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Preliminary Communication

Selective Synthesis of Cerebrosides: (2*S*, 3*R*, 4*E*)-1-*O*-β-D-galactopyranosyl-*N*-(2′*R* and 2′*S*)-2′-hydroxy-tetracosanoyl-sphingenine*

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The cerebrosides were first isolated by Thudicum in 1874 and the structures were established by Carter *et al.* in 1950 (for review, see [2]). In 1961 Shapiro and Flowers [3] reported the first total synthesis of a cerebroside **1** (Fig. 1) which was identified with the natural sample, only through comparison of their i.r. data. In order to confirm the absolute configuration at C-2′ of natural cerebroside **1**, we describe here an unambiguous synthesis of two stereoisomeric cerebrosides **1** and **2**, and found that the ^{1}H-NMR spectra of the synthetic **1** (Fig. 2) was completely identical with that of the natural cerebroside reported recently by Dabrowski *et al.* [4].

In planning the synthetic route, the target structures **1** and **2** were disconnected at the dotted lines to give three key synthetic intermediates **3**, **4** and **5** or **6** (Fig. 1).

Abbreviations: Bu, butyl; Ph, phenyl; *t*-$BuPh_2SiCl$, *t*-butyldiphenylsilyl chloride; MTPA, α-methoxy-α-trifluoromethylphenylacetic acid; THF, tetrahydrofuran.

*Part 36 in the series "Synthetic Studies on Cell-surface Glycans", for part 35, see [1].

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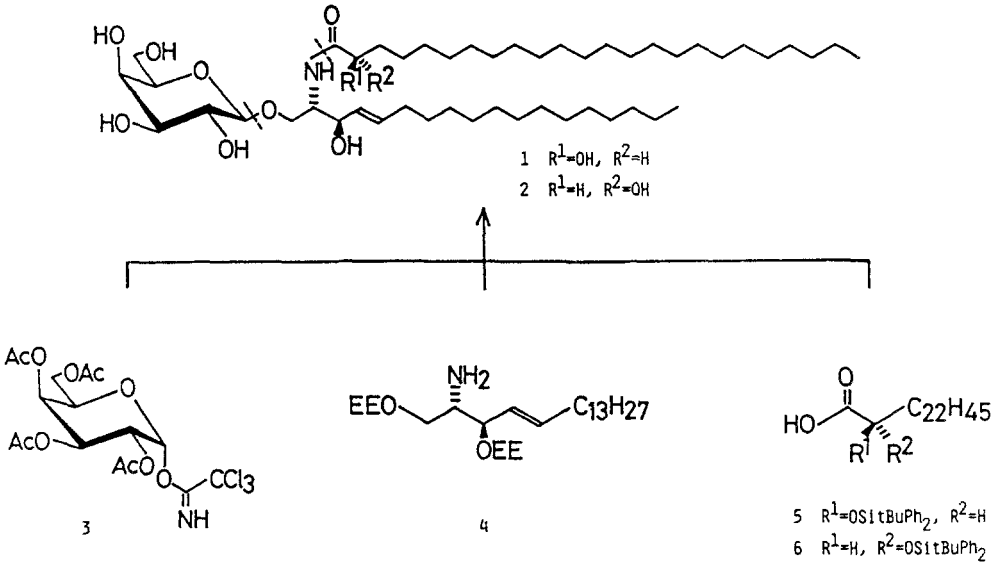

Figure 1. Key intermediates. Abbreviations: EE, ethoxyethyl; $SitBuPh_2$, *t*-butyldiphenylsilyl.

