The Enantioselective Synthesis of C_{18}-Sphingosines

by

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Chemistry

(Abstract)

Biocatalytic conversion of chlorobenzene to the corresponding homochiral cyclohexadiene cis-diol (113) allows, through careful symmetry-based planning, the stereodivergent synthesis of all sphingosine stereoisomers. This was achieved via the selective preparation of the appropriate diastereomer of azidoalcohol (118), were C-4 and C-5 correspond to C-3 and C-2 of the sphingosine skeleton, respectively (Scheme 1).
Scheme 1

D-erythro-sphingosine 1a  L-threo-sphingosine 1b  L-erythro-sphingosine 1c  D-threo-sphingosine 1d
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...to those who have given me unwavering support, through thick and thin
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IX. VITA
I. INTRODUCTION

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." Sphingosines constitute a group of related long-chain aliphatic 2-amino-1,3-diols, of which 2-amino-D-erythro-4E-octadecene-1,3-diol (I) occurs most frequently in animal glycosphingolipids, 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, which are of intense interest because of their diverse biological roles.

The intention of the synthetic project devised in this thesis is the enantioselective synthesis of all four stereoisomers of sphingosine from one chiral synthon. This goal was achieved using the versatile homochiral chlorocyclohexadiene-cis-diol synthon. The general utility of this synthon is apparent when one looks at the number of total syntheses of natural products that use this compound.

Chlorocyclohexadiene-cis-diol is one of many enantiomerically pure diols of type 113, accessible via the biocatalytic oxidation of aromatic compounds. Recently, it has been suggested that biotransformations could be used to remove toxic aromatic waste from the environment. It has been known for years that the soil bacterium Pseudomonas putida oxidatively degrades aromatic compounds to catechols and ultimately to muconates. In 1970 Gibson isolated a mutant strain of bacteria (Pseudomonas putida 39D) that arrested the oxidation of aromatics to catechols at the stage of the substituted cyclohexadiene-cis-diol. These bacterial metabolites are >95% enantiomerically pure. Industrially, aromatic waste such as benzene and chlorobenzene (over 3 million tons of each produced annually) could be converted into chiral synths of type 113.

It is this marriage of chemistry and microbiology that has allowed the field of biotransformations to grow so rapidly recently. Biotransformations or biocatalysis are chemical transformations mediated by either purified enzymes or by whole cell organisms (e.g. bacteria, fungi, plant or mammalian tissue culture). Included under this broad title are stereoselective hydrolysis, esterifications, oxidations, and reductions catalyzed by enzymes.
Organic synthesis is rich with examples of total syntheses of challenging and complex natural products, but upon examination of the aliphatic alkaloids called "sphingosines" one might be less excited. The molecule appears very simple, and it is. So the question arises, "Why are people still trying to make sphingosine?"

Since the first total synthesis in 1954, there have been at least another thirty syntheses. Over the years, overall yield has dramatically increased along with efficiency, to a point now of great desparity from the original synthesis. This has been accomplished mostly through great insight and to a lesser degree through the use of new reagents. The continued interest has been fueled by two main factors. One is the fact that only $D$-erythro-sphingosine exists in any significant amount in nature. Second, occasionally one of the other three isomers gives more encouraging results in biological assays. So, low natural abundance, lack of the other three isomers and diverse biological activities create a demand and keep the synthetic mind open to its synthesis. It is in this context that we became involved in the total synthesis of sphingosines, supported by interest from Genencor International, a biocatalysis-oriented company. What ensues is our story of the synthesis of sphingosines.
1. Sphingosines

1.1 Sphingolipid and Lysosphingolipid Structure

This thesis is concerned with the synthesis of sphingosines and necessitates a definition of their structures. Essential for the discussion of synthetic targets and bioactivity is a familiarity with the structures of sphingolipids and lysosphingolipids, which are themselves part of a larger category of biomolecules, namely the lipids.

Broadly defined, a lipid is any molecule of molecular weight between 100 and 5000 which has a substantial portion of its constitution as either aliphatic or aromatic in nature. Included are the hydrocarbons, steroids, soaps, detergents, and more complex molecules i.e. triacylglycerols, phospholipids, gangliosides, and lipopolysaccharides. The physical behavior of such chemically divergent molecules will be equally diverse. Indeed, one of the most interesting characteristics of lipids is their varied behavior in aqueous systems, ranging from almost total insolubility (e.g. paraffin oil, sterol esters) to nearly complete solubility (e.g. soaps, detergents, bile salts, and gangliosides). This particular aspect of lipids is important biologically because all cells exist in an aqueous milieu.5

The simplest sphingolipid is ceramide (Figure 1). All other sphingolipids have head groups attached at the C-1 carbon. Ceramides comprise sphingoid bases with an amide-linked fatty acyl chain, examples of which are stearoyl, oleoyl, palmitoyl, or linoleoyl residues. The stearoyl residue is the most commonly occurring fatty acyl chain. With the exception of sphingomyelin, which has a phosphoryl choline head group at C-1 linked through a phosphodiester bond, sphingolipids have a glycosidic bond at C-1. These sugar head groups can vary in complexity from a single glucose or galactose, as in cerebrosides, to more complex structures, such as lactosylceramide with two sugars, trinexosides with three, and higher order carbohydrate moieties. Certain subclasses are characterized by additional components, such as sulfatides, which contain sulfate, and gangliosides, which carry sialic acid residues. Lysosphingolipids are based on sphingosine in a manner analogous to the way sphingolipids are based on ceramide (Figure 1). For each parental sphingolipid there is a corresponding lysosphingolipid that has an identical head group at C-1, but that lacks the amide-linked fatty acyl group at C-2. All lysosphingolipids share two important structural features with sphingosine i.e. a charged amine at C-2 and a hydrophobic hydrocarbon tail. Note that sphingosine itself is a lysosphingolipid and not a sphingolipid.
<table>
<thead>
<tr>
<th>HEAD GROUP</th>
<th>SPHINGOLIPID</th>
<th>LYSOSPHINGOLIPID</th>
</tr>
</thead>
<tbody>
<tr>
<td>R = H</td>
<td>Ceramide</td>
<td>Sphingosine</td>
</tr>
<tr>
<td>R = Galactose</td>
<td>Galactocerebroside</td>
<td>Psychosine (Galactosylsphingosine)</td>
</tr>
<tr>
<td>R = Sulfogalactose</td>
<td>Sulfatide</td>
<td>Lysosulfatide (Sulfogalactosylsphingosine)</td>
</tr>
<tr>
<td>R = Ganglioside, e.g.</td>
<td>GM₂</td>
<td>Lyso GM₂</td>
</tr>
<tr>
<td>R = Phosphorylcholine</td>
<td>Sphingomyelin</td>
<td>Lysosphingomyelin</td>
</tr>
</tbody>
</table>

**Figure 1**
Although sphingosine is the predominant long-chain base in many sphingolipids and lysosphingolipids, other sphingoids may be present (Figure 2).\textsuperscript{6}

<table>
<thead>
<tr>
<th>STRUCTURE</th>
<th>n</th>
<th>NAME</th>
<th>OTHER NAMES</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>12</td>
<td>sphingosine</td>
<td>sphing-4-ene</td>
</tr>
<tr>
<td></td>
<td>12</td>
<td>sphinganine</td>
<td>dihydroxysphinganine</td>
</tr>
<tr>
<td></td>
<td>12</td>
<td>phytosphingosine</td>
<td>4-D-hydroxysphinganine</td>
</tr>
<tr>
<td></td>
<td>14</td>
<td>eicosasphingenine</td>
<td>\textit{D-erythro}-2-amino-4-\textit{trans}-eicosene-1,3-diol</td>
</tr>
</tbody>
</table>

Figure 2

1. Sphingosines
   1.2 Isolation and Structure Elucidation

Nerve tissue contains a high concentration of extremely complex lipid material. The three primary lipid constituents are cerebrosides,\textsuperscript{7} sphingomyelins,\textsuperscript{7} and gangliosides.\textsuperscript{8} The only practical source of the sphingolipids is the brain or spinal cord, although they occur in blood, liver, kidney, spleen, and other organs in small amounts. Brain is reported to contain 2.5 to 3.0 per cent of cerebroside\textsuperscript{9} and 1.0 to 3.0 per cent of sphingomyelin by weight.\textsuperscript{10} A variety of methods serve to isolate sphingolipids from nerve tissue. Practically all involve (1) dehydration of the fresh tissue, (2) extraction of the glycerophosphatides, and (3) extraction of the cerebrosides and sphingomyelins.\textsuperscript{11}

Thus Carter \textit{et al.}\textsuperscript{11} described the process using 100 pounds of freshly ground beef brain or spinal cord. The yield of crude sphingolipid from 45.4 kilos (100 pounds) of spinal cord ranged from 2.2 g to 2.5 g, compared to 1.3 g to 1.6 g when brain was used. The sphingolipids isolated from the spinal cord were of higher purity. For a brief procedural summary see reference 12.

The study of the chemical composition of the brain dates to the end of the eighteenth century. Foucroy made this task possible when he introduced organic solvents
(e.g. ethanol and ether) and mineral acids (e.g. H₂SO₄ and HNO₃). In 1793 he was able to isolate, among other things, a substance with properties like cholesterol from brain matter. It was not until 1812 that Vauquelin, Fourcroy's pupil, showed that phosphorus was inherent in brain matter. In 1834 Courbe isolated pure cholesterol via exhaustive extraction of brain matter. He also isolated a mixture of lipids, which was later referred to as protagon. In 1865 Liebreich hypothesized that the brain consisted of a single chemically distinct compound which he named protagon (Gr. protos, first; agonistes, a combatant). He described it as a white powdery brain substance which had been freed from fat and cholesterol by extraction with ethanol and ether. Nineteen years later Thudichum, now known as the chemist of the brain, published his work concerning protagon. His work showed that protagon was a mixture of phosphatides and cerebrosides and stated further that there were at least 14 adducts, which he isolated and analyzed. His contemporaries denounced his views, as Mathews points out, "Our knowledge of the chemical constitution of the brain is owing largely to Thudichum, a man of extraordinary care, accuracy, insight and industry, whose abilities were much underrated during his life. For, owing partly to an unusually combative nature, he alienated many of his colleagues and his work was neglected. There is now, however, no question that he was far in advance of all others in this difficult field and his 1901 book, entitled Die chemische Konstitution des Gehirns des Menschen und der Tiere, nach eigenen Forschungen bearbeitet, is a monument to his ability and insight." It was through extensive experimentation on the hydrolysis of phrenosine (a phosphorus-free entity of the brain), by Thudichum, that a parent base of "alkaloidal nature" was isolated. Of which he said, "in commemoration of the many enigmas which it presented to the inquirer" he gave the name sphingosine (Gr. sphingein, to bind tightly).

Thudichum suggested the first formulation for sphingosine, C₁₇H₃₅NO₂. In 1906 Thierfelder proved the presence of a double bond by adding Br₂. In 1912 Levene showed that sphingosine could be reduced to dihydrosphingosine, that it forms a dimethyl ether, and that all of its nitrogen is present as a primary amine. In the same year both Levene and Thierfelder independently made the triacetate and concluded that sphingosine was an unsaturated dihydroxy amine. One year later Levene's oxidative cleavage of the double bond yielded the aminotetrose 3 and the fatty acid 5 (upon further oxidation). This established the position of the double bond on the aliphatic chain. Unfortunately the oxidized fatty acid portion was misidentified as tridecanoic acid
4, when in actuality it was tetradecanoic acid 5 (Scheme 1). It was not until 1929 that Klenk\textsuperscript{26} reinvestigated the chromic acid oxidation and established that the fatty acid was indeed tetradecanoic acid 5. He also provided the correct empirical formula, C\textsubscript{18}H\textsubscript{37}NO\textsubscript{2}, for sphingosine.

\[
\begin{align*}
\text{X} & \quad \text{X} \\
\text{2} & \quad \text{12} \\
\text{X} & \quad \text{X} \\
\end{align*}
\text{Where X represents two alcohols and one primary amine of unknown position.}

\text{Scheme 1}

Several structural questions still remained. What was the relative order of the hetero atoms on C-1 through C-3? What was their relative stereochemistry, and finally, what was the geometry of the olefin? The assignment of the relative position of the hetero atoms was not a trivial one, because the first two assignments were incorrect (Scheme 2). In 1913 Levene\textsuperscript{27} made the first misassignment represented by structure 6. The second misassignment was made by Klenk\textsuperscript{28} when he suggested structure 7, in 1931.

\[
\begin{align*}
\text{H}_2\text{N} & \quad \text{OH} \\
\text{OH} & \quad \text{OH} \\
\end{align*}
\text{Scheme 2}

It was not until 1941 that Seydel\textsuperscript{29} proposed, in his dissertation, that the amino functionality resided on C-2. He came to this conclusion after periodate failed to cleave the assumed vicinal diol of the N-acetyldihydrosphingosine derivative of 7. One year later Carter\textsuperscript{30} studied the same problem with N-benzyldihydrosphingosine and found, as Seydel did, that periodate did not attack this molecule. Their independent conclusions were the same, namely sphingosine was a 2-amino-1,3-dihydroxy-4-octadecene.

In addition to the periodate reaction a few more reactions were performed that buttressed their structure proof (Scheme 3). Periodate cleavage of dihydrosphingosine 9 showed that all three hetero atoms were indeed on the first three carbons. In addition Carter found that the triacetate of sphingosine consumed 2.0 equivalents of hydrogen, implying that an allylic acetate was present. Hydrogenolysis of the allylic acetate formed olefin 11, which was further reduced to the fully saturated compound 12. Structure 12 was not identified until 1951 when Carter\textsuperscript{30c} confirmed the D-configuration of C-2 using
a chemical correlation study. He accomplished this feat by further derivatizing diacetate 12 to the known D-α-benzamidostearic acid. Two years later Carter\textsuperscript{31} published several papers concerning the synthesis of D-erythro-dihydrosphingosine and showed that the synthetic and natural dihydrosphingosine were identical in every respect.

\[
\begin{align*}
\text{HO} & \quad \text{NHBz} & \quad 8 & \quad \overset{\text{NaIO}_4}{\longrightarrow} & \quad \text{NO REACTION} \\
\text{HO} & \quad \text{NH}_2 & \quad 9 & \quad \overset{\text{NaIO}_4}{\longrightarrow} & \quad \text{H}_2\text{C}=\text{CH}_2 + \text{NH}_3 + \text{H}_2\text{O} + \text{H}_2 \\
\text{AcO} & \quad \text{NHAc} & \quad 10 & \quad \overset{\text{Pt, H}_2}{\longrightarrow} & \quad \text{AcO} \quad \text{NHAc} \quad 11 & \quad \longrightarrow & \quad \text{AcO} \quad \text{NHAc} \quad 12
\end{align*}
\]

Scheme 3

In 1947 Ohno provided a chemical correlation study which confirmed the trans geometry of the olefin in sphingosine.\textsuperscript{32} Degrading sphingosine to hexadecenal, he was then able to oxidize it to the known trans-hexadecenoic acid. This result was supported by Mislow\textsuperscript{33} who observed a strong infrared absorption at 10.3 μ, indicative of a trans double bond. A very informative and more detailed account of this structural proof can be found in Chemistry of Sphingolipids by David Shapiro.\textsuperscript{21}

1. Sphingosines

1.3 Bioactivity

At least 300 different sphingolipids are synthesized in various mammalian cell types. This diversity has intrigued investigators for many years. Thudichum, noted for his discovery of sphingosines,\textsuperscript{1} published his first report on the chemical constitution of the brain\textsuperscript{34} over 120 years ago (1874). In his last publication (1901), entitled "Die chemische Konstitution des Gehirns des Menschen und der Tiere," Thudichum summarizes his life-work on cerebrosides, sphingomyelins, and sphingosine.\textsuperscript{35,36}
In the latter half of the 1980's it was discovered that lysosphingolipids, the breakdown products of cellular sphingolipids, are biologically active. This bioactivity generated a new interest in the role of these molecules in cell physiology and pathology. Sphingolipid breakdown products, sphingosine and lysosphingolipids, inhibit protein kinase C, a pivotal enzyme in cell regulation and signal transduction. These compounds also affect significant cellular responses and exhibit anti tumor promoter activities in various mammalian cells. These molecules may play a role as endogenous modulators of cell function and possibly as second messengers.37

Until recently scientists were only able to allude to the important cellular functions of the vast array of complex cellular lipids. A recurring question was why these molecules had survived eons of evolution when a simple phospholipid would suffice in bilayer formation to delimit cells and to divide the cytoplasm into its organelles and compartments.37 The answer, while incomplete, has more to do with the breakdown products and metabolites of membrane lipids than with the lipids themselves. Many of these breakdown products and metabolites function predominantly in signal transduction as agonists or as second messengers. They include diacylglycerol,38 platelet activating factor,39 phosphatidic acid,40 arachidonic acid41 (4), prostaglandins,41 leukotrienes,41,42 (4,5) eicosanoids,41 thromboxanes,41 lipoxins,42 inositol phosphates,43 and inositol glycans.44 Although some of these, such as diacylglycerol and phosphatidic acid, are present constitutively in cells under resting conditions, most of the others are generated when cells are activated.44

A recent addition to this growing family of biologically and physiologically active lipids emerged from the investigation of sphingolipid breakdown products. When added to cells, sphingolipid-derived molecules, sphingosine and lysosphingolipids, elicit various pharmacological responses. Some of these responses are inhibition of platelet and neutrophil activation,45 inhibition of growth factor action,46 modulation of receptor function,47 and inhibition of phorbol ester-induced responses.48

A number of biological and pathological functions are attributed to different sphingolipids, they are summarized in Table 1 of the appendix.49
1. Sphingosine

1.4 Biosynthesis

Many nutritional experiments in animals have indicated that vitamin B₆ plays an undefined role in lipid metabolism, and particularly in the formation of polyunsaturated fatty acids. In an attempt to further assess these claims, Haskell and Snell conducted a careful comparison of the lipids of a yeast, *Hanseniaspora valbyensis*, grown with excess vitamins or deficient in vitamin B₆, pantothenic acid, or biotin. They found that the palmitoleic acid content of lipids from vitamin B₆-deficient cells was greatly reduced relative to that found in normal control cells, and that this effect was specifically related to the deficiency in vitamin B₆. These analyses also showed that the sphingolipid base content was substantially reduced as a result of the vitamin deficiency. The latter finding caught the attention of researchers interested in the metabolism of the sphingolipid bases.

