Carbohydrate Research, 144 (1985) C15–C17
Elsevier Science Publishers B.V., Amsterdam – Printed in The Netherlands

Preliminary communication

The structural assignment of *N*-acylkansosamine, 4,6-dideoxy-4-[(*R*)-2-methoxypropanamido]-3-*C*-methyl-2-*O*-methyl-L-mannopyranose, by chemical synthesis

JUJI YOSHIMURA*, KEN-ICHI SATO, AMJAD AQEEL, RHIDDI BIR SINGH, and HIRONOBU HASHIMOTO

Laboratory of Chemistry for Natural Products, Tokyo Institute of Technology, Nagatsuta, Midori-ku, Yokohama 227 (Japan)

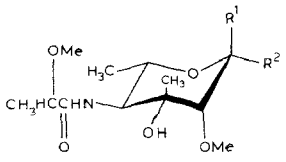
(Received September 19th, 1985; accepted for publication September 23rd, 1985)

The structure of a novel branched-chain amino sugar, *N*-acylkansosamine, isolated from the antigenic trehalose-containing lipooligosaccharides of *Mycobacterium kansasii*, was proposed[1] to be 4,6-dideoxy-4-(2-methoxypropanamido)-3-*C*-methyl-2-*O*-methyl-L-mannopyranose (**1**) by ^{1}H- and ^{13}C-n.m.r. spectroscopy and from mass-spectrometric data of derivatives. The *manno* configuration of **1** was ingeniously deduced from the hydrogen bonding between the hydroxyl group at C-3 and the carbonyl function of the 2-methoxypropanamido group. The absolute configuration based on the specific rotation, however, is uncertain, because such branched-chain sugars having closely related structures as nogalose (6-deoxy-3-*C*-methyl-2,4-di-*O*-methyl-L-mannopyranose) have the opposite sign[2]; furthermore, the contribution of the *N*-acyl moiety is difficult to assess. The configuration of the *N*-acyl group remains to be established. As spectral data of some derivatives are available[1], chemical synthesis should permit definitive structural assignment. This report endorses the proposed stereochemistry at C-3 and indicates from chemical synthesis that kansosamine has the L configuration and the configuration of the *N*-acyl group is *R*.

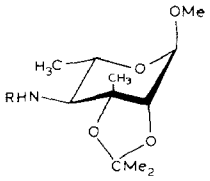
Recently, the authors synthesized[3] methyl α-L-sibirosaminide (methyl 4,6-dideoxy-3-*C*-methyl-4-methylamino-α-L-mannopyranoside), whose basic configuration is the same as that proposed for **1**. Methyl 4-amino-4,6-dideoxy-2,3-*O*-isopropylidene-3-*C*-methyl-α-L-mannopyranoside (**4**), which is one of the synthetic intermediates for methyl sibirosaminide, was obtained from L-rhamnose in 6 steps and was utilized as the starting material for the present synthesis.

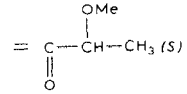
Coupling of **4** with (*S*)-2-methoxypropanoic acid with the aid of 3-(3-dimethylaminopropyl)-1-ethyl carbodiimide hydrochloride in dichloromethane for 1 h at room temperature gave quantitatively the corresponding (*S*) amide **5** as a syrup, $[\alpha]_D^{22}$ −76.8° (*c* 1.0, $CHCl_3$). Treatment of **5** with 70% acetic acid for 18 h at 80° gave quantitatively the *O*-deisopropylidenated product **6** as a syrup, $[\alpha]_D^{22}$ −119.5° (*c* 0.8, $CHCl_3$). Selective methylation of **6** with equimolar amounts of sodium hydride and methyl iodide in *N,N*-di-

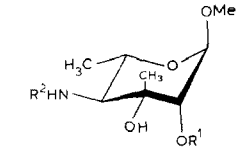
0008-6215/85/$03.30

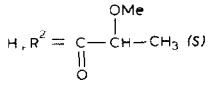
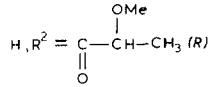
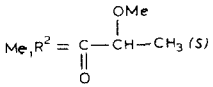
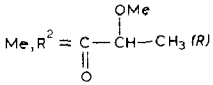


1 R^1, R^2 = H, OH
2 R^1 = OAc, R^2 = H
3 R^1 = H, R^2 = OAc



4 R = H
5 R = 



6 R^1 = H, R^2 =  (S)
7 R^1 = H, R^2 =  (R)
8 R^1 = Me, R^2 =  (S)
9 R^1 = Me, R^2 =  (R)

methylformamide at −5° gave, in 60% yield, the corresponding 2-methyl ether **8**, m.p. 134–136° (hexane), $[\alpha]_D^{22}$ −77.3° (*c* 0.8, $CHCl_3$); ^{1}H-n.m.r. data see Table I. As described for the (*S*) isomer, compound **4** was coupled with (*R*)-2-methoxypropanoic acid, which was prepared by optical resolution of the (*RS*) acid as the 1-phenylethylamine salt[4], to give the corresponding (*R*) amide (**7**), $[\alpha]_D^{22}$ −117.7° (*c* 0.5, $CHCl_3$). The amide **7** was then converted similarly into the 2-methyl ether **9**, m.p. 121–122° (hexane), $[\alpha]_D^{22}$ −35.6° (*c* 0.5, $CHCl_3$); ^{1}H-n.m.r. data see Table I. Comparison of the n.m.r.-spectra of **8** and **9** with that of methyl *N*-acyl-α-kansosaminide[1] clearly indicated that the spectrum of **9** was superposable on that of the natural compound. The n.m.r. data* were coincident for the two samples, as shown in Table I. Thus, the relative configuration was established as either L-*manno* with the (*R*) acyl group or D-*manno* with the (*S*) acyl group.