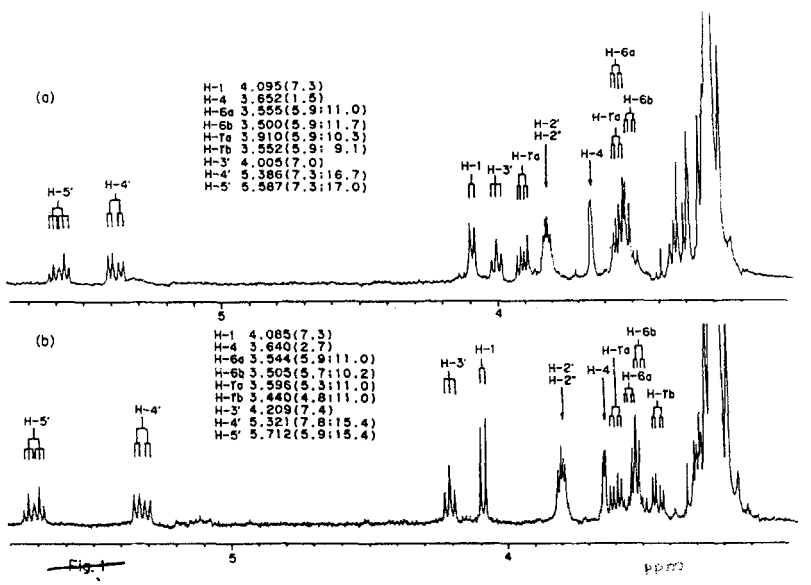

Figure 2. 400 MHz ^{1}H-NMR of (a) synthetic (2′*R*)-cerebroside **1**, and (b) synthetic (2′*S*)-cerebroside **2**. The spectra were recorded at 65°C in $^2H_6$-dimethylsulfoxide/^2H_2O, 49/1 by vol, for the sample after exchanging three times with ^2H_2O. Values of δ_H are expressed in ppm downward from internal standard tetramethylsilane. Values of $^3J_{HH}$ (Hz) are described in parentheses.

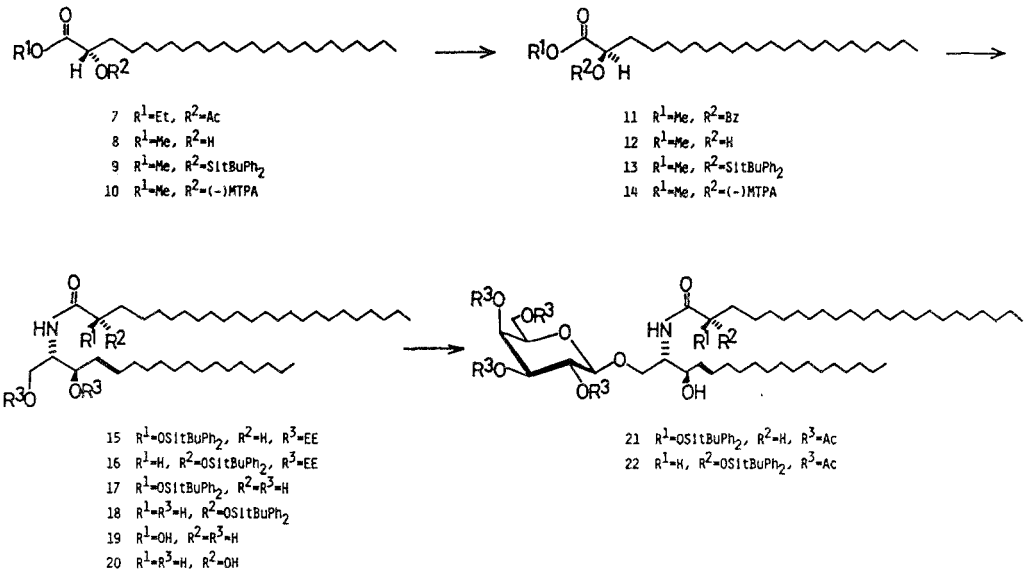

Figure 3. Synthetic scheme. Abbreviations: Bz, benzoyl; MTPA, α-methoxy-α-trifluoromethylphenylacetic acid.

The D-galactosyl donor **3** [5] and the (2*S*, 3*R*, 4*E*)-sphingenine derivative **4** [6] are already reported. The protected hydroxyacids **5** and **6** are readily prepared as follows (Fig. 3). Ethyl (2*S*)-2-acetoxy tetracosanoate **7**, $[\alpha]_D$ -14.2° (c 1.58, $CHCl_3$), obtainable [7] from (*S*)-(-)-malic acid was transformed into **6**, $[\alpha]_D$ +10.3° (c 0.91, $CHCl_3$) m.p. 49-50°C, R_F 0.50 in *n*-hexane/EtOAc, 4/1 by vol, in 77% overall yield in 3 steps: (i) NaOMe in MeOH/THF, 1/1 by vol, (ii) *t*-$BuPh_2SiCl$-imidazole in dimethylformamide, (iii) NaOH in MeOH/THF, 1/1 by vol. In a similar way, (*R*)-2-*t*-butyldiphenylsilyloxytetracosanic acid **5**, $[\alpha]_D$ -11.6° (c 0.29, $CHCl_3$), m.p. 48-49°C, could be prepared in 50% overall yield from **8** [the (-)-MTPA ester showed that **8** was obtained in 96.2%ee] in 4 steps (*via* **11**, **12** and **13**): (i) Ph_3P-PhCOOH-diethyl azodicarboxylate in THF [8], (ii) NaOMe in MeOH/THF, 1/1 by vol, (iii) *t*-$BuPh_2SiCl$/4-dimethylaminopyridine in pyridine, (iv) NaOH in MeOH/THF, 1/1 by vol.