The literature revealed an excellent rationale for the participation of pyridoxal phosphate (pyridoxal-P) in formation of the sphingolipids. Labeling experiments in animals had shown that sphingolipid bases arose from palmitate and serine, with loss of the carboxyl group of serine, a reaction reminiscent of the pyridoxal-P-dependent enzymatic decarboxylation of amino acids. One postulated course for this reaction was via a 3-keto intermediate, formed by replacing the carboxyl group of serine with the palmityl group. Such a reaction would be closely analogous to the formation of δ-aminolevulinic acid from succinyl CoA and glycine, a reaction known to require pyridoxal-P. In addition, Brady et al. had already achieved cell-free (in vitro) synthesis of dihydrosphingosine in brain microsomes from palmitaldehyde or palmityl CoA and serine, and had shown that this reaction was largely inhibited by dialysis of their preparations against cysteine (a good complexing agent for pyridoxal-P).

When it was found that a strain of *Hansenula ciferri*, a yeast, excretes relatively large quantities of dihydrosphingosine and phytosphingosine in the form of their acylated derivatives, a practical attempt at the biosynthesis problem could be started. These products arose from serine and palmitic acid. Using the knowledge gained from previous studies, Snell et al. proposed the biosynthesis of dihydrosphingosine followed the reaction sequence shown by equation 1 and 2 (Scheme 4).
The key features of this proposal are the previously unknown 3-keto intermediate 14, and the implicit assumption that two distinct enzymatic reactions are involved. From equations (1) and (2) it is evident that the proposed ketonic intermediate 14 should accumulate in the absence of TPNH (NADPH). Indeed, in the absence of TPNH the 3-keto intermediate 14 was formed. Separate experiments showed it to be inhibited by cysteine, i.e. reaction (1) appears to be pyridoxyl-P-dependent [PLP-E (pyridoxal-P enzyme)]. Thus Snell proposed a likely mechanism of formation (Scheme 5).

A question still remains: when was the unsaturation introduced at C-4 and C-5? To test their hypothesis, that the unsaturation was incorporated after formation of ketone
14, synthetic 3-keto-dihydrosphingosine was treated TPNH (i.e. same reaction conditions, but without palmitoyl CoA and serine). The only observed product was dihydrosphingosine, via reduction of the ketone. No sphingosine was found. Since these enzyme preparations form sphingosine in addition to dihydrosphingosine (sphinganine) when palmitoyl CoA and serine are the substrates, one can tentatively conclude that desaturation during the synthesis of sphingosine must occur at the fatty acyl CoA level,\textsuperscript{52} and not at the ketonic level (Scheme 6).\textsuperscript{56}

Scheme 6: Probable Course of Biosynthesis for Sphingosine and Dihydrosphingosine.
2. Syntheses

2.1 L-Serine Approaches

Many syntheses of optically pure sphingosines have relied on the use of L-serine as a chiral building block. The first application of L-serine to sphingosine synthesis was reported by Newman\textsuperscript{57} in 1973. Since then, exhaustive work using serine derivatives has culminated in highly efficient and diastereoselective synthesis of D-\textit{erythro}- and L-\textit{threo}-sphingosine. The starting materials for the syntheses to follow are shown in Table 1 and are commercially available.

<table>
<thead>
<tr>
<th>L-serine when R = H</th>
</tr>
</thead>
<tbody>
<tr>
<td>N-(t-butoxycarbonyl)-L-serine (N-Boc-L-serine) when R = CO\textsubscript{2}t-Bu</td>
</tr>
<tr>
<td>N-(benzyloxycarbonyl)-L-serine (N-Cbz-L-serine) when R = CO\textsubscript{2}CH\textsubscript{2}C\textsubscript{6}H\textsubscript{5}</td>
</tr>
<tr>
<td>L-Serine methyl ester hydrochloride</td>
</tr>
</tbody>
</table>

Newman\textsuperscript{57} published the first synthesis of sphingosine originating from L-serine. N-phthaloylation of L-serine followed by treatment with acetic anhydride gave a primary acetate. Treatment with thionyl chloride provided the acid chloride which was subsequently reduced to aldehyde 20 (Scheme 7). Treatment of aldehyde 20 with \textit{trans}-pentadecenylidiisobutylalane (\textit{trans}-vinyl-alane) gave the protected sphingosine 21 in predominantly the \textit{erythro} form.

![Scheme 7](image)

Thornton \textit{et al.}\textsuperscript{58} made \textit{erythro}- and \textit{threo}-N-oleoyl-D-sphingosines (ceramides) and galactosylerceramides (v cerebrosides) in 1981. Starting from L-serine the oxazoline aldehyde 22 was made in three steps and in 65% overall yield. The key step involved the addition of \textit{trans}-vinylalane to 22, yielding a 1:1 \textit{erythro/threo} ratio of protected sphingosines 23 and 24 in a combined yield of 50% (Scheme 8). In a one pot sequence from 23 or 24 the corresponding ceramides were made in 13% overall yield.
Scheme 8

In 1986 Rapoport et al.\textsuperscript{59} were the first to employ the lithium pentadecyne approach. The synthesis began with N-Cbz-L-serine which was converted to the protected L-serine isoxazolidide 25 in two steps and with 76% yield (Scheme 9). Lithium pentadecyne was added to isoxazolidide 25 (R\textsuperscript{1}= H) to give an ynnone in 90% yield. Reduction of the ynnone yielded propargylic alcohols 26 as a mixture of diastereomers. In an attempt to improve the diastereoselectivity of the reduction, a large variety of reducing reagents were investigated. Unfortunately no reducing agent gave high diastereoselectivities and high yields of propargylic alcohols 26. The best results provided a 5:1 ratio of diastereomers erythro 26a and threo 26b in a combined yield of 63% (53% erythro).

In an attempt to improve the diastereoselectivity and yield, an alternate approach was investigated. Addition of lithium ethyne to 25 (R\textsuperscript{1}= Si(Me)\textsubscript{2}-Bu) afforded the corresponding ynnone (racemized on chromatography with Florisil) in 60 - 80% yield. Reduction of the ynnone using NaBH\textsubscript{4} and CeCl\textsubscript{3} provided an 85% yield of 13 (Scheme 9) in a 6:4 ratio of diastereomers (accurate assignment as 27a vs 27b was not made). Acetonide formation revealed the major diastereomer to be 28. Terminal alkyne 28 was deprotonated and alkylated to give 29 in 35% yield from 28. Low yields and or low diastereoselectivity hampered this total synthesis. D-erythro-sphingosine was made in 22% overall yield in five steps. Note that 26a and 26b were only separable when silica gel treated with sodium borate was used (Boric acid impregnated silica gel has been used to differentiate diastereomeric 1,2 diols by TLC).\textsuperscript{60}
In 1988, Garner et al.\textsuperscript{61} used N-Boc-L-serine to make D-erythro-sphingosine triacetate in 39\% overall yield. This was accomplished in eight steps with 8:1 erythro/threo diastereoselectivity. The first three steps were used to form the acetonide of N-Boc-L-serinal 30. No further chromatographic purifications were necessary for the remainder of the synthesis. The key step, addition of lithium pentadecyne, yielding propargylic alcohols which were then reduced to sphingosine derivatives 31 and 32 (Scheme 10). This method afforded D-erythro-sphingosine in 39\% overall yield. Garner also investigated addition of Newman's\textsuperscript{62} trans-vinylalane to N-Boc-serinal acetonide 30 and found a 2:1 threo/erythro diastereoselectivity.

\textbf{Scheme 10}

In 1988 Herold\textsuperscript{63} used N-Boc-L-serine to make D-erythro- and L-threo-sphingosine triacetate, and their cis geometric isomers. In addition he made N-octadecanoyl-D-erythro-sphingosine, which is a ceramide or N-acylated sphingosine. The approach resembles that of Garner's in that both used lithium pentadecyne as the carbon nucleophile in the additions to N-Boc-serinal acetonide 30. By employing chelation control through judicious choice of Lewis acids, solvent, and temperature, Herold was able to add lithium pentadecyne with 95\% ds to give either erythro or threo propargylic alcohols 33 or 34 selectively (Scheme 11). The triacetate of D-
erythro-sphingosine or L-threo-sphingosine was obtained in eight steps with 18% and 21% yields, respectively. The alkynes were reduced to the trans olefins using sodium bis(2-methoxyethoxy)aluminum hydride (Red-Al) in Et₂O at 0 °C to room tempertaure in 65% yield.

![Scheme 11](image)

In 1988 a third independent synthesis of D-erythro- and L-threo-sphingosine was published, by Liotta et al.⁶⁴ These chemists also took advantage of the N-Boc-L-serinal acetonide 30 and lithium pentadecyne as Garner and Herold did. Using lithium pentadecyne in THF at -78 °C a 9:1 erythro/threo ratio of propargylic alcohols 33 and 34 was observed. Alcohols 33 and 34 gave a combined yield of 90% (81% erythro). Unique to this approach was the conversion of propargylic alcohol 33 to 34 using standard Mitsunobu conditions in 70% yield. Reduction of alkynes 33 or 34 with Na°/NH₃ proceeded in 90% yield when performed on a small scale (< 100 mg). When larger scale reductions were desired lithium aluminum hydride in refluxing 1,2 dimethoxyethane had to be employed. These large scale reductions proceeded in 70% yield. In six steps D-erythro-sphingosine was made in 28% yield and L-threo-sphingosine in eight steps and in 20% yield.

In 1990 Dondoni et al.⁶⁵ used N-Boc-L-serine to make the D-erythro-C20-sphingosine triacetate. This synthesis was accomplished in ten steps (six purifications) and with 12:1 erythro/threo diastereoselectivity, in an overall yield of 15%. In addition phytosphingosine was synthesized. These researches took advantage of a stable thiazole masked aldehyde, which could be deprotected at the appropriate time using mild conditions.⁶⁶ The subsequent aldehyde would then be subjected to Wittig olefination. Addition of 2-(trimethyl-silyl)thiazole to N-Boc-L-serinal acetonide 30 afforded the one carbon homologues, i.e. the masked aldehydes 35 and 36, with 85-90% anti-diastereoselectivity at the new asymmetric center (Scheme 12). Deprotection of thiazole 36 and Wittig olefination of the resulting aldehyde 37 gave the protected D-erythro-C20-sphingosine 38, which was deprotected and then converted to its triacetate for the purpose of identification.
L-Serine methylester hydrochloride was used by Polt et al.\textsuperscript{67} to prepare L-threo-sphingosine triacetate in 1992. The synthesis was accomplished in four steps, with > 95% diastereoselectivity and in an overall yield of 59%. Diastereoselective syntheses of sphingosine and C1-deoxygenated sphingosines, derived from L-alanine, of varying chain lengths were achieved. The key step involved the reaction of α-imino esters (O'Donnell's Schiff bases) with aluminum hydrides to produce acetal-like intermediates which underwent subsequent reaction with carbon nucleophiles. For the synthesis of L-threo-sphingosine (Scheme 13) the Schiff base methyl O-(tert-butyldimethylsilyl) -N-(diphenyl-methylene)-L-serinate (39), shown pre-coordinated as an aluminum acetal, was converted to the protected L-threo-sphingosine (40) in > 20:1 threolerythro selectivity in one pot and with 76% combined yield (72% threo).
This synthesis proceeded without epimerization, confirmed via use of Mosher's amide, and with a high degree of threo selectivity. Although some Boc-protected amino aldehyde derivatives show resistance to racemization, facile loss of optical activity in α-amino aldehyde derivatives is a general problem. Sphingosine derivative 40 was fully deprotected to L-threo-sphingosine using HCl in dioxane at 100 °C by Polt. The overall yield of L-threo-sphingosine was 59% from L-serine methylester hydrochloride.

The novelty and efficiency of sphingosine synthesis from serine has progressed by leaps and bounds since the first synthesis by Newman (Table 2). The syntheses have been arranged according to their overall percent yields.
<table>
<thead>
<tr>
<th>Author</th>
<th>Year</th>
<th>Starting Material</th>
<th>Product</th>
<th>Steps</th>
<th>Overall % Yield</th>
<th>Ref</th>
</tr>
</thead>
<tbody>
<tr>
<td>Polt</td>
<td>1992</td>
<td></td>
<td><em>D</em>-threo-sphingosine</td>
<td>5</td>
<td>59</td>
<td>67</td>
</tr>
<tr>
<td>Garner</td>
<td>1988</td>
<td></td>
<td><em>D</em>-erythro-sphingosine</td>
<td>6</td>
<td>39</td>
<td>61</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td><em>D</em>-threo-sphingosine</td>
<td>6</td>
<td>14</td>
<td></td>
</tr>
<tr>
<td>Liotta</td>
<td>1988</td>
<td></td>
<td><em>D</em>-erythro-sphingosine</td>
<td>6</td>
<td>28</td>
<td>64</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td><em>D</em>-threo-sphingosine</td>
<td>8</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td>Rapoport</td>
<td>1986</td>
<td></td>
<td><em>D</em>-erythro-sphingosine</td>
<td>5</td>
<td>22</td>
<td>59</td>
</tr>
<tr>
<td>Herold</td>
<td>1988</td>
<td></td>
<td><em>D</em>-erythro-sphingosine</td>
<td>8</td>
<td>18</td>
<td>63</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td><em>D</em>-threo-sphingosine</td>
<td>8</td>
<td>21</td>
<td></td>
</tr>
<tr>
<td>Thornton</td>
<td>1981</td>
<td></td>
<td>1-benzoyl-<em>D</em>-erythro-sphingosine</td>
<td>5</td>
<td>16</td>
<td>58</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>1-benzoyl-<em>D</em>-threo-sphingosine</td>
<td>5</td>
<td>16</td>
<td></td>
</tr>
<tr>
<td>Dondoni</td>
<td>1990</td>
<td></td>
<td><em>D</em>-erythro-sphingosine</td>
<td>10</td>
<td>15</td>
<td>65</td>
</tr>
<tr>
<td>Newman</td>
<td>1973</td>
<td></td>
<td><em>D</em>-erythro-sphingosine</td>
<td>6</td>
<td>*</td>
<td>57</td>
</tr>
</tbody>
</table>

All yields were calculated based on information obtained from the experimental sections of the cited papers and references therein, when available. * Many reactions were reported without a percent yield.
2.2 Sugar Approaches

A crucial problem with the L-serine approach is the introduction of the second asymmetric center. One way to avoid unwanted mixtures of diastereomers is to begin the synthetic sequence with the stereocenters already established. The chiral pool of sugars become attractive building blocks for such an endeavor.

The first sphingosine synthesis from sugars was accomplished by Reist et al.\textsuperscript{70} in 1970. Starting with 3-amino-3-deoxy-1,2:5,6-di-isopropylidene-\(\alpha\)-D-allolufuranose\textsuperscript{71} the synthesis of D-\textit{erythro}-sphingosine was accomplished in 8 steps with a 5\% overall yield. The key step was the Wittig olefination of aldehyde 41 which provided the coupled \textit{trans} product 42 in 60\% yield (Scheme 14). The strong band at 970 cm\(^{-1}\) in the infrared spectrum was used to assign the geometry of the olefin. Upon examination of the spectral data for olefin 42 it was found that the methyl carbamate had been converted to the ethyl carbamate. The authors reasoned that phenyl lithium had attacked the solvent (ether) to produce ethylene and an ethoxy anion, which then proceeded to transesterify the methyl carbamate. Reist previously published a synthesis of D-dihydrosphingosine,\textsuperscript{72} from the same starting material.

![Scheme 14](image)

\(\text{RHN}^+\text{O}^\text{O}^\text{O}^\text{O}^\text{O}^\text{O} \xrightarrow{\text{phosphonium salt}} \text{C}_{13}\text{H}_{27}\text{RHN}^+\text{O}^\text{O}^\text{O}^\text{O}^\text{O}^\text{O} \xrightarrow{\text{phenyl lithium}} \text{RHN}^+\text{O}^\text{O}^\text{O}^\text{O}^\text{O}^\text{O}\)

It was not until 1985 that sphingosines were again synthesized via a sugar route. Schlosser \textit{et al.}\textsuperscript{73} were able to convert D(+)\textit{mannose} and D(+)\textit{ribose}-1,4-lactone into D-\textit{erythro} and L-\textit{threo}\textit{-sphingosine} respectively. Schlosser states that the key intermediates were the two epimers of 4\textit{-bromo-3-methoxymethyl-2-methoxymethoxymethyl-2,3-dihydrofurans 44a and 44b (Scheme 15). Treatment of 44 with \textit{n-butyllithium} gave the alkynes 45 which were subsequently alkylated to give the sphingosine skeleton. In twenty four steps and in 1.3\% overall yield D-\textit{erythro}-sphingosine was made, while the preparation of L-\textit{threo}-sphingosine took twenty steps and proceeded in 1.6\% overall yield.
Reagents and conditions: (i) (1) Li, NH₃, (2) CICH₂OCH₃, EtN(iPr)₂, (3) Br₂, CCl₄, (4) DBU, THF; (ii) n-BuLi, THF.

