94° (c=0.94, CHCl₃)]. The protective benzyl and phenyl groups of 10a were removed stepwise by hydrogenolysis catalyzed by 10% Pd-on-carbon at 45°C for 5 h and PtO₂ at room temperature for 16 h in methanol⁸⁾ to yield 2a⁶⁾ [56%, mp 116-118°C, $[\alpha]_D^{25}$ +33.5° (c=0.40, CHCl₃)]. Similarly, the diacylated compounds (10b-d)^{6,9)} were obtained by simultaneous acylation of the amino and hydroxyl groups of 8 with the corresponding fatty acids in the presence of dicyclohexylcarbodiimide and 4-dimethylaminopyridine in CH₂Cl₂ at room temperature for 16 h and the respective monosaccharide analogs of lipids A (2b-d)^{6,10)} were afforded by hydrogenolysis as described above for 2a.

Preliminary studies of the biological activity of 2a-d revealed that the order of potency of the endotoxic lethal activity was 2a>2c>2b>2d. The antitumor effect on the ascites form of Ehrlich carcinoma in mice was also observed.¹¹⁾

REFERENCES AND NOTES

- 1) Lipids A and Related Compounds. III., Part II; K. Ikeda, S. Nakamoto, T. Takahashi, and K. Achiwa, Chem. Pharm. Bull., submitted.
- 2) T. Shiba, and S. Kusumoto, "Chemistry of Endotoxin," Vol. I, ed. by E. Th. Rietschel, Elsevier, New York, 1984, pp. 284-302.
- 3) a) K. Takayama, N. Qureshi, and P. Mascagni, J. Biol. Chem., 258, 12801 (1983);
b) M. Imoto, S. Kusumoto, T. Shiba, H. Naoki, T. Iwashita, E. T. Rietschel, H. W. Wollenweber, C. Galanos, and O. Luderitz, Tetrahedron Lett., 24, 4017 (1983);
c) U. Seydel, Lindner, H. W. Wollenweber, and E. T. Rietschel, Eur. J. Biochem., 145, 505 (1984).
- 4) a) M. Imoto, H. Yoshimura, N. Sakaguchi, S. Kusumoto, and T. Shiba, Tetrahedron Lett., 26, 1545 (1985), and the references therein; b) S. Kusumoto, M. Yamamoto, and T. Shiba, Tetrahedron Lett., 25, 3727 (1984); c) T. Takahashi, C. Shimizu, S. Nakamoto, K. Ikeda, and K. Achiwa, Chem. Pharm. Bull., 33, 1760 (1985).
- 5) a) M. Kiso, H. Ishida, and A. Hasegawa, Agric. Biol. Chem., 48, 251 (1984);
b) D. Charon, R. Chaley, A. Malinvaud, M. Mondange, and L. Szabo, Biochem., 24 2736 (1985).
- 6) Satisfactory analytical and spectral data were obtained for this compound.
- 7) S. Kusumoto, H. Yoshimura, M. Imoto, T. Shimamoto, and T. Shiba, Tetrahedron Lett., 26, 909 (1985).

-
- 8) C. Dioleg, M. Mondange, S. R. Sarfati, L. Szabo, and P. Szabo, *J. Chem. Soc., Perkin Trans. 1.*, 1984, 275.
- 9) 10b: mp 55-57°C, $[\alpha]_D^{21} -6.36^\circ$ (c=0.88, CHCl₃).
10c: mp 62-64°C, $[\alpha]_D^{21} -9.75^\circ$ (c=0.80, CHCl₃).
10d: mp 60-63°C, $[\alpha]_D^{20} -11.5^\circ$ (c=1.00, CHCl₃).
- 10) 2b: mp 172-174°C, $[\alpha]_D^{25} +5.65^\circ$ (c=0.46, CHCl₃).
2c: mp 155-157°C, $[\alpha]_D^{24} +10.5^\circ$ (c=0.40, CHCl₃).
2d: mp 157-159°C, $[\alpha]_D^{24} +19.5^\circ$ (c=1.24, CHCl₃).
- 11) T. Shimizu, S. Akiyama, T. Masuzawa, Y. Yanagihara, S. Nakamoto, T. Takahashi, K. Ikeda, and K. Achiwa, *Chem. Pharm. Bull.*, submitted.

(Received July 20, 1985)