Chemoenzymatic Synthesis of D-*erythro*-C_{18}- and L-*threo*-C_{18}-Sphingosines

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Summary: Biocatalytic conversion of chlorobenzene to the corresponding homochiral cyclohexadiene *cis*-diol allows, through careful symmetry-based planning, the stereodivergent synthesis of two sphingosine stereoisomers, the natural isomer **1** and the L-*threo* isomer **2**, from selectively prepared diastereomers of azido alcohol **5**.

In 1876, Thudichum, a London surgeon–chemist, described the chemical composition of the brain and alluded to the presence of cerebroside (or cerebral galactoside) and its exact chemical composition, including a unique aliphatic alkaloid called "sphingosine."[1] Sphingosines constitute a group of related long-chain aliphatic 2-amino 1,3-diols, of which 2-amino-D-*erythro*-4(*E*)-octadecene-1,3-diol (**1**) occurs most frequently in animal glycosphingolipids,[2] the glycosides of *N*-acylsphingosines, or ceramides. The structural variation inherent in fatty acids, sphingosines, and carbohydrates results in a great number of chemically distinct glycosphingolipids,[2] which are of intense interest because of their diverse biological roles.[3] These include inhibition of protein kinase C[3d] and the transfer of information between developing cells in vertebrates.[3b] Recently, galactosyl ceramide has been shown to be a receptor for HIV binding in cells lacking the CD4 receptor.[4]

Many syntheses of optically pure sphingosines have relied on the use of L-serine as a chiral building block. The work of Newman,[5a] Garner,[5b] Rapoport,[5c] and Polt[3b] exemplifies the exhaustive work using serine aldehyde derivatives in their approaches to the title compound **2**. A highly stereoselective synthesis of L-*threo*-sphingosine, the unnatural isomer **2**, has been reported by Polt and co-workers.[3b] Noteworthy syntheses of the natural isomer, **1**, are those of Whitesides *via* the stereospecific hydration of chlorofumaric acid with fumarase[6] and

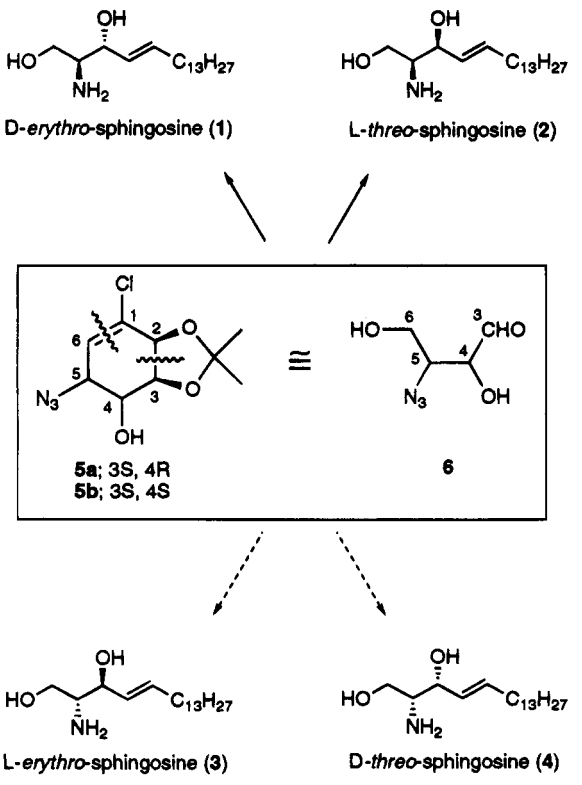
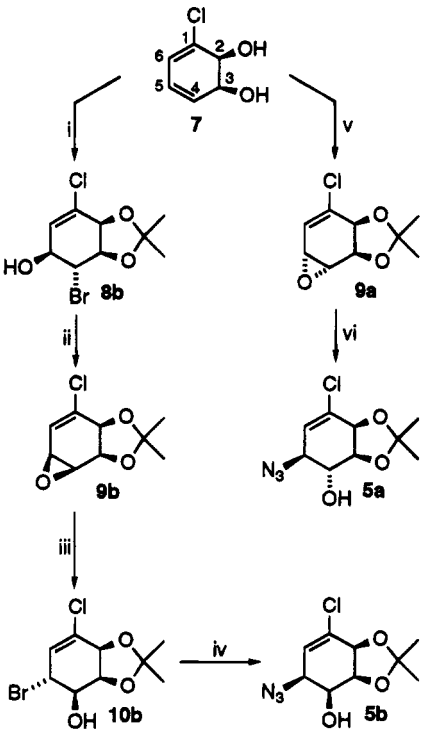


Figure 1. General design of sphingosines.