Scheme 15

Three efficient syntheses of D-erythro-sphingosine were published in the first six months of 1986. Independently, Schimdt, Kiso, and Ogawa employed very similar approaches. Schmidt and Zimmermann published a total synthesis of D-erythro-sphingosine from D-galactose in seven steps with an overall yield of 7%. The crucial step was the Wittig olefination of 2,4-O-benzylidene-D-threose 46 (Scheme 16) to yield the trans-olefin 47 in 68% yield. Conversion of the free alcohol intrins-olefin 47 to an azide (via the triflate) with inverse stereochemistry, followed by deprotection provided azido D-erythro-sphingosine. Despite the low overall yield the synthesis is an efficient one. If the first step is eliminated (formation of 4,6-O-benzylidene-D-galactose from D-galactose, 26% yield) the overall yield increases to 28%. In 1988 a detailed paper with full experimental procedures was published. In this publication Schmidt reports that 2,4-O-benzylidene-D-threose 46 can be made from D-galactose and D-arabinose, and further shows that 2,4-O-isopropylidene-D-threose 48 can be made from D-xylose and D-glucose.

Scheme 16

Kiso et al. employed the same synthetic strategy as Schmidt, the only differences being the initial isopropylidene protecting group vs Schimdt's benzylidene protecting group and the Wittig condensation. Using Schlosser Wittig betaine-ylid conditions, 2,4-O-isopropylidene-D-threose 48, available from 3,5-O-isopropylidene-D-xylofuranose or 4,6-O-isopropylidene-D-galactopyranose was converted to 49 in 40% yield (35% cis-olefin isolated, Scheme 16). Thus from D-xylose, D-erythro-sphingosine was made in seven steps and in 6% overall yield. Unlike
many researchers Kiso did not photoisomerize the cis-olefin 49 (these photoisomerizations generally proceed in high yield) to the trans-olefin 49, lowering his overall yield. As with Schmidt’s synthesis the lowest yielding step is the first one, namely the formation of the 3,5-O-isopropylidene-xylofuranose from D-xylose. Kiso also converted his sphingosine product into several ceramides.

Ogawa et al. used D-glucose to arrive at aldehyde 50 in three steps. Use of standard Wittig condensation procedures provided a 42% of the trans olefin 51, and 47% of the cis olefin (Scheme 17). The cis geometric isomer was photoisomerized to the trans olefin 51, so that an 87% yield of 51 could be realized from aldehyde 50. Ogawa was interested in the synthesis of ceramides, thus these researchers never made azido D-erythro-sphingosine or D-erythro-sphingosine, but instead a related compound 52 was made (Scheme 18). Thus, from D-glucose, protected azido D-erythro-sphingosine 52 was made in eleven steps and in 14% overall yield. Noteworthy was the reduction of azide 52, accomplished in 90% yield using sodium borohydride in refluxing isopropanol.

![Scheme 17](image)

![Scheme 18](image)

In 1993 Yadav et al. synthesized D-erythro- and L-threo-sphingosine from D-xylose and D-arabinose respectively. The key reaction was a base-induced double elimination of chloride 53 followed by alkylation of the terminal alkyne to give high yields (53a → 54a in 78% yield) of the corresponding propargylic alcohols 54 (Scheme 19). While the β-alkoxy chloride elimination to give propargylic alcohols 54 is interesting this synthesis offered no improvement over past syntheses. For both series fourteen steps were needed and an estimated overall yield of < 10% and > 5% was assumed based on the available data, as many yields were not reported in the paper.
In 1994 additional syntheses of sphingosines were published. First Murakami et al.\textsuperscript{83} made D-erythro-sphingosine and phytosphingosine from D-glucosamine hydrochloride. After nine steps epoxytosylate 55 was made and then coupled with a dodecyl cuprate to give epoxide 56 (Scheme 20). The epoxide was then opened with NaI to give the corresponding iodohydrin 57, from which either D-erythro-sphingosine or phytosphingosine was made. This synthesis in 15 steps provides D-erythro-sphingosine in 19\% overall from the hydrochloride of D-glucosamine.\textsuperscript{84}
Late in 1994 Wu et al.\textsuperscript{85} published a formal synthesis of D-erythro-62 and L-threo-sphingosine 63. The strategy was to make the four stereoisomers of 1,2,3-trihydroxy-4E-octadecene 64-67 and then to protect them as their 1,3-O-benzylidene-4-octadecene-1,2,3 triols, subject them to Mitsunobu conditions and then liberate the desired sphingosine 60-63 (Scheme 21). To this end they investigated the syntheses of 64 - 67 from sugars (Table 3).

\begin{table}[ht]
\centering
\begin{tabular}{lll}
\hline
Triol & Starting material & Overall % yield, \# of steps \\
\hline
64 & D-mannose & 39\%, 6 \\
65 & L-tartaric acid & 13\%, 8 \\
65 & D-xylose & 20\%\textsuperscript{†}, 7 \\
66 & D-xylose & 9\%, 5 \\
66 & D-glucose & 27\%, 5 \\
66 & D-galactose & 10\%, 5 \\
67 & D-glucose & 35\%, 4 \\
\hline
\end{tabular}
\caption{Table 3}
\end{table}

\textsuperscript{†} No % yield given for one step, so it was arbitrarily assigned 50\%.
While the syntheses from sugars do not suffer from mixtures of diastereomers, they generally lack the higher overall yields associated with the L-serine approaches. Many times the initial protection of the sugar is the lowest yielding step. If this fact is taken into account, then many of these syntheses are highly efficient and noteworthy. The sugar-based syntheses have been organized in tabular form with overall percent yield as the criterion for their relative order (Table 4). Note that several (1,2,3)-trihydroxy-4E-octadecene have been included in the table, some representing formal syntheses.

<table>
<thead>
<tr>
<th>Author</th>
<th>Year</th>
<th>Starting Material</th>
<th>Product</th>
<th>Steps</th>
<th>%</th>
<th>Ref</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wu</td>
<td>1994</td>
<td>D-glucose</td>
<td>(1,2R,3R)-trihydroxy-4E-octadecene</td>
<td>5</td>
<td>27</td>
<td>85</td>
</tr>
<tr>
<td></td>
<td></td>
<td>D-glucose</td>
<td>(1,2R,3S)-trihydroxy-4E-octadecene</td>
<td>4</td>
<td>35</td>
<td>85</td>
</tr>
<tr>
<td></td>
<td></td>
<td>D-mannose</td>
<td>(1,2S,3R)-trihydroxy-4E-octadecene</td>
<td>6</td>
<td>39</td>
<td>85</td>
</tr>
<tr>
<td></td>
<td></td>
<td>D-xylose</td>
<td>(1,2S,3S)-trihydroxy-4E-octadecene</td>
<td>7</td>
<td>20</td>
<td>85</td>
</tr>
<tr>
<td>Murakami</td>
<td>1994</td>
<td>D-glucosamine</td>
<td>D-erythro-sphingosine</td>
<td>13</td>
<td>19</td>
<td>83</td>
</tr>
<tr>
<td>Ogawa</td>
<td>1986</td>
<td>D-glucose</td>
<td>1,3-diethoxymethoxy-D-erythro-sphingosine</td>
<td>11</td>
<td>14</td>
<td>80</td>
</tr>
<tr>
<td>Kiso</td>
<td>1986</td>
<td>D-galactose</td>
<td>D-erythro-sphingosine</td>
<td>7</td>
<td>10</td>
<td>76</td>
</tr>
<tr>
<td>Schmidt</td>
<td>1986</td>
<td>D-galactose, D-xylose, D-arabinose, or D-glucose</td>
<td>D-erythro-sphingosine</td>
<td>7</td>
<td>7</td>
<td>74</td>
</tr>
<tr>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>75</td>
</tr>
<tr>
<td>Reist</td>
<td>1970</td>
<td>D-glucosamine</td>
<td>D-erythro-sphingosine</td>
<td>7</td>
<td>6</td>
<td>70</td>
</tr>
<tr>
<td>Yadav</td>
<td>1993</td>
<td>D-xylose</td>
<td>D-erythro-sphingosine</td>
<td>14</td>
<td>&lt;10</td>
<td>81</td>
</tr>
<tr>
<td></td>
<td></td>
<td>D-arabinose</td>
<td>D-threo-sphingosine</td>
<td>14</td>
<td>&lt;10</td>
<td>81</td>
</tr>
<tr>
<td>Schlosser</td>
<td>1985</td>
<td>D-mannose</td>
<td>D-erythro-sphingosine</td>
<td>24</td>
<td>1</td>
<td>73</td>
</tr>
<tr>
<td></td>
<td></td>
<td>D-ribono-1,4-lactone</td>
<td>D-threo-sphingosine</td>
<td>20</td>
<td>1</td>
<td>73</td>
</tr>
</tbody>
</table>
2.3 First Total Synthesis and Miscellaneous Approaches

Seventy-two years after the initial isolation of sphingosine by Thudichum, the first total synthesis of the alkaloid sphingosine appeared and confirmed the assigned structure. Shapiro et al.\textsuperscript{86} began their synthesis with a Knoevenagel-Doebner condensation of myristryl aldehyde (tetradecyl aldehyde) with malonic acid. The crucial reaction took place four steps later when ester 68 was treated with benzenediazonium chloride in the presence of sodium acetate and ammonium chloride to give hydrazone 69 (Scheme 22). This was the first time the Japp-Klingemann reaction\textsuperscript{87} was used on such a substrate. At the time the coupling of a diazonium salt with acyl substituted aceto-acetic esters was known, but had not been carried out with \(\alpha,\alpha\)-diacyl ester in which one of the acyl groups is \(\alpha,\beta\)-unsaturated. This synthesis can be performed on tens of grams and proceeded in 3\% overall yield to give \(DL\)-erythro-sphingosine in nine steps.

\[
\begin{array}{c}
\text{C}_{13}\text{H}_{27}\text{O}_2\text{C} \rightarrow \text{PhN} & \text{C}_{13}\text{H}_{27}\text{O}_2\text{C} \rightarrow \text{C}_{13}\text{H}_{27}\text{O} \\
\text{68} & \text{69} \\
\end{array}
\]

Scheme 22

In 1983, Vasella et al.\textsuperscript{88} published a synthesis of \(D\)-erythro-sphingosine. Of the several key reactions, the first was the generation of enynol 70 (Scheme 23), \textit{via} a Pd coupling reaction. Sharpless epoxidation of 70 proceeded in high yield and enantiomeric purity. This epoxy alcohol was then converted to carbamate 71a or imidate 71b (Scheme 24). Intramolecular cyclization of carbamates 71a gave the desired regiochemical opening of the epoxide to afford oxazolidinone 72a. This oxazolidinone was then converted to \(D\)-erythro-sphingosine. The trichloroimidate 71b preferred to cyclize with the opposite regiochemical opening and was used to synthesize the 3-amino-2-hydroxy-\(L\)-erythro-isomer of sphingosine.

\[
\begin{array}{cccc}
\text{C}_{13}\text{H}_{27} & \rightarrow & \text{C}_{13}\text{H}_{27} & \rightarrow \\
\text{B} & \text{OH} & \text{OH} & 4 \text{ steps} \\
\text{80\% yield} & 39\% \text{overall yield} & & \\
\end{array}
\]

Scheme 23


In 1986 Vasella published a second generation synthesis of D-erythro-sphingosine. The synthesis of enynol 70 was improved so that the palladium coupling was no longer needed (Scheme 23). In addition the percent yield of both the Sharpless epoxidation and the reduction of the alkyne to the trans alkene were increased. D-erythro-Sphingosine was synthesized in 50% overall yield and in six steps.

Scheme 24

In 1986 a (±)-erythro-sphingosine synthesis was reported by Cardillo et al. A Wittig-Horner olefination of tetradecylaldehyde 73 yielded the diene ester 74. Lithium aluminum hydride reduction of diene ester 74, formation of the trichloroimidate and treatment of the diene trichloroimidate with N-iodo-succinimide gave the corresponding 4,5-dihydro-1,3-oxazine 75 (Scheme 25). These researchers noted the same problem that

Scheme 25

Vasella et al. observed with imidate 71b (Scheme 24). Acidic hydrolysis of 75 afforded the ammonium salt, which upon treatment with base formed aziridine 76. This aziridine opened exclusively at the allylic sight with Amberlyst A 26 (in the acetate form) to give racemic acetamido-sphingosine in 32% yield and in seven steps.
In 1987 George Whitesides et al.\textsuperscript{90} was investigating the breadth of synthetically useful substrates accepted by fumarase. Thus chlorofumaric acid 77 was stereospecifically hydrated to give L-\textit{threo}-chloromalic acid 78, in \( \geq 99.5 \% \) ee (Scheme 26). L-\textit{threo}-chloromalic acid 78 was converted in nine steps to give oxazolidinone 79. Julia coupling\textsuperscript{91} yielded the desired sulfone acetate 80, which gives olefin 81. From acid 78, D-\textit{erythro}-sphingosine was made in thirteen steps and in 3\% overall yield.

![Scheme 26]

Another synthesis of sphingosine was published in 1988 by Nicolaou et al.\textsuperscript{92} Starting with the chiral oxazolidinone 82, the corresponding boron enolate was condensed with an \( \alpha,\beta \)-unsaturated aldehyde to afford derivative 83 in 72\% yield. Nucleophilic displacement of bromine with an azide group, silyl protection of the allylic alcohol, followed by reduction with LiBH\(_4\), gives the very important sphingosine derivative 84 in 52\% yield from 82. From oxazolidinone 82 D-\textit{erythro}-sphingosine was made in 42\% yield and in six steps (Scheme 27).

![Scheme 27]
Scheme 28

Takano et al.\(^\text{93}\) took advantage of a Katsuki-Sharpless epoxidation of (R,R) and meso-1,2-divinylethylene glycols to give epoxides 85 and 86 (Scheme 28). After four steps epoxide 85 was converted to diol 87. Oxidative cyclization of 87 using 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) in the presence of 4 Å molecular sieves gave 1,3-bezylidene 88 (Scheme 29). Mesylation of the free alcohol and treatment with laurly magnesium bromide, CuI gives the 1,3-benzylidene-azido-sphingosine. Thus epoxide 85 is converted to L-threo-sphingosine in 11% overall yield and in 10 steps. In a similar manner epoxide 86 was converted to D-threo-sphingosine in 16% overall yield in 10 steps.

Scheme 29

Somfai and Olsson\(^\text{94}\) in 1993 made a derivative similar to Nicolaou's azide 84 (Scheme 27) although in much lower yield and in twice the number of steps. Starting with diethyl-D-tartrate diol, diol 89 was made in two steps and in quantitative yield (Scheme 30). Treatment with diborane yields the unsymmetrical triol 90 and protection of the 1,2 vicinal diol affords 91. The key step is the oxidation of alcohol 91 to the aldehyde which is not purified further, but subjected to Wittig conditions using freshly prepared phenyl lithium to ensure trans-selectivity.\(^\text{149b}\)

In 1994 Solladie-Cavailo and Koessler\(^\text{95}\) published a four-step diastereo- and enantioselective synthesis of D-erythro-sphingosine. Utilizing (+)-(R,R,R)-hydroxypinanone 93 as a chiral template, iminoglycin ate 94 was synthesized (Scheme 31). Formation of the titanium enolate of 94, followed by addition of E-hexadecenal afforded imino ester 95 in 60% yield. Note: 20% of both the iminoglycinate 94 and E-hexadecenal were recovered. Hydrolysis of imino ester 95 afforded compound 96 in 85% yield. The chiral auxillary (+)-(R,R,R)-hydroxypinanone 93 was also recovered, in 80% yield. Reduction of the ester moiety in 96 afforded D-erythro-sphingosine in 35 - 40% overall yield, based on recovered starting materials.
Scheme 30

Scheme 31
All the miscellaneous syntheses have been arranged according to their overall percent yield in Table 5.

<table>
<thead>
<tr>
<th>Author</th>
<th>Year</th>
<th>Starting Material</th>
<th>Product</th>
<th>Steps</th>
<th>Overall Yield</th>
<th>Ref</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vasella</td>
<td>1983</td>
<td>pentadecyne</td>
<td>D-threo-sphingosine</td>
<td>6</td>
<td>50%</td>
<td>88</td>
</tr>
<tr>
<td>Nicolaou</td>
<td>1988</td>
<td></td>
<td>D-erythro-sphingosine</td>
<td>6</td>
<td>42%</td>
<td>92</td>
</tr>
<tr>
<td>Solladie</td>
<td>1994</td>
<td>(+)-(R,R,R)-hydroxypinanone</td>
<td>D-erythro-sphingosine</td>
<td>4</td>
<td>35%</td>
<td>95</td>
</tr>
<tr>
<td>Cardillo</td>
<td>1986</td>
<td>tetradecyl-aldehyde</td>
<td>(±)-sphingosine</td>
<td>7</td>
<td>32%</td>
<td>89</td>
</tr>
<tr>
<td>Somfai</td>
<td>1993</td>
<td>diethyl-D-tartrate</td>
<td>D-erythro-sphingosine</td>
<td>13</td>
<td>31%</td>
<td>94</td>
</tr>
<tr>
<td>Takano</td>
<td>1991</td>
<td>meso-1,2-divinylethylene-glycol</td>
<td>L-threo-sphingosine</td>
<td>10</td>
<td>11%</td>
<td>93</td>
</tr>
<tr>
<td>Whitesides</td>
<td>1987</td>
<td>chlorofumaric acid</td>
<td>D-threo-sphingosine</td>
<td>10</td>
<td>16%</td>
<td></td>
</tr>
<tr>
<td>Shapiro</td>
<td>1954</td>
<td>tetradecyl-aldehyde</td>
<td>(±)-sphingosine</td>
<td>9</td>
<td>3</td>
<td>86</td>
</tr>
</tbody>
</table>
3.0 Vinlyc oxirane openings with carbon and heteroatom nucleophiles.

Unlike isolated oxiranes which have two possible sites for nucleophilic attack vinyl oxiranes exhibit three (Scheme 32). Vinyl oxiranes, endowed with all of the reactivities of the former, also lend themselves to a whole new class of reactions, namely palladium catalyzed nucleophilic additions. A brief overview of the regio- and stereospecificity of these reactions ensues.