Further confirmation of the structure was performed by conversion of **9** into the corresponding 1-acetate. The glycoside **9** was hydrolyzed with 0.5M sulfuric acid at 85° to give a quantitative yield of the free sugar **1** as a syrup, $[\alpha]_D^{26}$ +5.6° (*c* 1.1, H_2O, equilibrium); lit.[1] $[\alpha]_D$ +14.7° (*c* 0.02, H_2O). Furthermore, acetylation of **1** with acetic anhydride in pyridine gave a mixture of 1-acetates (**2** and **3**, in the ratio of 4 to 3), which was separated by preparative t.l.c. with 7:3 ether–acetone. The ^{1}H-n.m.r. as well as ^{13}C-n.m.r. data of the α anomer **2** coincided again completely with those reported (Tables I and II), thus reconfirming the structure of *N*-acylkansosamine. Thus, the structure of *N*-acylkansosamine was assigned as 4,6-dideoxy-4-[(*R*)-2-methoxypropanamido]-3-*C*-methyl-2-*O*-methyl-L-mannopyranose. There remains a little ambiguity concerning the absolute configuration because of the low value of the specific rotation. As a change of the anomeric ratio could reverse the sign of rotation, decisive determination of the absolute configuration awaits chiro-optical data for natural *N*-acylkansosamine derivatives.

*^{1}H-N.m.r. data for natural methyl *N*-acyl-α-kansosaminide were kindly provided by P. J. Brennan.

TABLE I

^{1}H-N.M.R. CHEMICAL SHIFTS (δ) AND COUPLING CONSTANTS (Hz) OF *N*-ACYLKANSOSAMINE DERIVATIVES

Compound	*H-1* ($J_{1,2}$)	*H-2*	*H-4* ($J_{4,5}$)	*H-5* ($J_{5,6}$)	*H-6*	*Me-3*	*NH* ($J_{4,NH}$)	*H-1'* ($J_{1',2'}$)	*H-2'*	*OMe*	*OAc*	*OH*
2	6.21d (1.5)	3.12d	4.08t (10.0)	3.80dq (5.8)	1.23d	1.31s	6.44bd (10.0)	3.80q (6.8)	1.41d	3.40s 3.54s	2.11s	3.00bs
3	5.80s (<1.0)	3.23d	4.03t (10.0)	3.54dq (6.2)	1.27d	1.22s	6.40bd (10.0)	3.80q (7.0)	1.40d	3.37s 3.65s	2.15s	3.15bs
Reported[a]	6.19d (1.5)	3.09d	4.05t (10.4)	3.78m (6.1)	1.22d	1.28s	6.41d (10.2)	3.78m (6.8)	1.39d	3.37s 3.52s	2.09s	2.93bs
8	4.78bs (1.2)	3.10d	4.00t (10.0)	3.63dq (6.0)	1.18d	1.27s	6.47d (10.0)	3.78q (6.4)	1.39d	3.40 3.45 3.52		
9	4.77bs (1.3)	3.07d	4.01t (10.2)	3.65dq (6.0)	1.23d	1.27s	6.42d (10.2)	3.79q (6.4)	1.40d	3.38 3.39 3.50		
Reported[b]	4.78d (1.1)	3.08d	4.00t (10.3)	3.65dq (6.2)	1.24d	1.27s (10.3)	6.42d	3.79q	1.41d	3.38 3.39 3.50		

[a] For *N*-acyl-1-*O*-acetyl-α-kansosamine[1]. [b] For methyl *N*-acyl-α-kansosaminide[1].

TABLE II

^{13}C-N.M.R. CHEMICAL SHIFTS (δ) OF 1-*O*-ACETYL-*N*-ACYL-α-KANSOSAMINE

	C-1	*C-2*	*C-3*	*C-4*	*C-5*	*C-2'*	*OMe*	*CMe*		*C=O*
Reported[a]	91.1	78.4	71.0	55.4	82.7	69.3	57.4	18.1	18.4	168.9
							59.2	18.8	21.2	174.0
Synthesized[b]	91.2	78.4	71.1	55.4	82.7	69.3	57.4	18.1	18.4	168.9
							59.2	18.8	21.2	174.0

[a] See ref. 1. [b] Compound **2**.

ACKNOWLEDGMENTS

We thank Professor P.J. Brennan and Dr. T. Fujiwara for discussions and for spectral data.

REFERENCES

1 S. W. Hunter, T. Fujiwara, R. C. Murphy, and P. J. Brennan, *J. Biol. Chem.*, 259 (1984) 9729–9734.
2 N. Hong, M. Funabashi, and J. Yoshimura, *Carbohydr. Res.*, 96 (1981) 21–28; B. K. Bhuyan and C. G. Smith, *Proc. Natl. Acad. Sci. U.S.A.*, 54 (1965) 566–572.
3 J. Yoshimura, K. Sato, and R. B. Singh, *Chem. Lett.*, (1985) 69–70.
4 P. Newman, *Optical Resolution Procedures for Chemical Compounds*, Vol 2, Part I, Manhattan College, New York, 1981, p. 82.