400 MHz ^{1}H-NMR of the (-)MTPA esters [9], (*S*)-**10** and (*R*)-**14**, which were obtainable, respectively, from **6** and **5** in 3 steps; (i) CH_2N_2, (ii) Bu_4NF in THF, (iii) (-)-MTPA chloride in pyridine, showed that (*S*)-**10** and (*R*)-**14** were obtained in 97.0% ee and 95.4% ee, respectively, and that no significant racemization occurred during these transformations.

A solution of **4** and (*R*)-acid **5** in CH_2Cl_2 was treated with 1,3-dicyclohexylcarbodiimide in the presence of hydroxy benzotriazole to give a 92% yield of completely protected (2*S* 3*R*, 4*E*, 2′*R*)-ceramide **15**, R_F 0.52 and 0.45 in *n*-hexane/EtOAc, 4/1 by vol, which was solvolysed in MeOH/CH_2Cl_2, 1/1 by vol, in the presence of Amberlist® 15 to afford a 67% yield of the desired product **17**, $[\alpha]_D$ +6.4° (c 1.13, $CHCl_3$), m.p. 44-45°C, R_F 0.30 in *n*-hexane/EtOAc, 7/3 by vol. Treatment of **17** with Bu_4NF in THF afforded (2*S*, 3*R*, 4*E*, 2′*R*)-ceramide

19, $[\alpha]_D$ +8.7° (c 1.13, $CHCl_3$/MeOH, 9/1 by vol), m.p 98-99°C, R_F 0.45 in $CHCl_3$/MeOH, 9/1 by vol.

In a similar manner, **4** was condensed with (*S*)-acid **6** to give a 90% yield of completely protected (2*S*, 3*R*, 4*E*, 2'*S*)-ceramide **16**, R_F 0.62 and 0.55 in *n*-hexane/EtOAc, 4/1 by vol, which was further transformed into the oily glycosyl acceptor **18** (62%), $[\alpha]_D$ -6.4° (c 1.91, $CHCl_3$), R_F 0.46 in *n*-hexane/EtOAc, 7/3 by vol. Desilylation of **18** with fluoride anion gave an 80% yield of (2*S*, 3*R*, 4*E*, 2'*S*)-ceramide **20**, $[\alpha]_D$ -11.1° (c 1.41, $CHCl_3$/MeOH, 9/1 by vol), m.p. 100-101°C, R_F 0.51 in $CHCl_3$/MeOH, 9/1 by vol.

Crucial glycosylation of (2'*R*)-ceramide **17** with galactopyranosyl trichloroacetimidate **3** in $CHCl_3$ in the presence of BF_3-ether and molecular sieves 4Å according to the method of Schmidt and Michel [5] afforded a 33% yield of the protected (2'*R*)-cerebroside **21**, $[\alpha]_D$ +1.6° (c 1.12, $CHCl_3$), R_F 0.45 in $CHCl_3$/MeOH, 49/1 by vol. Complete deprotection of **21**, by (i) Bu_4NF in THF, (ii) NaOMe-MeOH, afforded a 62% yield of (2'*R*)-cerebroside **1**, $[\alpha]_D$ +8.5° (c 0.40, $CHCl_3$/MeOH, 1/1 by vol), R_F 0.37 in $CHCl_3$/MeOH, 17/3 by vol. Similarly, (2'*S*)-ceramide **18** was transformed into the protected cerebroside **22** (31%), $[\alpha]_D$ -2.8° (c 1.09, $CHCl_3$), R_F 0.61 in $CHCl_3$/MeOH, 49/1 by vol, which was further deprotected to afford (2'*S*)-cerebroside **2** (78%), $[\alpha]_D$ -25.2° (c 0.48, $CHCl_3$/MeOH, 1/1 by vol), R_F 0.48 in $CHCl_3$/MeOH, 17/3 by vol.

In conclusion, (2'*R*) and (2'*S*)-cerebrosides **1** and **2** were synthesized in a stereo-controlled way and their ^{1}H-NMR data (Fig. 2) provided the conclusive evidence for the stereochemical assignment of the natural cerebroside.

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