![Scheme 32](image)

**Scheme 32**

Nucleophilic $S_N2$ opening of epoxides affords trans products. When unsymmetrical epoxides are ring opened with nucleophiles, a mixture of regioisomeric products generally ensues. Unsymmetrical cyclic epoxides, on the other hand, give products resulting from only one of two possible trans-diaxial openings.\(^96\) The exclusive formation of one product is the result of the stereoelectronic demands inherent in the transition states. For example, the conformationally rigid cyclic epoxide 97 (Scheme 33), gives only one product 101.\(^96\) The transition state in the $S_N2$ reaction requires that the reaction at C-2 must give the twist-boat intermediate 98 which would lead to the diequatorial product 99. When attack is at C-3 the chair intermediate 100 is required and the diaxial product 101 ensues. This is a classic example of a kinetically controlled reaction product. Intermediate 100 is lower in energy than 98 and thus the higher energy diaxial product 101 is formed preferentially over the lower energy diequatorial product 99. In general, when the energy gap between the two possible transition states (e.g. 98 & 100) is large, exclusive formation of one product is found. As the energy gap between the two transitions states decreases, formation of the other regioisomer becomes possible. The same principles apply to vinyl oxiranes, as will be seen in the following paragraphs.
The addition of carbon nucleophiles to vinyl oxiranes has been studied extensively. Organocuprate and Grignard reagents add stereoselectively in a $S_N2'$ manner (vinyllogous attack) to yield allylic alcohols.\textsuperscript{97} Table 6 demonstrates the stereoselective \textit{anti} addition of organocuprates and the preponderance for retention of the initial olefinic geometry.\textsuperscript{97b} Organocuprate addition to cyclic vinyl oxiranes proceeds in an analogous \textit{anti} $S_N2'$ mode. \(\beta\)-Attack has been observed but usually is a secondary product to vinyllogous attack and can generally be suppressed by the use of cyanocuprate reagents (Scheme 34). These stereoelectronic effects cease to dominate when stericly demanding reactive centers are involved.\textsuperscript{97a,98} Extensive work has been published concerning organocuprate additions to 1,3-cyclopentadiene, 1,3-cyclohexadiene, and 1,3-cycloheptadiene monoepoxides.\textsuperscript{97a,98,99}

Vinyl and phenyl organostannanes couple with vinyl oxiranes in good yield to give allylic alcohols.\textsuperscript{100} Other organostannanes either fail to react (allyl, benzyl, and alkyl) or do so by other pathways. The most common mode of addition is \textit{anti} $S_N2'$, although \(\beta\)-addition sometimes occurs. The reactions are palladium catalyzed and are believed to proceed as indicated in Scheme 35.
Table 6. $S_{N}2'$ Cuprate Addition to Vinyl Oxiranes

\[
\begin{align*}
102a & \quad \xrightarrow{i \text{ or } ii} \quad 103a + 104a \\
102b & \quad \xrightarrow{i \text{ or } ii} \quad 103b + 104b
\end{align*}
\]

<table>
<thead>
<tr>
<th>Vinyl Oxirane</th>
<th>Ratio of 103:104</th>
<th>Yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Trans 102a</td>
<td>84:16</td>
<td>81</td>
</tr>
<tr>
<td>cis 102a</td>
<td>1:99</td>
<td>88</td>
</tr>
<tr>
<td>Trans 102b</td>
<td>14:86</td>
<td>95</td>
</tr>
<tr>
<td>cis 102b</td>
<td>97:3</td>
<td>95</td>
</tr>
</tbody>
</table>

i. LiMe$_2$Cu, THF/Et$_2$O (4:1), 0°C; ii. LiMeCuCN, Et$_2$O, 0°C

\[ R = \text{H, SiMe}_2\text{tBu}, \quad \text{1,3-cyclohexadiene-monoepoxide} \]

![Scheme 34](image)
Scheme 35

In contrast to the organometallic carbanions mentioned so far, the addition of stabilized carbanions proceeds under neutral conditions. Malonate-type additions to cyclic and acyclic vinyl oxiranes, using palladium catalysis affords syn S_n^2' products (Scheme 36). The syn addition is explained by initial coordination of Pd (0), followed by deprotonation of the malonate species by the alkoxyanion of the former epoxide. Attack by the malonate anion then occurs at the least sterically hindered face of the π-allylpalladium complex. When the Pd catalyst is absent, products arising from anti β-attack are observed (Scheme 36).

Scheme 36

The heteroatomic opening of vinyl oxiranes has been examined extensively. This brief discussion will be limited to alcohols, acids, anhydrides, esters, thiols, amines, and azide nucleophiles. Treatment of 1,3-cyclohexadiene monoepoxide with alcohols or amines in the presence of alumina affords products of β-addition (Scheme 37). When
acids are also used. Under Pd(0) catalysis (e.g. tetrakis(triphenylphosphine)palladium) cyclic and acyclic vinyl oxiranes can afford a diverse number of mono- and diprotected cis-2-(cyclo)alkene-i,4-diols (Table 7).\textsuperscript{101} The chemistry is closely related to the malonate "type" addition found in Scheme 36, were the zwitterion 102 is the same. In fact the premise for the addition is the same. The oxygen in 102 can function as an alkoxide base while the electron deficient allylic system reacts with nucleophiles in a manner analogous to normal π-allylpalladium complexes.\textsuperscript{101a} Thus any nucleophile containing a proton of lower acidity than an alcohol should in theory add to 102, Table 6, in a syn S_N2' fashion. The nucleophilicity of the alkoxide anion in 102 has also been exploited, entries 3-5.\textsuperscript{101a} Tenaglia\textsuperscript{103} notes that direct amination, with primary amines, of complexes of type 102 do not occur, "because of the occurrence of stable unreactive intermediate palladium complexes." The use of secondary amines however does not have this short coming, entry 7 produced 110 in 92% yield.\textsuperscript{104} Note that the syn S_N2' additions in Table 7 do not exclude acyclic cases, the transformation of 102 to 110 is solely for illustrative purposes.
Table 7. Palladium Catalyzed Heteroatomic Addition to Vinyl Oxiranes

<table>
<thead>
<tr>
<th>Entry number</th>
<th>Nucleophile</th>
<th>R</th>
<th>R'</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>R'CO₂H</td>
<td>R'CO₂</td>
<td>H</td>
<td>fa)</td>
</tr>
<tr>
<td>2</td>
<td>ArOH</td>
<td>ArO</td>
<td>H</td>
<td>fa)</td>
</tr>
<tr>
<td>3</td>
<td>AcOAr</td>
<td>OAr</td>
<td>Ac</td>
<td>fa)</td>
</tr>
<tr>
<td>4</td>
<td>(R''CO)₂O</td>
<td>R''CO₂</td>
<td>R''CO</td>
<td>fa)</td>
</tr>
<tr>
<td>5</td>
<td>R₃''SiOCOR'''</td>
<td>R₃''SiO</td>
<td>R''CO</td>
<td>fb)</td>
</tr>
<tr>
<td>6</td>
<td>NaN₃</td>
<td>N₃</td>
<td>H*</td>
<td>g</td>
</tr>
<tr>
<td>7</td>
<td></td>
<td></td>
<td>H</td>
<td>h</td>
</tr>
<tr>
<td>8</td>
<td></td>
<td></td>
<td>H</td>
<td>h</td>
</tr>
<tr>
<td>9</td>
<td></td>
<td></td>
<td>H</td>
<td>e,h</td>
</tr>
</tbody>
</table>

* Note that H₂O, in the reaction mixture, served as the proton source.

The reaction of vinyl oxiranes with sulfur nucleophiles is unique in that α-, β-, and S₉2' addition can be controlled by selecting the appropriate conditions. For example vinyl epoxide 111 under Lewis acid conditions afforded the β-addition product in 52% yield. Note that upon treatment of the same epoxide with a catalytic amount of n-BuLi α addition dominates. Interestingly S₉2' addition of thiol phenol to vinyl oxirane 112 can be controlled to give exclusively cis or trans olefins (Scheme 38).
Scheme 38
III. DISCUSSION

1. Introduction - Strategy

The objective of this investigation was the synthesis of all four stereoisomers of sphingosine 1. We reasoned that this could be accomplished from one common starting material, namely (2R,3S)-2,3-dihydroxy-1-chlorocyclohexa-4,6-diene\textsuperscript{105} (in future text referred to as chlorodiol 113 (Scheme 39). Although several possibilities exist, our retrosynthetic analysis was biased by a working knowledge of the reactivity of chlorodiol 113 and similar diols.\textsuperscript{106} The synthesis presented here is the culmination of the chemical knowledge gained during the investigation of the original proposal.

\[\text{Sphingosine 1} \quad \rightarrow \quad \text{lactol 122}\]

\[\text{chlorodiol 113} \quad \leftarrow \quad \text{azidoalcohol 118}\]

Scheme 39

Crucial to our success would be the synthesis of all four C4 and C5 stereoisomers of azidoalcohol 118 (Scheme 39). Earlier work on chlorodiol 113 suggested the use of an acetonide protecting group.\textsuperscript{107} This would combine a high degree of steric hindrance on the β-face with an unsymmetrical diene unit in the cyclohexene ring (Scheme 40). Any subsequent chemical transformations performed on the diene would then be biased toward the α-face.\textsuperscript{107} Because of the stereoelectronic features of the diene, the C4–C5 double bond is more electron rich, and thus reacts with electrophiles preferentially over the Cl–C6 double bond. Thus, exploitation of acetonide 114, should allow for the high degree of regio- and stereocontrol needed to synthesize all four diastereomers of sphingosine in an enantiomerically pure form.

The stereocenters of sphingosine would be established at the cyclic stage, were the high degree of stereocontrol needed is more easily achieved. This could be accomplished by
synthesizing the four diastereomers of azidoalcohol 118 (Scheme 39). Azidoalcohols 118 could then be exhaustively cleaved to provide lactols 122 (Note: The carbon atoms of chlorodiol 113 have retained the same numbers in compounds 118 and 122 of Scheme 39). A Wittig reaction of lactol 122 with the ylide of tetrade cyltri phenyl phosphonium bromide could then provide an azidosphingosine product. Simple reduction of the azide moiety would then provide the natural product, sphingosine.

Carbons C1 through C4 of sphingosine 1 would originate from the chlorodiol 113 (Scheme 39). Treatment of the vinyl chloride moiety, in 118, with ozone, followed by reductive work-up should provide the primary alcohol of sphingosine. Carbons C2 and C3 of sphingosine correspond to C5 and C4 of azidoalcohol 118, respectively. Oxidative cleavage of C2–C3 of azidoalcohol 118 should provide an aldehyde at C3 which corresponds to C4 of sphingosine. Wittig olefination will provide the rest of the aliphatic skeleton of sphingosine.
2. Results

2.1 A Misassigned Structure

Based on the literature data, an assumption was made that treatment of epoxide 115 with NaN₃ would result in a homo allylic opening of the epoxide to provide azidoalcohol 116 (Scheme 40). Our original goal was to synthesize the four C4 and C5 stereoisomers of type 116, a regioisomer of azidoalcohol 118 (Scheme 39).

Facile protection of chlorodiol 113, followed by epoxidation (m-CPBA) afforded epoxy acetone 115 in a regio- and stereospecific manner. It was reported that epoxy acetone 115 upon treatment with NaN₃ and NH₄Cl in the polar solvent mixture of 1,2-dimethoxyethane, ethanol, and water, provided azidoalcohol 116 (Scheme 40). Using this reaction protocol, an azidoalcohol believed to have structure 116 was obtained.

\[ \text{Cl} \overset{\text{OH}}{\text{OH}} \quad 113 \quad \overset{\text{i}}{\rightarrow} \quad \text{Cl} \overset{\text{O}}{\text{O}} \quad 114 \quad \overset{\text{ii}}{\rightarrow} \quad \text{Cl} \overset{\text{O}}{\text{O}} \quad 115 \quad \overset{\text{iii}}{\rightarrow} \quad \text{Cl} \overset{\text{1}}{\text{1}} \overset{\text{3}}{\text{3}} \overset{\text{N₃}}{\text{O}} \quad 116 \]

Scheme 40

Reagents and Conditions: (i) 2,2-dimethoxypropane, cat. p-TsOH, CH₂Cl₂; (ii) m-CPBA, CH₂Cl₂, 0 °C; (iii) NaN₃, NH₄⁺Cl⁻, 1,2-dimethoxyethane, EtOH, H₂O, 70 °C.

The relevance of azidoalcohol 116 is apparent in light of its retrosynthetic analysis (Scheme 41). Although azidoalcohol 116 could be dismantled in several different ways, we decided to cleave the C2–C3 bond first. Removal of the acetone, in 116, would afford a vicinal diol 117 (Scheme 42), which could then be oxidatively cleaved, i.e. with NaIO₄. To this end, azidoalcohol 116 was deprotected to afford a vicinal diol, believed to have structure 117.

\[ \text{HO} \overset{\text{3}}{\text{4}} \overset{\text{5}}{\text{6}} \overset{\text{CH₃}}{\text{12}} \quad 1d \quad \overset{\text{→}}{\text{}} \quad \text{Cl} \overset{\text{1}}{\text{1}} \overset{\text{3}}{\text{3}} \overset{\text{N₃}}{\text{O}} \quad 116 \]

Scheme 41
Scheme 42
Reagents and Conditions: (i) Amberlyst 15 (wet) ion-exchange resin-strongly acidic, Aldrich Chemical Co.

Careful scrutiny of the spectral data, obtained for this novel compound 117, revealed inconsistencies. The proton spectrum of the presumed triol 117 was recorded in DMSO-d$_6$ (Figure 4). Addition of D$_2$O eliminated the exchangeable protons, largely simplifying the resonance patterns (Figure 5). The connectivity assignment began with the readily identifiable olefinic proton H$_A$ (δ 5.72 d, J = 2.6 Hz, 1H). The only other proton resonance showing the same coupling constant (J = 2.6 Hz) was at 3.93 ppm (J = 2.6, 7.8 Hz, 1H) and therefore must be H$_B$, residing on carbon C5. This was supported by selective irradiation experiments. Surprisingly, the resonance pattern of this proton (H$_B$) appeared to be unchanged upon addition of D$_2$O (see Figures 4 and 5), indicating that no alcohol functionality was present on carbon C5. Since an azide functionality was clearly present, as indicated by the IR spectrum, it seemed logical that the azide resided on carbon C5. Further scrutiny of the spectrums confirmed the observations discussed and left us with only one conclusion, i.e. azidoalcohol 116 is NOT formed under the reaction conditions found in the literature$^{108}$ (Scheme 40). Instead its regioisomer 118b was the main product (Figure 3).

![Diagram of azidoalcohol 116 and 118b](image)

**Figure 3.** Regioisomeric Azidoalcohols 116 and 118b
Figure 4. Triol 119b in DMSO-d$_6$ - Recorded at 270 MHz
Figure 5. Triol 119b in DMSO-$d_6$ With a Drop of D$_2$O - Recorded at 270 MHz
Figure 6. The expected Triol 117 and the Actual Triol formed 119b

The article,\textsuperscript{108} which contains the misassigned structure, describes a large variety of nucleophilic openings of epoxide 115. The authors state that all nucleophiles examined, except azide, opened epoxide 115 at the allylic site. After examining all of our data, we now believed that azide had also attacked at the allylic site, as all the other nucleophiles had. Recognizing this fact we were able to explain all of the inconsistencies we were finding with the spectral data we presumed was for triol 117 (Figure 6).

The article stated, that a homo allylic opening with azide (top pathway Scheme 43) had occurred, but we now knew that an allylic opening had occurred to give azidoalcohol 118b which we converted to triol 119b. The \textsuperscript{1}H NMR analysis revealed the regiochemistry of the product, but its absolute stereochemistry was still debatable.

Scheme 43

Reagents and Conditions: (i) NaN\textsubscript{3}, NH\textsubscript{4}Cl, DME/ethanol/water (3:3:2), 65 °C; (ii) Amberlyst 15 (wet) ion-exchange resin-strongly acidic, Aldrich Chemical Co.

While it was assumed that the epoxide 115 was opened in a S\textsubscript{N}2 'like' fashion by attack of azide, further proof was needed. Attempts to define the relative stereochemistry of the
azidoalcohol 119b by interpretation of the 1H NMR coupling constants of H_B and H_C (Figure 4 and 5), suggested a trans relationship between H_B and H_C (J= 7.8 Hz) and between H_C and H_D (J= 10.2 Hz). These data support structure 119b.

In order to unequivocally prove the regio- and stereochemistry of azidoalcohol 118b, a chemical correlation study was undertaken. Since benzylaminoalcohol 120b (Scheme 44) was a known compound,108 it was our intention to degrade both 120b and azidoalcohol 118b to trihydroxyamine hydrochloride 121. Hydrogenolysis of azidoalcohol 118b using Adams' catalyst afforded the ammonium salt 121 in 86% yield (Scheme 44). When benzylamino alcohol 120b was subjected to the same reaction conditions a mixture of products corresponding to different levels of reduction was obtained. Pearlman's catalyst (20% Pd(OH)_2) gave similar results. In contrast, catalytic transfer hydrogenolysis provided ammonium salt 121 in 72% yield. The two products were indistinguishable by 1H NMR, 13C NMR, and [α]_D comparison. This confirmed the structure of azidoalcohol 118b.

![Scheme 44](image)

Reagents and conditions: (i) 80 psi H_2, PtO_2, MeOH; (ii) ammonium formate, 10% Pd–C, CH_3OH reflux.

While the chemical correlation study was ongoing we were able to grow a suitable crystal of azidoalcohol 118b, for x-ray crystallographic analysis. The data obtained confirmed all of our previous findings (Figure 7).