Scheme 1[a]



[a] Reagents and conditions: (i) (1) 2,2-dimethoxypropane, catalytic *p*-TsOH, CH_2Cl_2, (2) NBS, DME, H_2O, 0 °C; (ii) NaOH (1 equiv), Bu_4NHSO_4, CH_2Cl_2 reflux; (iii) LiBr (1.1 equiv), ethyl acetoacetate (2.5 equiv), THF, 35 °C; (iv) NaN_3, DMSO; (v) (1) 2,2-dimethoxypropane, catalytic *p*-TsOH, CH_2Cl_2, (2) *m*-CPBA, CH_2Cl_2; (vi) NaN_3, $NH_4^+Cl^-$, 1,2-dimethoxyethane/EtOH/H_2O, 70 °C.

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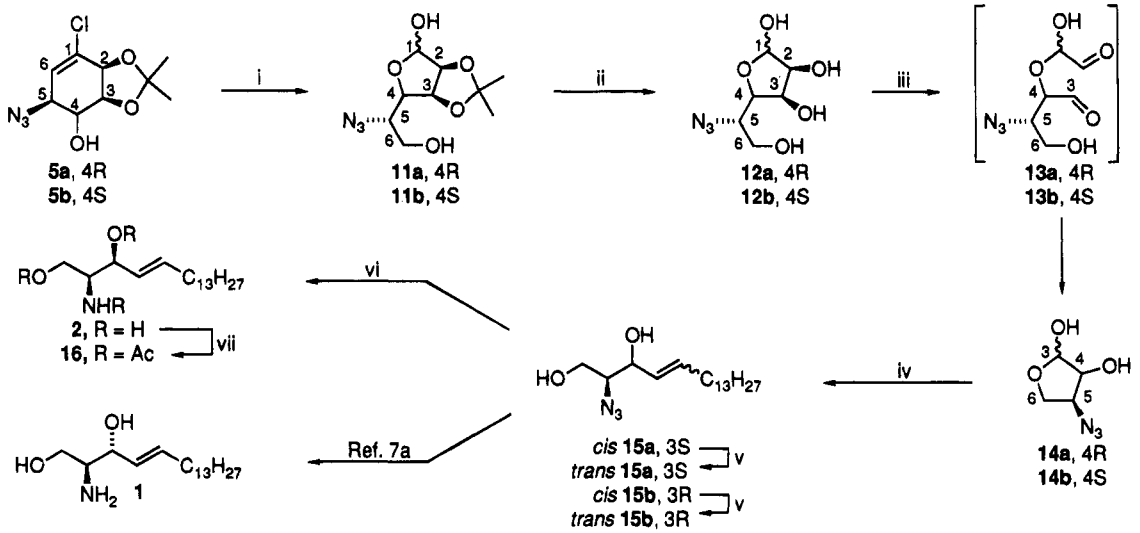
Scheme 2[a]



[a] Reagents and conditions: (i) (1) O_3, CH_3OH, −78 °C, (2) $NaBH_4$, CH_3OH, −55 °C to rt; (ii) Amberlyst 15 (wet) ion-exchange resin-strongly acidic, Aldrich Chemical Co.; (iii) $NaIO_4$, H_2O; (iv) *n*-tetradecylphosphonium bromide (3.2 equiv), n-BuLi (2.8 equiv), THF, rt; (v) hν, PhSSPh, dioxane, hexane; (vi) H_2S, pyridine/H_2O; (vii) acetic anhydride, pyridine.

Schmidt's[7] highly efficient synthesis from D-galactose. In addition, Takano[8] synthesized L-*erythro*- and D-*threo*-sphingosine, **3** and **4**, respectively, *via* a Katsuki–Sharpless asymmetric epoxidation reaction. Other syntheses have been reviewed.[9]

We envisioned the synthesis of all four isomers of the C_{18}-sphingosines by manipulation of the C4–C5 olefin in diene diols of type **7**, Scheme 1, available by enzymatic oxidation of chlorobenzene with toluene dioxygenase from the whole cells of *Pseudomonas putida* 39D.[10] The stereoselective establishment of the C4 and C5 centers of **5** and the subsequent cleavage of the C1–C6 olefin followed by the scission of the C2–C3 diol would provide the sphingosine synthon **6**, Figure 1. In this paper we report on the synthesis of sphingosines **1** and **2** by a method of potential generality: it should be noted that the chirality set during the enzymatic dioxygenation immolates, prior to its removal, the configuration of all future stereocenters through stereospecific functionalization of the C4–C5 olefin in **7**.