It should be noted, that a large number of reaction conditions were examined in the hopes of synthesizing azidoalcohol 116 from epoxide 115, but all attempts failed. Azidoalcohol 116 was later synthesized in our group using an alternative route.106
2. Results

2.2 The Synthesis of Four Azidoalcohol Stereoisomers.

Having the structure of azidoalcohol 118b firmly established, we needed to modify our proposed synthesis which was no longer valid. The original synthetic proposal called for oxidative cleavage (treatment with O₃, followed by dimethylsulfide) of the C1–C6 double bond, of 116, and a reductive cleavage (treatment with NaIO₄, followed by NaBH₄) of the diol-moiety, C2-C3 (Scheme 45). The new strategy would require the reductive cleavage (treatment with O₃, then NaBH₄) of the C1-C6 double bond, in 118b, and oxidative cleavage (NaIO₄) of the diol-moiety (C2-C3) to afford the enantiomeric hemiacetal 122b (Scheme 45). Depending on the diastereomer of azidoalcohol 118 chosen, all four diastereomers of lactol 122 should be accessible.

![Scheme 45](image)

With the new synthetic strategy in hand, our goal became the synthesis of two isomeric epoxides, 115 and 125. Their nucleophilic opening and subsequent manipulation could open access to all four stereoisomers of azidoalcohol 118. Thus, methods were needed to synthesize both α- and β-epoxides 115 and 125 respectively (Scheme 46). Procedures leading to both epoxides 115¹⁰⁹ and 125¹¹⁰ had already been developed in our group. Full characterization of epoxide 125 and compounds leading to it were left until this project.
Scheme 46. Reagents and conditions: (i) 2,2-dimethoxypropane, catalytic \( \rho \)-TsOH, \( \text{CH}_2\text{Cl}_2 \); (ii) NBS, \( \text{DME/H}_2\text{O} \) (3:2), 0 °C; (iii) NaOH (1.1 equiv), \( \text{Bu}_4\text{NHSO}_4 \) (1.0 equiv), \( \text{CH}_2\text{Cl}_2 \) reflux; (iv) \( m \)-CPBA, \( \text{CH}_2\text{Cl}_2 \); (v) \( \text{NaN}_3, \text{NH}_4^+\text{Cl}^- \), \( \text{DME/MeOH/H}_2\text{O} \) (3:3:2), 65 °C; (vi) LiCl (5.0 equiv), ethyl acetoacetate (3.0 equiv), THF, 45 °C; (vii) LiBr (1.1 equiv), ethyl acetoacetate (2.0 equiv), THF, 35 °C; (viii) \( \text{NaN}_3 \) (3.0 equiv), DMF; (ix) \( \text{NaN}_3 \) (15 equiv), DMSO.
Exposure of acetonide 114 to N-bromosuccinimide (NBS), in the presence of H₂O, led predominately, but not exclusively, to bromohydrin 124 in 30% yield (Scheme 47). The minor isomer 127 was produced in 3% yield and was shown to cyclize to the known epoxide 115, to aid in its identification. The electrophilic bromine species attacks the more electron rich olefin, predominately from the sterically less hindered α-face. Subsequent nucleophilic attack by H₂O at the allylic site yields the corresponding trans bromohydrins 124 and 127. Their exposure to base furnished epoxides 125 and 115 respectively.

![Scheme 47](image)

Reagents and Conditions: (i) NBS, catalytic p-TsOH, 1,2-dimethoxyethane, H₂O; (ii) NaOH, Bu₄NHSO₄, CH₂Cl₂ reflux.

Azidoalcohols 118b and 118d should be directly accessible from epoxides 115 and 125 respectively (Scheme 46). Indeed, azidoalcohols 118b and 118d were obtained in high yield upon exposure of the epoxides (115 and 125) to NaN₃. The synthesis of the two remaining azidoalcohols 118a and 118c presented a greater challenge. Inversion of stereochemistry at the allylic site of halohydrins 128 and 129 would be necessary to form the cis azidoalcohols 118a and 118c.

When bromohydrin 128 was treated with NaN₃ (Table 8) an intra- and intermolecular competition of epoxide formation vs azide introduction ensued. This competition could be controlled by varying the concentration of NaN₃ present. Surprisingly, depending on the excess/concentration of sodium azide used, azidoalcohol 118a or 118d could be obtained as the major product. When the concentration of azide did not greatly exceed that of the bromohydrin 128, intramolecular closure prevailed and epoxide 125 was formed. Subsequent opening of the epoxide, with azide, yields the trans azidoalcohol 118d (Scheme 48). On the other hand a large excess of NaN₃, compared to the halohydrin, leads preferentially to an intermolecular displacement of the halogen by azide and afforded the cis azidoalcohol 118a as the major product (Table 8).
Table 8. Reactivity of Halohydrins to Sodium Azide

<table>
<thead>
<tr>
<th>Halohydrin, where X equals</th>
<th>Molarity of NaN₃</th>
<th>Molarity of NaN₃/SM</th>
<th>Equiv. ratio</th>
<th>Solvent</th>
<th>Final temp.</th>
<th>% of 118a</th>
<th>% of 118d</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chlorine</td>
<td>0.18</td>
<td>0.70</td>
<td>3.89</td>
<td>DMF</td>
<td>85 °C</td>
<td>12.7</td>
<td>77.3</td>
</tr>
<tr>
<td>Bromine</td>
<td>0.20</td>
<td>0.80</td>
<td>4.00</td>
<td>DMF</td>
<td>90 °C</td>
<td>14.4</td>
<td>75.6</td>
</tr>
<tr>
<td>Chlorine</td>
<td>0.19</td>
<td>5.7</td>
<td>30.0</td>
<td>DMSO</td>
<td>40 °C†</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Bromine</td>
<td>0.61</td>
<td>9.7</td>
<td>16.0</td>
<td>DMSO</td>
<td>40 °C†</td>
<td>74.8</td>
<td>21.8</td>
</tr>
</tbody>
</table>

SM starting material (halohydrin); † An ultrasound sonicator was used to maintain homogeneity. The temperature of the bath was 40 °C.

Before the optimum conditions (last row of Table 8) to synthesize 118a were found, a variety of different approaches were examined. Thus, attempts at oxidizing azidoalcohol 118b (Scheme 46) to the corresponding ketone and then reducing it with a hydride reagent preferentially from the least hindered, α-face, failed at the ketone stage. The alcohol of 118b turned out to be highly resistant to oxidation, Swern conditions and tetra-n-propylammonium per ruthenate (TPAP) failed to produce the ketone. In still another attempt to exploit 118b, the tosylate was formed. Treatment of the tosylate with a nucleophilic source of oxygen (NaOH and KNO₂) also failed to produce azidoalcohol 118a.

Attempts to protect the alcohol moiety of 128 (Scheme 48), prior to bromide displacement by azide, also failed. The acetyl, trimethylsilyl and benzyl protecting groups were investigated. Formation of the acetate proceeded in high yield, but attack by azide proved troublesome. Slow reaction times and a complex mixture of products forced us to abandon this work. Treatment of the trimethylsilyl ether of bromohydrin 128 with azide resulted in deprotection of the silyl group. Attempts to form the benzyl ether of 128 were unsuccessful, only the β-epoxide 125 was formed.

In yet another attempt to synthesize azidoalcohol 118a, β-epoxide 125 was treated with iodine, and triphenylphosphine in CH₂Cl₂ to give the corresponding iodo hydridr. The iodo hydridr showed visible signs of decomposition after 20 minutes at room temperature. However if immediately treated with sodium azide in DMSO, the desired azidoalcohol 118a was
exclusively formed. Even though this sequence was fruitful, the reaction conditions shown in Table 8 were more reliable.

The synthesis of the last azidoalcohol 118d went effortlessly, in 85%, via the direct opening of epoxide 125 with NaN₃ (Scheme 46). Interestingly, 118d could also be accessed directly from bromohydrin 124¹¹⁴ (Scheme 49). The spectral data for the two independent synthesizes of azido alcohol 118d were indistinguishable. When 118d is synthesized from bromohydrin 124, the overall yield from chlorodiol 113 is 27%. Using the route shown in Scheme 46 the overall yield from chlorodiol 113 is 13%.

\[
\begin{align*}
\text{Cl} & \quad \text{HO} \\
\begin{array}{c}
\text{Br} \quad 124 \\
\end{array} & \quad \text{NaN₃, DMSO} \\
\text{70 °C, 90% yield} & \quad \text{N₃} \\
\text{OH} \quad 118d \\
\end{align*}
\]

**Scheme 49**

In an attempt to take advantage of this unique reaction (Scheme 49) bromohydrin 124 was treated with NaBr in DMSO in the hopes of forming bromohydrin 128 (Scheme 46). Unfortunately none of the desired bromohydrin 128 was observed. Similar results were observed when NaCl was used.

That all four azidoalcohols 118a-d were accesible from chlorodiol 113, was now firmly established. The efficiency of their synthesis is shown in Table 9. It was now our intent to synthesize a stereoisomer of sphingosine. To this end we chose 118b to be carried through to L-threo-sphingosine.
<table>
<thead>
<tr>
<th>Compound</th>
<th>Number of steps from chlorodiol 113</th>
<th>Overall % yield from chlorodiol 113</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image" alt="Chemical Structure 118a" /></td>
<td>five steps</td>
<td>11%</td>
</tr>
<tr>
<td><img src="image" alt="Chemical Structure 118b" /></td>
<td>three steps</td>
<td>48%</td>
</tr>
<tr>
<td><img src="image" alt="Chemical Structure 118c" /></td>
<td>four steps</td>
<td>53%</td>
</tr>
<tr>
<td><img src="image" alt="Chemical Structure 118d" /></td>
<td>three steps</td>
<td>28%</td>
</tr>
</tbody>
</table>
2. Results

2.3 Exhaustive Cleavage of Azidoalcohols 7a & 7b -
Synthesis of D-erythro-C\textsubscript{18} and L-threo-C\textsubscript{18}-Sphingosine.

Since azido alcohol \textbf{118b} could be synthesized in high yield it was chosen for our initial investigation, which would ultimately lead to the unnatural L-threo-sphingosine. Content that L-threo-sphingosine could be synthesized from an aldehyde of type \textbf{D} (Figure 8), several possibilities for its synthesis were considered. Ozonolysis of the C1-C6 double bond in compound \textbf{A} would lead to compounds of type \textbf{B} and subsequent periodate cleavage (C2-C3), after deprotection, would yield aldehyde \textbf{D}. Alternatively, periodate cleavage of \textbf{118b} would form compounds of type \textbf{C}, ozonolysis of which should yield aldehyde \textbf{D}. Compound \textbf{D} the open from of lactol \textbf{122} would then be subjected to Wittig olefination, providing the skeleton of sphingosine. (Note: For the synthesis of a particular stereoisomer of sphingosine, all compounds leading to its synthesis will carry the same letter designation. If another stereoisomer of sphingosine is synthesized using stereoisomeric compounds they will have the same numbers, but a different letter. For example all compounds which could potentially led to the synthesis of D-erythro-sphingosine \textbf{1a} will bear the letter "a".)
Figure 8. Proposed Route to Sphingosine.
To this end, alcohol 118b was protected as an acetate 130b to provide the first substrate for the ring cleavage experiments. Scheme 50 summarizes the cleavage chemistry performed. Acetonide 130b was deprotected to afford diol 131b, which upon treatment with NaIO₄ in H₂O gave at least four compounds as evidenced by the complicated ¹H NMR spectrum of the crude product mixture. Readily identifiable aldehyde and lactol resonances suggested the presence of dialdehyde 132b, along with a cyclic dihemiacetal (presumably via the addition of one equivalent of water, Scheme 51). An attempt to simplify the reaction product mixture via reduction (NaBH₄, both normal and Luche's conditions¹¹⁵) proved fruitless and the route was abandoned.

![Diagram](image)

**Scheme 51**

Disappointed by the outcome of the periodate cleavage reaction, we focused our attention on the cleavage of the vinyl chloride moiety in 131b. Ozonolysis in MeOH, followed by reductive work-up with NaBH₄, did not yield methyl ester 133b (Scheme 50). The large number of products formed (as evidenced by the TLC) made this particular route unattractive for our goal.

Since we were unable to synthesize any compounds of interest from 131b, we decided to examine the cleavage of the vinyl chloride moiety in 130b. Ozonolysis (methanol, 5.0 equivalents of sodium bicarbonate), followed by NaBH₄ work-up, surprisingly gave vinyl azide 135b (Scheme 50). Evidently an elimination occurred, in which acetate served as the leaving group.
Scheme 50. Reagents and Conditions: (i) Amberlyst 15 (wet) ion-exchange resin, strongly acidic, Aldrich Chemical Co., (ii) 1.0 M HCl, 80% H₂O, 20 °C, (iii) i NaOAc, H₂O, 16 °C, (iv) 1.0 M O₂ (excess), (v) i NaOH, MeOH, rt, (vi) NaBH₄, MeOH, rt.

Reagents: (i) Amberlyst 15 (wet) ion-exchange resin, strongly acidic, Aldrich Chemical Co., (ii) 1.0 M HCl, 80% H₂O, 20 °C, (iii) i NaOAc, H₂O, 16 °C, (iv) 1.0 M O₂ (excess), (v) i NaOH, MeOH, rt, (vi) NaBH₄, MeOH, rt.
The stage at which this elimination occurred was not clear. Ozonolysis of compounds containing a vinyl chloride moiety (e.g. 130b) generate HCl when the latent alkanoyl chloride is attacked by methanol, the solvent. To maintain a pH close to neutral, sodium bicarbonate is sometimes added. In this particular case, after NaBH₄ work-up of the ozonide at -20 °C, sodium bicarbonate or sodium carbonate could have easily deprotonated the acidic α-proton of aldehyde 137b (Scheme 50). Elimination of acetate via the enolate of 137b might explain the formation of the α,β-unsaturated aldehyde 135b.

This hypothesis is strengthened by the observation that in the absence of sodium bicarbonate aldehyde 137b is the main product (none of the elimination product 135b was found). Interestingly even under the forceful conditions of the excess NaBH₄ the ozonide reduction, of 130b, always provided the aldehyde 137b and none of the corresponding alcohol. However, if the crude aldehyde 137b was isolated (after acidic work-up - pH= 3.5) and treated with NaBH₄ at 0 °C, a seven-membered lactol 139b ensued. If the reduction was carried out at room temperature a five-membered lactol 138b was formed.

The cleavage experiments performed on acetate 130b (Scheme 50) provided us with further important information. When sodium cyanoborohydride was used to reduce the ozonide of 130b, methyl ester 134b was isolated in 82% yield. We expected the corresponding vicinal diol 133b, obtained from acetonide hydrolysis of 134b, to provide lactol 122b when treated with NaIO₄ (Scheme 52). Unfortunately, after the acetonide protecting group was removed, a migration of the acetyl group occurred. Several different reaction conditions were examined, but no improvements were observed. The propensity of this acetate to migrate made isolation of triol 133b impossible.

![Scheme 52](image)

At this point it became clear that a different protecting group had to be utilized. When the ozonide of 130b was reductively worked-up in the absence of sodium bicarbonate, lactol 138b was obtained (Scheme 50). Obviously the acetate protecting group of 130b was also cleaved under the reaction conditions, and the resulting free alcohol participated in an intramolecular
lactolization. The possibility of an "intramolecular protection" of this alcohol was extremely appealing and thus considered further.

Thorough examination of this "protecting group" revealed an obvious problem. At the stage of compound 141b (Scheme 53) the "protecting group" would be connected to the alcohol via a hemiacetal bond. Thus, once the vicinal diol of 140b was cleaved, the protecting group would be readily hydrolyzed under the reaction conditions. In an attempt to overcome this lability problem, compound 144b was considered. At the oxidation level of an ester, the glyoxalate moiety (in square) might be stable under the reaction conditions and yield lactol 122b, where R = glyoxalate. Investigation of the synthesis of 142b therefore appeared worthwhile.

![Scheme 53](image)

Reagents and conditions: (i) Amberlyst 15 (wet) ion-exchange resin-strongly acidic, Aldrich Chemical Co.; (ii) NaIO₄, H₂O.

Since the C-4 acetate was cleaved during the NaBH₄ work-up of the ozonide of 130b, we expected direct ozonolysis of alcohol 118b to provide lactol 138b in an analogous fashion. Indeed, under these conditions ozonolysis of alcohol 118b afforded lactol 138b in yields varying from 60 - 80%. To obtain lactone 142b, the ozonide of 118b was reduced with sodium cyanoborohydride¹¹⁶ (Scheme 54). Acid catalyzed hydrolysis of the acetonide in 142b followed by periodate cleavage failed to give 122b (R = glyoxalate, Scheme 53), but instead yielded 122b (R = H). Even though our initial strategy warranted protection of the hydroxy group in lactol 122b, where R = H, we decided to evaluate its potential as a Wittig substrate.
Scheme 54
Reagents and conditions: (i) O₃ (excess), MeOH, -78 °C; (ii) NaBH₄, MeOH, -55 °C to rt; (iii) NaBH₃CN, MeOH, pH = 3.0, 0 °C.

Wittig olefination, completing the carbon skeleton of sphingosines, was examined in great detail. Even though a control reaction, using known lactol 145 (synthesized from the chlorodiol 113, 117 Table 10) gave olefins 146 and 147 in almost quantitative yield, with substrate 122b (R = H) the overall yield of azidosphingosine was never greater than 30% (Table 11). In theory, a minimum of three equivalents of base and tetradeceyltriphénylphosphonium bromide is required for the reaction. The results suggest that a slight excess of the phosphonium salt is required or the reaction yield will be seriously compromised.

Azidoalcohol 118a (precursor to naturally occurring D-erythro-sphingosine) was exhaustively cleaved, in the same manner as 118b, to give the diastereomeric lactol 122a. Thus 118a was converted to lactol 138a, which was then transformed into lactol 122a (see top pathway, Scheme 53, for reaction type). When lactol 122a (Table 11) was treated under the same optimized Wittig olefination conditions as 122b similar yields of azidosphingosines 148a and 149a were observed.
Table 10. Wittig Olefination of Lactol 48

<table>
<thead>
<tr>
<th>Equiv of salt§</th>
<th>Equiv of base</th>
<th>Temperature</th>
<th>Solvent</th>
<th>% Yield Z/E</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.2</td>
<td>2.25 of n-BuLi</td>
<td>room temp.</td>
<td>THF</td>
<td>80 / 15</td>
</tr>
<tr>
<td>2.2</td>
<td>4.12 PhenylLi</td>
<td>-35°C</td>
<td>THF</td>
<td>6 / 61</td>
</tr>
</tbody>
</table>

§ Equiv of salt, refers to equivalents of tetradecyltriphenylphosphonium bromide.

Table 11. Wittig Olefination of Lactol 122

<table>
<thead>
<tr>
<th>Lactol</th>
<th>Ref.</th>
<th>Equiv of salt§</th>
<th>Equiv of base</th>
<th>Temperature</th>
<th>Solvent</th>
<th>% Yield Z/E</th>
</tr>
</thead>
<tbody>
<tr>
<td>122b</td>
<td>117a</td>
<td>4.0</td>
<td>4.0 n-BuLi</td>
<td>room temp.</td>
<td>THF</td>
<td>Δ</td>
</tr>
<tr>
<td>122b</td>
<td>118†</td>
<td>1.15</td>
<td>4.6 PhenylLi</td>
<td>-35 °C</td>
<td>THF/toluene</td>
<td>0.9 / 1.8</td>
</tr>
<tr>
<td>122b</td>
<td>†</td>
<td>1.20</td>
<td>5.5 PhenylLi</td>
<td>-78 °C</td>
<td>THF/toluene</td>
<td>Δ</td>
</tr>
<tr>
<td>122b</td>
<td>119*</td>
<td>4.0</td>
<td>8.0 Dimsyl anion</td>
<td>room temp.</td>
<td>DMSO</td>
<td>Δ</td>
</tr>
<tr>
<td>122b</td>
<td>120</td>
<td>2.0</td>
<td>2.0 Na amylate</td>
<td>room temp.</td>
<td>THF/toluene</td>
<td>10 / 7</td>
</tr>
<tr>
<td>122b</td>
<td>122a</td>
<td>3.2</td>
<td>2.7 n-BuLi</td>
<td>room temp.</td>
<td>THF</td>
<td>24 / 6†</td>
</tr>
<tr>
<td>122b</td>
<td>3.2</td>
<td>2.7 n-BuLi</td>
<td>room temp.</td>
<td>THF</td>
<td>14 / 4</td>
<td></td>
</tr>
</tbody>
</table>

§ Equiv of salt, refers to equivalents of tetradecyltriphenylphosphonium bromide. † Denotes modified Schlosser-Wittig conditions. * The dimsyl anion was generated by the addition of 8.0 equiv of NaH to DMSO (the reaction solvent). ‡ Note this yield was not repeatable, subsequent reactions gave yields very similar to those of lactol 122a, i.e. combined yields (Z and E) of 20%. Δ Neither cis- or trans-azidosphingosine were observed.
It is known that aldehydes with α-hydroxy groups generally give poor yields in Wittig reactions.\textsuperscript{121} This alone could account for our poor yields, but we also questioned the inherent stability of these lactols under the basic conditions of a Wittig reaction. Although many Wittig reactions have been performed on lactols,\textsuperscript{119} none could be located in the literature with a disposition of functional groups found in our lactols (Figure 9). Under the reaction conditions it was assumed that both acidic protons were removed. Once the lactol was opened, structure F, an intramolecular nucleophilic displacement of azide could ensue, forming epoxides G and H. Due to the insolubility of LiN\textsubscript{3} or NaN\textsubscript{3} in the reaction medium, azide would effectively be removed from the reaction. Further experiments and data collection would be required to support the suppositions of Figure 9. Possibly, the phosphonium ylide itself may attack the azide, complicating the picture further. Generally, these Wittig reactions did not show a satisfactory mass recovery.

\textbf{Figure 9. Possible Decomposition Route of Lactol 122b}

\[ \text{Sphingosine contains a } \text{trans} \text{ olefin, yet the Wittig reactions we performed gave predominantly the } \text{cis} \text{-azidosphingosine product. Therefore } \text{cis} \text{-azidosphingosines 148a (D-erythro-series) and 148b (L-threo-series) were photoisomerized to } \text{trans} \text{-azidosphingosines 149a and 149b by means of a Hanovia 450 W lamp, Pyrex filter, and diphenyl disulfide.}^{79}\text{ Reduction of } \text{trans} \text{-azidosphingosine 149b with hydrogensulfide gave L-threo-sphingosine 1b, which was not isolated, but instead treated with acetic anhydride and pyridine to provide its triacetate, indistinguishable from an authentic sample. } \text{trans} \text{-Azidosphingosine 149a, a known compound, displayed } ^1\text{H NMR and } [\alpha]_D^{24} = -34.2^\circ \text{ (c 1.58, CHCl}_3)\text{ [lit.}^{122}[\alpha]_D^{20} = -32.9^\circ \text{ (c 4.0, CHCl}_3)]\text{ in agreement with the literature values. The synthesis of D-erythro-azidosphingosine 149a constitutes a formal synthesis of D-erythro-sphingosine.} \]
2. Results

2.4 Exhaustive Cleavage of Azidoalcohol 118d - Synthesis of D-threo-C18 Sphingosine

To meet our goal of synthesizing all four stereoisomers of sphingosine, D-threo- and L-erythro-sphingosine needed to be synthesized. This goal could have been met by repeating the protocols developed for the synthesis of the first two sphingosines. Instead we embarked on a second generation synthesis of sphingosine. Our original goal of synthesizing a lactol of type 122 with the alcohol protected had never been met. With this objective in mind we set our sights on such a synthesis, in the hopes of improving the Wittig olefination reaction. The protecting group chosen would have to be stable under the acetonide deprotection conditions, as well as the basic conditions of a Wittig reaction, and finally be easily removed at the azidosphingosine stage (Scheme 55).

![Scheme 55]

It was decided that a tert-butyldiphenylsilyl (TBDPS) ether protecting group would fulfill the requirements necessary. To this end, alcohol 118d was protected as its tert-butyldiphenylsilyl ether (TBDPS ether) 150d and subsequently treated with ozone, to provide methyl ester 151d (Scheme 56).
Scheme 56
Reagents and conditions: (i) tert-butyldiphenylsilyl chloride (1.97 equiv), imidazole (2.39 equiv), THF; (ii)
(1) O₃ (excess), MeOH, -78 °C, (2) NaBH₄ (excess), MeOH, 0 °C to room temperature.

Acetonide 151d, upon treatment with Amberlyst 15 (wet) strongly acidic ion-exchange resin, provided the desired vicinal diol 152d, although in poor yield (44%). The remaining mass-balance consisted of three new compounds (153d - 155d, Scheme 57). Full characterization of these new compounds showed that deprotection of the acetonide was not a problem. Instead it suggested that the product, 152d, was inherently unstable under the acidic reaction conditions. The product distribution was essentially unchanged when THF/H₂O/CH₃CO₂H, THF/H₂O/CF₃CO₂H (see Scheme 58 for an example found in the literature¹²⁴); and/or CH₃OH/H₂O/HCl systems were examined. This result was unexpected, especially in light of the result shown in Scheme 58.¹²⁴ In any event the products 152d through 155d are readily explained. It seems almost without question that deprotection of the acetonide to form vicinal diol 152d occurs first. The TBDPS group then migrating to the adjacent alcohol. This migration provides what we believe is triol 153d, based on the ¹H NMR spectrum. If triol 42d is stored neat or exposed to silica gel (short columns of silica gel and fast elution are advised for purification) lactones 154d and 154d are formed. Full characterization of diol 153d was not attempted because of its unstable nature. It is interesting to note that when pure lactone 154d or 155d was placed in DMSO-d₆, a 1:1 ratio of the two lactones (154d and 155d) immediately ensued, as evidenced by ¹H NMR, and that this ratio does not change over a period of a couple of days. Yet when pure lactone 154d or 155d is placed in CDCl₃ no sign of the other lactone is evident by ¹H NMR.
Scheme 57

Acidic reaction conditions
In an attempt to thwart the silyl migration of diol 152d, some nontraditional reagents were examined. Thus when 151d was treated with dimethylaluminum chloride the results were very encouraging (TLC), but upon work-up, the four products were found as usual. Finally, when 1% I₂ in MeOH (w/v) was employed an 80% yield of 152d was obtained. No further optimization of this reaction was carried out.

![Scheme 58](image)

Reagents and conditions: (i) THF/H₂O/CF₃CO₂H (20:5:1), 0 °C → rt.

Proceeding with the synthesis, vicinal diol 152d was treated with NaIO₄ to provide lactol 156d in 75% yield (Scheme 59). The silyl migration product 157d was also observed.

![Scheme 59](image)

From azidoalcohol 118d, TBDPS protected lactol 156d was synthesized in 46% overall yield. For comparison lactol 122b, R = H, was synthesized from azidoalcohol 118b in the same overall yield, 46%. Disappointed with the results of earlier Wittig olefination reactions, we looked forward to the Wittig olefination of lactol 156d with great anticipation. Confident that success was at hand, modified Schlosser-Wittig conditions (2.2 equiv of phosphonium salt, 4.40 equiv of phenyllithium) were examined first (Table 12). These conditions destroyed the starting lactol and none of the desired product was observed. Amazed by the reactivity of this lactol, the previous conditions (Table 11) using sodium amylate and n-BuLi were examined. Thus the best conditions afforded a combined yield of cis- and trans-azidosphingosine 148d and 149d in 13% after Wittig olefination and tert-butylidiphenylsilyl ether cleavage. Photoisomerization of cis-azidosphingosine 148d provided trans-azidosphingosine 149d and was found to have ¹H NMR and [α]D²⁵ = -2.4° (c 0.27, CHCl₃) in agreement with the literature values.93
Table 12. Wittig Olefination Conditions Performed on Lactol 156d (R = TBDPS)

![Chemical Structures](image)

<table>
<thead>
<tr>
<th>Equiv of salt</th>
<th>Equiv of base</th>
<th>Temperature</th>
<th>Solvent</th>
<th>158d &amp; 159d</th>
<th>Dimer †</th>
<th>Silanol †</th>
<th>trans-2-hexadecene</th>
<th>PØ₃</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.20</td>
<td>4.40 PhenylLi</td>
<td>-50 to 0 °C</td>
<td>THF</td>
<td>0%</td>
<td>§</td>
<td>57%</td>
<td>not present</td>
<td>§</td>
</tr>
<tr>
<td>1.77</td>
<td>1.77 Na amylate</td>
<td>rt</td>
<td>THF</td>
<td>9.0%</td>
<td>§</td>
<td>43%</td>
<td>§</td>
<td>§</td>
</tr>
<tr>
<td>1.77</td>
<td>1.77 Na amylate</td>
<td>0 °C to rt</td>
<td>THF</td>
<td>3.7% ‡</td>
<td>20.0%</td>
<td>47%</td>
<td>§</td>
<td>§</td>
</tr>
<tr>
<td>3.50</td>
<td>3.50 Na amylate</td>
<td>0 °C to rt</td>
<td>THF</td>
<td>13% ‡</td>
<td>19.1%</td>
<td>=19%</td>
<td>12%</td>
<td>11.5%</td>
</tr>
<tr>
<td>2.20</td>
<td>2.0 of nBuLi</td>
<td>rt</td>
<td>THF</td>
<td>0%</td>
<td>§</td>
<td>66%</td>
<td>not present</td>
<td>§</td>
</tr>
<tr>
<td>3.00</td>
<td>2.7 of nBuLi</td>
<td>rt</td>
<td>THF*</td>
<td>=5%</td>
<td>=5%</td>
<td>=40%</td>
<td>not present</td>
<td>=5%</td>
</tr>
<tr>
<td>3.50</td>
<td>3.2 of nBuLi</td>
<td>-78 °C to rt</td>
<td>Toluene</td>
<td>0%Δ</td>
<td>20.0%</td>
<td>§</td>
<td>not present</td>
<td>§</td>
</tr>
<tr>
<td>4.00</td>
<td>3.6 of nBuLi</td>
<td>rt</td>
<td>THF</td>
<td>0%ΔΔ</td>
<td>§</td>
<td>§</td>
<td>not present</td>
<td>§</td>
</tr>
</tbody>
</table>

† The dimer refers to 14-octacosene (C₂₈H₅₆). † The silanol is tert-butyldiphenylsilanol. § Was not looking for or did not isolate. ‡ Percent yield determined by treating the crude product with F⁻ and isolating the corresponding azidosphingosine. * Deoxygenated the solvent. Δ 36% of the silylmigrated product 157d and 35% of starting lactol isolated. ΔΔ TLC shows silylmigrated product 157d, within minutes, after 15 hrs neither starting lactol or 157d remains.
A common observation while performing these Wittig reactions was that of silyl migration. After five minutes of reaction time compound 157d (Scheme 59) was always present in the reaction mixture. The work-up of these reactions was not a trivial task. Simple addition of water caused silyl ether cleavage. In an effort to suppress this unwanted silyl cleavage subsequent reactions were chilled to -50 °C before quenching with saturated NH₄Cl thus suppressing the silyl ether cleavage.

Finally the formation of a dimer was noted, namely 14-octacosene (C₂₈H₅₆). Closer scrutiny of the reaction conditions revealed, that it was forming during the phosphonium ylide formation, i.e. before the lactol was added. The normal protocol for Wittig formation was as follows. To tetradecyltriphenylphosphonium bromide, under argon, freshly distilled solvent was added. After cooling this solution in an ice-bath, base was added. Regardless of the base used a color change ensued (clear to a dark orange or yellow). The phosphonium ylide was then stirred for 30 minutes at 0 °C, at which time the ice-bath was removed. Once at room temperature the reaction was stirred for an additional 30 minutes. The temperature of the ylide solution was then adjusted to the desired reaction temperature. It has been suggested that perhaps dissolved oxygen is the culprit of this dimer formation (Scheme 60). Deoxygenation of the solvent diminished the dimer formation (Table 12, entry 6). Further experimentation would be needed to draw more accurate conclusions.

Scheme 60
IV. SUMMARY, CONCLUSION and OUTLOOK

The utility of chlorodiol 113 as a general synthon for sphingosine synthesis has been unequivocally shown. The synthesis of three out of the four possible stereoisomers of sphingosine from one starting material is a feat in its own, and bodes well for the enantiodivergent accessibility of the fourth isomer from chlorodiol 113.

At present the Achilles’ tendon of this total synthesis is the Wittig olefination of lactols 122a, 122b, and 156d. To date the olefination has been plagued by a myriad of set backs. Further work after this dissertation will try to address some of the following questions. Are lactols of type 122 and 156 inherently unstable to basic reaction conditions? Is the azide attacked by the phosphonium ylides? Can a stable protecting group be found and still be easily removed at the end of the synthesis? If an appropriate protecting group is found will it increase the Wittig olefination yields? These are few of the questions which need to be addressed.

The fourth isomer will be synthesized after the this dissertation is submitted. The strategy will entail protection of azidoalcohol 118c as a benzyl ether. Using the same set of reactions used on the previous isomers, lactol 122c, where R = benzyl, will be subjected to the Wittig olefination. If no improvement in yield is apparent, the azide (of lactol 122c) will be reduced and protected and this new lactol will then be subjected to Wittig olefination. Depending on the outcome, most of the above questions will be answered. Below are Schemes 61 and 62 which summarize what has been accomplished during this investigation.
Scheme 61
Scheme 62

**122a**

**122b**

**122c**

**156d**

**148a & 149a**

**148b & 149b**

**148c & 149c**

**148d & 149d**

Triacetate of D-erythro-sphingosine

Triacetate of L-threo-sphingosine

Triacetate of L-erythro-sphingosine

Triacetate of D-threo-sphingosine
V. EXPERIMENTAL

General: All reactions were carried out in an argon atmosphere with standard techniques for the exclusion of air and moisture. Glassware used for moisture sensitive reactions were flame dried under vacuum. All solvents were reagent grade. Anhydrous solvents were dried immediately before use. THF and toluene were distilled from sodium benzophenone ketyl. A one-piece reflux apparatus was used when reflux conditions were needed (it was custom-made in a glass shop and consists of a jointless connection of a round bottom flask and a reflux condenser), it can be substituted for by using a condenser and a round bottom flask.

Dry oxygen containing about 2.5% ozone was introduced at a rate of 4L/min into the solution of a substrate.

TLC plates were visualized by immersion in a vanillin stain, followed by warming on a hot plate. Flash chromatography was carried out on Merck Kieselgel 60 silica gel (230 - 400 mesh). The impure products were impregnated on silica gel and then loaded onto the column.

$^1$H NMR and APT $^{13}$C NMR were recorded at 270 MHz and 68 MHz. Proton and carbon chemical shifts are reported in parts per million (ppm) relative to CDCl$_3$ ($^1$H NMR, 7.24 and $^{13}$C NMR, 77.0 ppm - middle peak of the triplet). Elemental analyses were performed by Atlantic Microlabs, Inc.