Chlorobenzenedihydrodiol (**7**), Scheme 1, was protected as an acetonide[11a-c] and converted to epoxide **9a**,[11d,12c] whose stereospecific opening with sodium azide afforded azido alcohol **5a**.[12] Azido alcohol **5b**, required for the synthesis of the natural isomer **1**, was prepared from the *syn*-epoxyacetonide **9b** as shown in Scheme 1. Reaction of **9b** with LiBr gave bromohydrin **10b** (94%), whose subsequent nucleophilic substitution with azide furnished azido alcohol **5b** (75%). Separate treatment of azido alcohols **5a** and **5b** with excess ozone at −78 °C, followed by excess $NaBH_4$ (carefully monitored by TLC), gave the azido-D-gulose (**11a**) and azido-D-allose (**11b**) in 70% yield, Scheme 2. Deprotection of **11** furnished **12**, whose cleavage with $NaIO_4$ afforded the azido-L-threose (**14a**) and azido-L-erythrose (**14b**) *via* **13** and its concomitant loss of the glyoxalate (65% overall yield from **11**).

Direct Wittig olefination of **14** under optimized conditions[13] gave azidosphingosine **15**. Lactol **14a** gave *cis* **15a** and *trans* **15a** in 24.4% and 6.1% yield, respectively, while **14b** gave *cis* **15b** and *trans* **15b** in 14.0% and 3.8% yield, respectively. *cis*-Azidosphingosines **15a** and **15b** were photoisomerized to *trans*-azidosphingosines **15a** and **15b** by means of a Hanovia 400 W lamp, Pyrex filter, and diphenyl disulfide in a 4:1 mixture of hexanes and dioxane.[14] Thus, from **14a** and **14b** a 20.7% yield of *trans*-azidosphingosine **15a** and 12.1% yield of *trans*-azidosphingosine **15b** were realized, respectively.

Reduction of *trans*-azidosphingosine (**15a**) gave **2**, which was acylated to **16** and shown to be indistinguishable (vide ^{1}H-NMR and $[\alpha]_D$) from an authentic sample. *trans*-Azidosphingosine (**15b**) displayed ^{1}H-NMR and $[\alpha]^{24}_D$ −34.17 (*c* 1.58, $CHCl_3$) [lit.[7a] $[\alpha]^{20}_D$ −32.9 (*c* 4.0,

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(13) Many Wittig olefination conditions were examined for the elaboration of **15**. The data suggest that the number of equivalents of phosphonium salt should be greater than that of the base; otherwise, the yield is greatly reduced. Schlosser–Wittig and modified Schlosser–Wittig conditions gave no product or <3% combined yield of *cis* and *trans*-sphingosines. The Conia–Dauben protocol (Conia, J.-M.; Limasset, J.-C. *Bull. Soc. Chem. Fr.* **1967**, *6*, 1936. Dauben, W. G.; Walker, D. M. *J. Org. Chem.* **1981**, *46*, 1103) and the Takai procedure (Takai, K.; Kakiuchi, T.; Kataoka, Y.; Utimoto, K. *J. Org. Chem.* **1994**, *59*, 2668) will be investigated for further improvements of this reaction.
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$CHCl_3$)] in agreement with the literature values and has been previously reduced to D-*erythro*-sphingosine (**1**).[7a]

The preparation of **1** and **2** bodes well for the potential of synthon **6** as a chiral pool reagent for a fully general method of synthesis for sphingosines. Optimization of these syntheses and the preparation of **3** and **4** from the appropriate isomers of **5** will be reported in the near future.

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Supplementary Material Available: Experimental procedures and ^{1}H- and ^{13}C-NMR spectra for compounds **5b**, **8b**, **9b**, **10b**, **11a**, **11b**, **14a**, **14b**, **15a**, and **15b** (31 pages). This material is contained in libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be ordered from ACS; see any current masthead page for ordering information.