(3S,4S,5S,6S)-3-azido-1-chloro-5,6-O-isopropylidene-1-cyclohexene-4,5,6-triol (118a). To a 50 mL Erlenmeyer flask was added bromohydrin 128 (6.039 g, 21.30 mmol), NaN$_3$ (22.00 g, 338.4 mmol, 15.9 eq) and then DMSO (35.0 mL). The mixture solidified immediately, so it was broken into much smaller pieces (CAUTION: A metal spatula can initiate an explosion with dry NaN$_3$ and possibly with these thick pastes of NaN$_3$). The ensuing thick paste was sonicated for 1.5 hrs and then more DMSO (15.0 mL) was added. The reaction vessel was sonicated for another 2.5 hrs, at which point no bromohydrin 128 was apparent by TLC. Distilled H$_2$O was added until all the NaN$_3$ dissolved and then the solution was extracted with EtOAc (X5). The combined organics were dried with MgSO$_4$, filtered, and volatiles removed. Column chromatography using gradient elution (hexanes/EtOAc, 8.5:1.5 and ending with 7:3) gave azidoalcohol 118a (3.915 g, 15.94 mmol) as a clear oil in 74.8% yield and an epimeric azidoalcohol [(3R,4S,5S,6S)-3-azido-1-chloro-5,6-O-isopropylidene-1-cyclohexene-4,5,6-triol (118d)] (1.143 g, 4.652 mmol) as a brown oil (pure by $^1$H NMR) in 21.8% yield. $R_f$ = 0.35 (hexanes/EtOAc, 2:1); [α]$_D^{23}$ = +288.8 (c 1.06, CHCl$_3$); IR (neat) ν 3420, 3060, 2995, 2910, 2090, 1635 cm$^{-1}$; $^1$H NMR (CDCl$_3$) δ 6.00 (d, J = 5.0 Hz, 1H), 4.49 (m, 2H), 4.02 (m, 2H), 2.77 (br.s, 1H), 1.51 (s, 3H), 1.39 (s, 3H); $^{13}$C NMR (CDCl$_3$) δ 137.4 (C), 111.6 (C), 121.7 (CH), 75.9 (CH), 75.4 (CH), 67.0 (CH), 58.1 (CH), 27.4 (CH$_3$), 25.7 (CH$_3$); MS (Cl, 70 eV) $m/z$ (rel. intensity) 246 (M$^{+}$+1, 3.0), 232 (19), 230 (53), 220 (42), 218 (92),
205 (23), 203 (60), 182 (39), 59 (100); **Anal. calc**d for C$_9$H$_{12}$ClN$_3$O$_3$: C, 44.00; H, 4.92; N, 17.1. **Found:** C, 43.91; H, 4.93; N, 16.93.

(3R,4S,5S,6S)-3-azido-1-chloro-5,6-O-isopropylidene-1-cyclohexene-4,5,6-triol (118d). Epoxide 125 (250 mg, 1.23 mmol), 1,2-dimethoxyethane (13.0 mL), ethanol (10.0 mL), H$_2$O (8.0 mL), Na$_2$N$_3$ (320 mg, 4.92 mmol, 4.00 equiv), and NH$_4$Cl (264 mg, 4.94 mmol, 4.00 equiv) were added to a rb flask in the order stated. The rb flask was equipped with a reflux condenser and the apparatus placed in an oil bath at 55 °C. After 60 min the reaction was allowed to cool to rt and then had H$_2$O (40.0 mL) added to it. This was extracted with EtOAc (X3), the organic extracts were combined, dried with MgSO$_4$, filtered, and volatile organics removed. Column chromatography of this crude product using gradient elution (Hex/EtOAc 3:1 → 2:1) afforded azido alcohol (118d) (262.1 mg, 1.07 mmol) in 87% yield as a clear oil. This clear oil turns to a dark brown oil within 5 hrs at rt, yet no decomposition is noticable by $^1$H NMR. An analytical sample was obtained using column chromatography with an acetone/CH$_2$Cl$_2$ solvent system (1% acetone in CH$_2$Cl$_2$ → 2% acetone in CH$_2$Cl$_2$). $R_f$ = .45 (hexanes/EtOAc, 2:1); $[\alpha]_D^{22}$ = -120.1 (c 1.00, CHCl$_3$); IR (neat) ν 3430, 2995, 2930, 2895, 2100, 1645, 1225, 1040 cm$^{-1}$; $^1$H NMR (CDCl$_3$) δ 5.81 (d, J = 2.0 Hz, 1H), 4.57 (m, 2H), 4.27 (td, J = 1.8, 8.6 Hz, 1H), 3.81 (dt, J = 2.1, 8.1 Hz, 1H), 2.54 (d, J = 7.5 Hz, -OH), 1.44 (s, 3H), 1.43 (s, 3H);

$^{13}$C NMR (CDCl$_3$) δ 134.2 (C), 111.1 (C), 123.9 (CH), 76.7 (CH), 76.3 (CH), 72.0 (CH), 60.9 (CH), 27.3 (CH$_3$), 26.3 (CH$_3$); MS (Cl, 70 eV) $m/z$ (rel. intensity) 246 (M$^+$+1, 3.0), 160 (3.0), 145 (20), 133 (3.0), 117 (3.0), 114 (4.0), 101 (15.0), 96 (15.0), 59 (100); **Anal. calc**d for C$_9$H$_{12}$ClN$_3$O$_3$: C, 44.00; H, 4.92; N, 17.11. **Found:** C, 44.34; H, 4.96; N, 16.96.

(1R, 2S, 3S, 6S)-6-Azido-4-chloro-cyclohex-4-ene-1,2,3-triol (119b). To a solution of azidoalcohol (118b) (513 mg, 2.088 mmol) in CH$_3$OH (25 mL) was added 1.6 grams of Amberlyst 15(wet) ion-exchange resin. After 8 days no more azidoalcohol remained by TLC. The solution was filtered through a pad of celite and the solvent was evaporated to give 453.9 mg of crude product. Purification by column chromatography (hexane/ethylacetate, 1:4) gave 11mg of starting material and 405.7 mg (1.973 mmol, 97%) of azidotriol as a white solid. For data collection a portion was recrystallized from ethylacetate. $R_f$ = 0.47 (Ethylacetate); mp 137-138 °C; $[\alpha]_D^{20}$ = -11.0 (c 1.1, CH$_3$OH); IR (KBr) ν 3290, 3190, 2900, 2840, 2060, 1640, 1430, 1240 cm$^{-1}$; $^1$H NMR (DMSO) δ 5.72 (1H, d, J 2.6), 5.51 (1H, d, J 6.1), 5.38 (1H, d, J 5.2), 4.98 (1H, d, J 6.1), 3.98 (1H, m, J 4.1, 6.1), 3.93 (1H, m, J 2.6, 7.8), 3.62 (1H, ddd, J 5.2, 7.8, 10.2), 3.45 (1H, ddd, J 4.1, 6.1, 10.2); $^{13}$C NMR (DMSO) δ 135.4 (C), 125.0 (CH), 71.4 (CH), 71.0 (CH), 70.0 (CH), 63.6 (CH); MS (Cl 70 eV) $m/z$ (rel. intensity) 237 (5), 212
(5), 182 (8), 180 (26), 178 (24), 160 (30), 145 (80), 116 (80), 114(100); **Anal. calcd for C₆H₈N₃O₃Cl: C, 35.05; H, 3.92; Found: C, 35.19; H, 3.87.**

**1R, 2R, 3R - Cyclohexanetriol-4S-amine:** Method #1: To a steel vessel was added azido alcohol (118b) (396mg, 1.612 mmol), CH₃OH (14mL), and PtO₂ (45mg, 0.1982 mmol). The vessel was charged with hydrogen and purged, charged again and maintained at 80 psi. After 6h no starting material remained by TLC, the solution was filtered through Celite and the solvent evaporated to give a light brown oil. To the oil was added aqueous HCl (1 mL, 2.42 N), followed by 4 mL of CH₃OH with stirring (5 min). The solution was evaporated to dryness and CHCl₃ (2 x 15mL) was added and subsequently evaporated. The crude product was then dissolved in CH₃OH (1-2 mL) and the product was precipitated from solution by adding hexane, then ether dropwise. The lightly pink precipitate was precipitated again to give the salt as a white precipitate (255.9 mg, 86%).

Method #2: To a one-piece reflux apparatus was added benzylationmo alcohol (120) (249.0 mg, 0.804 mmol), followed by CH₃OH (4 mL), 10% Pd/C (180 mg) and ammonium formate (250.0 mg, 3.965 mmol). The reaction mixture was brought to reflux and after 10 min. no starting material remained by TLC. The solution was filtered through Celite and evaporated to give a brown oil. The crude oil was worked-up as described in method #1 using 1 mL of 1.21 N HCl, a white precipitate ensued (105.8 mg, 72%). **Rᵣf= 0.0 (ethyl acetate); mp > 200 °C; [α]D²⁴=-26.24 (c 1.0, H₂O); IR (KBr) ν 3410, 3320, 3050, 2940, 1660, 1595, 1515, 1075 cm⁻¹;¹H NMR (DMSO) δ 7.96 (3H, bs), 5.36 (1H, bs), 4.84 (1H, bs), 4.60 (1H, bs), 3.77 (1H, s), 3.52 (1H, t, J 9.8), 3.16 (1H, d, J 8.4), 2.74 (1H, bs), 1.67 (3H, m), 1.41 (1H, m);¹³C NMR (DMSO) δ 74.5 (CH), 70.5 (CH), 68.0 (CH), 53.5 (CH), 27.4 (CH₂), 22.4 (CH₂); MS (CI 70 eV) m/z (rel. intensity) 148 (100), 112 (30), 85 (11), 84(9); **Anal. calcd for C₆H₁₄ClN₂O₃: C, 39.24; H, 7.69; Found: C 39.24; H 7.70**

**General procedure for the formation of lactol 122a and 122b:** A 0.1 M soln of lactol 138 in distilled H₂O had amberlyst 15(wet) ion-exchange resin (4.0 weight eq. based on 138) added. The temp was raised to 65 °C and after 5h the rxn was finished by TLC. The solution containing crude product 140b was passed through a fritted glass filter and the pH adjusted to 7.0 (+ or - 0.5) with sat. Na₂CO₃H. This solution was diluted with H₂O until the molarity was lowered to 0.05 M. NaIO₄ (1.0 eq., based on 11) was added and the rxn flask was totally enclosed with aluminum foil, to exclude all light. Rxn time varies for epimers, 122a needs 0.5
hrs, **122b** needs > 2 hrs. The reaction soln was saturated with NaCl and the product was extracted with EtOAc/isopropanol (1:1) until no more product could be detected in the aqueous layer by TLC. The combined organic extracts had NaCl added to them, were then decanted, dried with MgSO₄, filtered and solvent removed. The crude product was chromatographed using hexanes/EtOAc 1:4, producing **122** as a clear oil.

**(2R)-hydroxy-(3S)-azido-γ-butyrolactol (122b).** This compound was obtained in 64% yield from **138b** following the general procedure. Rf = 0.35 (CH₂Cl₂/acetone, 3:1); [α]D²³ = + 39.58 (c 1.2, acetone); IR (film) ν 3400, 2105, 1660, 1640 cm⁻¹; ¹H NMR (DMSO) δ 6.38 (m, 2H), 5.60 (d, J = 4.39 Hz, 1H), 5.30 (d, J = 6.83 Hz, 1H) 5.06 (t, J = 5.42, 4.52 Hz, 1H), 5.00 (dd, J = 4.6, 1.3 Hz, 1H), 4.02 (m, 3H), 3.80 (m, 2H), 3.70 (m, 1H), 3.40 (m, 2H); ¹³C NMR (DMSO) δ 102.5 (CH), 95.7 (CH'), 80.4 (CH), 75.8 (CH'), 65.3 (CH), 64.7 (CH'), 68.3 (CH₂), 66.7 (CH₂); MS (CI 70 eV) m/z (rel. intensity) 128 (15), 118 (8), 103 (6), 88 (93), 85 (54), 73 (30), 60 (100); **Anal. calcd for C₄H₇N₃O₃:** C, 33.11; H, 4.86; **Found:** C, 33.37; H, 4.98.

**(2S)-hydroxy-(3S)-azido-γ-butyrolactol (122a).** This compound was obtained in 68% yield from **138a** following the general procedure. Rf = 0.29 (CH₂Cl₂/acetone, 3:1); [α]D²⁵ = + 5.47 (c 1.17, acetone); IR (film) ν 3400, 2950, 2900, 2500, 2105, 1725, 1650 cm⁻¹; ¹H NMR (DMSO) δ 6.39 (d, J = 5.2 Hz, -OH), 5.70 (d, J = 5.1 Hz, -OH), 5.00 (dd, J = 5.2, 2.3 Hz, 1H), 3.95 (m, 3H), 3.63 (dd, J = 9.0, 4.0 Hz, 1H); ¹³C NMR (DMSO) δ 101.8 (CH), 77.2 (CH₂), 67.5 (CH₂), 61.1 (CH); MS (CI 70 eV) m/z (rel. intensity) 128 (45), 118 (8), 103 (12), 100 (13), 85 (68), 72 (46), 60 (100); **Anal. calcd for C₄H₇N₃O₃:** C, 33.11; H, 4.86.

**(3S,4R,5R,6S)-4-bromo-1-chloro-5,6-O-isopropylidene-1-cyclohexene-3,5,6-triol (124).** To chlorocyclohexadienediol **113** (5.00g, 34.11 mmol) in acetone (20 mL, HPLC grade) was added 2,2-dimethoxypropane (30 mL) and cat. p-toluenesulfonic acid monohydrate (175 mg). After 20 min. at RT, no more diol **113** remained by TLC. Separatory funnel washing with sat. NaHCO₃ was followed by CH₂Cl₂ extraction of the aqueous layer. The combined organic extracts were dried with MgSO₄, and solvent removed. The crude acetonide product was placed under high vacuum for 15 min. and then had 1,2-dimethoxyethane (140 mL) and distilled
H₂O (35 mL) added to it. This soln. was cooled in an ice/NaCl-bath and then NBS (1.6 eq., based on 113), 54.58 mmole, 9.715 g) was added. The reaction was then maintained at 0 °C for 10 hrs, at which point the rxn was finished by TLC (note this rxn has not been optimized). To the soln. was added brine (100 mL) and EtOAc. The EtOAc extracts were combined and dried with MgSO₄. Removal of volatiles gave 4.20 g of crude product, this residue was dissolved in hot hexanes and decanted to another flask. Upon cooling a white precipitates forms [this is an undesired diepimeric bromohydrin (270 mg, 0.952 mmole, 2.8% yield from diol 113) which is difficult to remove by chromatography, Rf= 0.30 (hexanes/EtOAc, 4:1)], the soln. is once again decanted hexanes removed to give an oily residue. Column chromatography of the residue using acetone/CH₂Cl₂ (1:50) gives the desired bromohydrin 124 (2.968 g, 10.47 mmole, 30.7% from diol 7) as a clear oil. Rf= .35 (hexanes/EtOAc, 4:1); mp = 43-47 °C; [α]D²³ = +17.24 (c 1.16, CHCl₃); IR (neat) ν 3485, 3000, 2940, 1650, 1380, 1220, 1060 cm⁻¹; ¹H NMR (CDCl₃) δ 6.10 (d, J = 4.4 Hz, 1H), 4.61 (dm, J = 2.27 Hz, 2H), 4.35 (dddd, J = 9.1, 4.4, 4.2 Hz, 1H), 4.27 (m, 1H), 2.88 (d, J = 9.1 Hz, 1H), 1.51 (s, 3H), 1.41 (s, 3H); ¹³C NMR (CDCl₃) δ 133.1 (C), 122.2 (C), 126.9 (CH), 78.1 (CH), 75.3 (CH), 70.2 (CH), 48.6 (CH), 27.9 (CH₃), 26.4 (CH₃); MS (Cl, 70 eV) m/z (rel. intensity) 285 (0.25), 233 (M⁺, 0.40), 281 (0.27), 269 (40.0), 267 (38), 211 (20), 209 (65), 207 (50), 181 (23), 145 (23), 128 (21), 59 (100); Anal. calcd for C₉H₁₂BrClO₃: C, 38.12; H, 4.266. Found: C, 38.00; H, 4.26.

(1S,4S,5S,6S)-3-chloro-4,5-O-isopropyldiene-7-oxa-bicyclo[4.1.0]hept-2-ene-4,5-diol (125). To an argon flooded one-piece reflux condenser was added bromohydrin 124 (1.650 g, 5.829 mmol) and CH₂Cl₂ (25 mL), followed by n-tetrabutylammoniumhydrogensulfate (195.2 mg, 0.575 mmol) and NaOH (300.0 mg, 5.749 mmol, note NaOH pellets were crushed in a crucible, weighed and immediately added). This refluxed for 1h and then stirred at rt for another 12h at which point the reaction was finished by TLC. The reaction was quenched with saturated NH₄Cl and the aqueous layer was extracted with CH₂Cl₂ (X2). The combined CH₂Cl₂ extracts were dried with MgSO₄, filtered, and solvent removed. Column chromatography using gradient elusion (hexanes/EtOAc, 9:1 and ending with 7:3) gave epoxide 125 (805.7 mg, 3.98 mmol) as a white solid in 68.3% yield. Recrystallization from hexanes gave an analytical sample. Rf= .46 (hexanes/EtOAc, 2:1); mp= 89-90 °C; [α]D²³ = -111.9 (c 1.05, CHCl₃); IR (neat) ν 3070, 2990, 2910, 2875, 1650, 1375, 1255, 1210, 1060, 1040 cm⁻¹; ¹H NMR (CDCl₃) δ 6.40 (d, J = 4.3 Hz, 1H), 4.59 (dd, J = 6.7, 1.8 Hz, 1H), 4.52 (dd, J = 6.8, 2.6 Hz, 1H), 3.61 (ddd, 4.2, 2.6, 1.7 Hz, 1H), 3.48 (t, J = 4.2 Hz, 1H), 1.52 (s, 3H), 1.40 (s, 3H); ¹³C NMR (CDCl₃) δ 135.7 (C), 109.1 (C), 125.6 (CH), 75.5 (CH), 74.0 (CH), 54.5 (CH), 49.6 (CH), 27.1 (CH₃), 25.3 (CH₃); MS (Cl, 70 eV) m/z (rel. intensity) 203 (M⁺+1, 1.0), 187 (80.0), 145 (60), 117.
(3R,4R,5S,6S)-3-bromo-1-chloro-5,6-O-isopropylidene-1-cyclohexene-4,5,6-triol (128). To an argon flooded one-piece reflux condenser was added syn-epoxyacetonide 125 (5.000 g, 24.67 mmol) and distilled THF (80.0 mL), followed by lithium bromide (2.570 g, 29.60 mmol, 1.2 eq) and ethylacetocetate (6.24 mL, 49.34 mmol, 2.0 eq). After 4 hrs at 30 °C, no epoxide remained by TLC (do not use hexane/EtOAc mixtures to monitor rxn, epoxide and bromohydrin cospost, use acetone/CH₂Cl₂). The rxn soln was washed with saturated NH₄Cl (X3), then dried with MgSO₄, filtered, and volatiles removed, the viscous residue was then left under high vacuum at rt for 48 hrs (to remove most of the ethylacetocetate). Column chromatography using gradient elution (acetone/CH₂Cl₂, 1:99 and ending with 3:97 or hexanes/EtOAc, 9:1 and ending with 7:3) gave bromohydrin 10b (6.535 g, 23.22 mmol) as a white solid in 94.1% yield. Recrystallization from hexanes gave an analytical sample. Rf= 0.40 (hexanes/EtOAc, 2:1); mp= 66-67 °C; [α]D²³ = -89.3 (c 1.00, CHCl₃); IR (KBr) ν 3420, 3070, 2995, 2935, 2905. 1630, 1370, 1220 cm⁻¹; ¹H NMR (CDCl₃) δ 6.06 (d, J = 2.7 Hz, 1H), 4.75 (ddd, J = 7.7, 2.6, 1.8 Hz, 1H), 4.64 (dd, J = 5.4, 1.7 Hz, 1H), 4.60 (dd, J = 5.4, 2.4 Hz, 1H), 4.06 (dd, J = 7.7, 2.4 Hz, 1H), 1.43 (s, 3H), 1.41 (s, 3H); ¹³C NMR (CDCl₃) δ 133.7 (C), 111.2 (C), 126.4 (CH), 76.1 (CH), 75.8 (CH), 72.7 (CH), 48.8 (CH), 27.2 (CH₃), 26.1 (CH₃); MS (Cl, 70 eV) m/z (rel. intensity) 287 (7.5), 285 (22), 283 (M⁺,19), 269 (14), 267 (11), 209 (16), 203 (25), 147 (32.4), 145 (90), 59 (100); Anal. calcd for C₉H₁₂BrClO₃: C, 38.12; H, 4.27. Found: C, 38.29; H, 4.23.

General procedure for the formation of lactols 138a and 138b. A 0.4 M soln of azido alcohol 118 in methanol was cooled in a dry-ice/acetone bath and an excess of O₃/O₂ was bubbled through the solution until a persistent blue color was apparent, indicating the presence of O₃ and the fact that it is no longer being consumed. Nitrogen was then bubbled through the solution for half an hour at -78 °C. The reaction temperature was then raised to -55 °C and 3.0 eq. of NaBH₄ were added slowly so that the rxn temperature never rose above -30 °C (note the reduction is routinely done with the round bottom open to the atmosphere). This solution is allowed to come to rt over a 6h period slowly. If the reduction is not complete by TLC analysis, more NaBH₄ should be added at rt very slowly; use 0.3 eq. of NaBH₄, stir for 0.5 hrs, check by TLC. If not complete repeat this routine until the reduction is complete, note over reduction can occur. The reaction was acidified using aqueous HCl (1.0 M) until pH = 4.5 (+ or - 0.5). Extract with EtOAc (X4), wash combined organics with brine, dry with MgSO₄, remove volatiles. Column chromatography using gradient hexanes/EtOAc (6.5:3.5 ending with 1:1) gives a clear oil in 70 - 80% yield.

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(2S,3S,4R)-2,3-O-Isopropylidene-4-((1'S)-1'-azido-2'-hydroxyethyl)-γ-butyrolactol (138b). Note the equilibrium between the two possible anomic lactols is evident in the $^1$H NMR and $^{13}$C NMR, but one anomic form dominates in an approximate ratio of 9 to 1 (conc. of 10 - 20 mg in 0.5 mL of CDCl$_3$). $R_f$ = .29 (hexanes/EtOAc, 1:1); $[\alpha]_D^{21}$ = + 31.03 (c 0.97, CHCl$_3$); IR (neat) ν 3400, 2995, 2940, 2115, 2095, 1755, 1725 cm$^{-1}$; $^1$H NMR (CDCl$_3$) δ 5.40 (s, 1H), 4.73 (dd, J = 5.9, 3.6 Hz, 1H), 4.61 (d, J = 5.9 Hz, 1H), 4.24 (dd, J = 8.8, 3.4 Hz, 1H), 4.11 (br.d, J = 2.0 Hz, 1H), 3.82 (m, 2H), 3.71 (m, 1H), 2.92 (br.t, J = 5.5 Hz, 1H), 1.44 (s, 3H), 1.29 (s, 3H); $^{13}$C NMR (CDCl$_3$) δ 112.9 (C), 100.9 (CH), 85.9 (CH), 79.9 (CH), 79.5 (CH), 63.5 (CH), 62.1 (CH$_2$), 25.9 (CH$_3$), 24.8 (CH$_3$); MS (Cl, 70 eV) m/z (rel. intensity) 246 (M$^+ + 1$, 0.5), 230 (12), 218 (2), 202 (2.5), 159 (20), 73 (25), 59 (100); HRMS calcd for C$_{13}$H$_{22}$N$_{2}$O$_3$: Found: ....

(2S,3S,4S)-2,3-O-Isopropylidene-4-((1'S)-1'-azido-2'-hydroxyethyl)-γ-butyrolactol. (138a). Note the equilibrium between the two possible anomic lactols is evident in the $^1$H NMR and $^{13}$C NMR. Also the number of protons listed for the splitting patterns of the $^1$H NMR will not always be integers because the pair of anomic lactols are present in an approximate ratio of 2 to 1 (when approximately 20 mg of compound are in 0.5 mL of CDCl$_3$). If the conc. is decreased (approximately 3 mg of compound in 0.5 mL of CDCl$_3$) the anomic ratio changes to approximately 3 to 1. $R_f$ = .26 (hexanes/EtOAc, 1:1); $[\alpha]_D^{23}$ = + 11.7 (c 0.94, CHCl$_3$); IR (neat) ν 3430, 2995, 2950, 2100, cm$^{-1}$; $^1$H NMR (CDCl$_3$) δ 5.44 (s, 1H), 5.38 (d, J = 3.83 Hz, 0.5H), 4.83 (dd, J = 6.0, 1.1 Hz, 1H), 4.72 (dd, J = 6.8, 2.8 Hz, 0.5H), 4.67 (dd, J = 6.9, 3.9 Hz, 0.5H), 4.61 (d, J = 5.92 Hz, 1H), 4.1 (dd, J = 5.5, 2.8 Hz, 0.5H), 4.06 (dd, J = 9.36, 1.22 Hz, 1H), 3.97 (dd, J = 11.8, 4.0 Hz, 1H), 3.81 (m, 2.5 H), 3.62 (m, 1.5H), 1.55 (s, 3.15H), 1.46 (s, 3H), 1.38 (s, 1.5H), 1.31 (s, 3H); $^{13}$C NMR (CDCl$_3$) δ 115.2 (C), 112.9 (C), 103.2 (CH), 96.5 (CH), 86.0 (CH), 85.7 (CH), 82.3 (CH), 80.9 (CH), 80.7 (CH), 79.5 (CH), 64.6 (CH), 64.2 (CH), 62.9 (CH$_2$), 62.4 (CH$_2$) 26.5 (CH$_3$), 26.2 (CH$_3$), 24.9 (CH$_3$); MS (Cl, 70 eV) m/z (rel. intensity) 246 (M$^+ + 1$, 4), 218 (23), 202 (25), 200 (25), 188 (41), 159 (64), 142 (61), 69 (69), 59 (100).

General procedure for the formation of 148a,b and 149a,b. To a flame dried round bottom under Ar was added n-tetradecltriphenylphosphonium bromide (3.20 eq.). After 5 min. under vacuum the RB was flooded with Ar. THF was added until a 1.0 M soln was obtained. This soln was cooled with an ice bath and n-BuLi (2.75 eq., 2.0 M in hexanes) was added giving a brownish/yellow color. After stirring for 5 min the ice-bath was removed and the soln was allowed to come to rt. After stirring at rt for 15 min the lactol (1.0 eq.) in THF (0.7 M) was added. The round bottom originally containing the lactol was rinsed with additional THF and this was immediately transferred to the reaction flask. The rxn continued to stir at rt and was monitored by
TLC. TLC's at 1, 2, 3, and 5 hrs revealed no apparent change in product or starting material (a small amount remains) intensity, thus after 5 hrs the rxn was quenched with saturated NH_4Cl soln. Extraction with EtOAc (X3), combination of the organic extracts, drying with MgSO_4, filtration, and removal of the volatile solvents gave a viscous residue. Gradient elution using Hexane/EtOAc (beginning with 15% EtOAc and ending with 30% EtOAc) gave cis and trans azidosphingosines in approximately a 4 to 1 ratio respectively, by ^1H NMR.

(2S,3S,4E)-2-azido-4-octadecen-1,3-diol (149b). A 6.1% yield of the trans product was observed. **R_f** = 0.45 (hexane/EtOAc, 2:1); [α]_D^24 = +3.11 (c 0.53, CHCl_3); **IR** (neat) ν 3360, 2920, 2855, 2095, 1520 cm\(^{-1}\); **1H NMR** (CDCl_3) δ 5.78 (dd, J = 15.4, 6.7, 0.7 Hz, 1H), 5.50 (dd, J = 15.4, 7.1, 1.4 Hz, 1H), 4.19 (t, J = 6.5 Hz, 1H), 3.80 (dd, J = 11.5, 4.3 Hz, 1H), 3.68 (dd, J = 11.5, 6.3 Hz, 1H), 3.45 (dt, J = 6.0, 4.3 Hz, 1H), 2.04 (q, J = 6.7 Hz, 2H), 1.63 (br.s, >2H), 1.22 (m, >2H), 0.86 (t, J = 6.6 Hz, 3H); **13C NMR** (CDCl_3) δ 135.5 (CH), 128.4 (CH), 73.6 (CH), 67.8 (CH), 63.0 (CH_2), 32.3 (CH_2), 31.9 (CH_2), 29.7 (CH_2), 29.5 (CH_2), 29.3 (CH_2), 29.2 (CH_2), 29.0 (CH_2), 22.7 (CH_2), 14.0 (CH_3); **MS** ; Anal. calcd for C_{18}H_{35}N_3O_2: C, 66.42; H, 10.84; N, 12.91; **Found**: C, 66.87; H, 10.70; N, 12.41.

(2S,3S,4Z)-2-azido-4-octadecen-1,3-diol (148b). The cis isomer was produced in 24.4% yield. **R_f** = 0.52 (hexane/EtOAc, 2:1); **1H NMR** (CDCl_3) δ 5.64 (dt, J = 11.1, 7.5 Hz, 1H), 5.44 (tt, J = 9.9, 1.5 Hz, 1H), 4.53 (m, 1H), 3.79 (m, 1H), 3.65 (m, 1H), 3.44 (dt, J = 6.3, 4.1 Hz, 1H), 2.06 (m, 4H), 1.23 (m, >2H), 0.86 (t, J = 6.6 Hz, 3H); **13C NMR** (CDCl_3) δ 135.7 (CH), 127.7 (CH), 68.4 (CH), 68.1 (CH), 62.8 (CH_2), 31.9 (CH_2), 29.64 (CH_2), 29.56 (CH_2), 29.5 (CH_2), 29.3 (CH_2), 28.0 (CH_2), 22.7 (CH_2), 14.0 (CH_3);

(2S,3R,4E)-2-azido-4-octadecen-1,3-diol (149a). A 3.8% yield of the trans product was observed. **R_f** = 0.38 (hexane/EtOAc, 2:1); [α]_D^24 = -34.1 (c 1.58, CHCl_3); **1H NMR** (CDCl_3) δ 5.80 (ddt, J = 15.4, 6.7, 0.7 Hz, 1H), 5.51 (ddt, J = 15.4, 7.3, 1.3 Hz, 1H), 4.23 (t, J = 6.4 Hz, 1H), 3.77 (m, 2H), 3.45 (q, J = 5.3 Hz, 1H), 2.05 (q, J = 7.0 Hz, 2H), 1.94 (br.s, 2H), 1.23 (m, >2H), 0.86 (t, J = 6.8 Hz, 3H); **13C NMR** (CDCl_3) δ

(2S,3R,4Z)-2-Azido-4-octadecen-1,3-diol (148a). A 14.0% yield of the cis isomer was observed. **R_f** = 0.43 (hexanes/EtOAc, 2:1); **1H NMR** (CDCl_3) δ 5.67 (ddt, J = 11.0, 7.5, 0.8 Hz, 1H), 5.45 (tt, J = 10.9, 1.5 Hz, 1H), 4.58 (dddt, J = 8.8, 5.8, 0.9 Hz, 1H), 3.77 (m, 2H), 3.49 (q, J = 5.4 Hz, 1H), 2.19 (br.s, 2H), 2.08 (m, 2H), 1.23 (m, >2H), 0.85 (t, J = 0.85, 3H)

(3R,4S,5S,6S)-3-Azido-4-tbutylidiphenylsilyl-1-chloro-5,6-O-isopropylidenecyclohex-1-ene-4,5,6-triol (150d). To a one-piece reflux apparatus was added azido alcohol 118d (2.302 g, 9.37 mmol) as a solution in THF (10.0 mL), an additional 4.0 mL of THF was used to rinse the solution rb and this was placed in the reflux apparatus. tButylidiphenylsilylchloride (5.074 g, 18.46 mmol, 1.97 equiv) and imidazole (1.524 g, 22.37
mmol, 2.387 equiv.) were then added. The apparatus was then placed in a 75 °C oil bath with stirring. After 4 h of heating the reaction was allowed to cool to rt, and then stirred for another 12 h. Note: as the reaction proceeds a white precipitate accumulates. Work-up: The solution was filtered through celite and the reaction rb and filter are rinsed with methylene chloride, the filtrate is washed with sat. ammonium chloride, and then with brine. The combined organic layers were then dried with MgSO₄, filtered, and evaporated to give 7.214 g of a viscous brown oil. Purification by flash chromatography using gradient elution (100% hexanes → 3% ethylacetate in hexanes) provided 3.846 g (7.945 mmol, 85%) of the silyl ether as a very viscous clear oil. \( R_f = 0.45 \) (hexanes/EtOAc, 9:1); \([\alpha]_D^{24} = -62.5 \) (c 0.95, CHCl₃); \( \text{IR (neat)} \) ν 3075, 3050, 2985, 2930, 2895, 2860, 2105, 1430, 1380, 1225 cm⁻¹; \( ^1H \text{ NMR (CDCl}_3) \) δ 7.78 (m, 4H), 7.40 (m, 6H), 5.73 (m, J = 1.7 Hz, 1H), 4.37 (d, J= 8.7 Hz, 1H), 4.10 (m, 2H), 3.68 (dt, J = 8.8 Hz, 1H), 1.42 (s, 3H), 1.27 (s, 3H), 1.09 (s, 9H); \( ^13C \text{ NMR (CCl}_3) \) δ 133.9 (C), 133.8 (C), 132.2 (C), 110.6 (C), 19.4 (C), 136.0 (CH), 135.9 (CH), 130.2 (CH), 129.9 (CH), 128.0 (CH), 127.6 (CH), 76.6 (CH), 76.4 (CH), 72.8 (CH), 61.7 (CH), 27.5 (CH₃), 26.8 (CH₃), 26.2 (CH₃); \( \text{MS Cl m/z} \) (rel. intensity) 484 (M⁺, 0.52), 426 (17), 400 (15), 399 (11), 398 (38), 386 (10), 385 (36), 384 (27), 383 (100), 378 (34), 368 (29), 348 (21), 340 (24), 320 (27), 305 (37); \( \text{Anal. calcd for C}_25\text{H}_{30}\text{ClN}_3\text{O}_3\text{Si} \): C, 62.03; H, 6.25; N, 8.68. \( \text{Found: } \) C, 62.41; H, 6.52, 8.35.

\( (2S,3R,4S,5R)-5-\text{Azido-4-} \text{tbutyldiphenylsilyloxy-1,2-O-isopropyldenehexanoicmethylester-2,3,4-triol (151d)} \). To a rb was added vinyl chloride (150d) (3.740 g, 7.73 mmol) followed by MeOH (51 mL). This solution was heated gently to completely dissolve the vinyl chloride. The solution was then cooled in a dry ice/acetone bath and ozone was bubbled through the solution. After 20 min the solution was saturated with O₃, as indicated by the characteristic blue color. TLC indicated no starting material remained. The generation of ozone was terminated and the reaction rb had Ar bubbled through it for 20 min at -78 °C. The reaction flask was then placed in an ice-bath. With the rb open to the atmosphere several portions of NaBH₄ (Note: use lumps of NaBH₄ vs powder when possible. If powder is used, add slowly \( i.e. \) over \( \equiv 3 - 5 \) min) were added. 580 mg (15.33 mmol, 1.98 equiv) of NaBH₄ were added first, \( \equiv 20 - 30 \) min transpired and then more NaBH₄ (220 mg) was added. 25 min passed and another portion of NaBH₄ (240 mg) was added, an additional 25 min transpired. At this point the ice-bath was removed, and another 20 min passed and then two more portions of NaBH₄ (340 mg, wait 20 min, then 107 mg) were added. Note: it is crucial to monitor this reaction by TLC after each addition of NaBH₄; depending on the reaction more or less NaBH₄ may be needed. Work-up: Add distilled H₂O (150 mL) and then acidify with 1.2 N HCl acid, to a pH of 3.5 ± 0.5. Immediately extract the aqueous solution with EtOAc (X4), wash the combined EtOAc
extracts with brine (X2), dry with MgSO₄, filter, and evaporate the EtOAc. This procedure provided an oil which was purified by flash chromatography, using gradient elution (15% EtOAc in hexanes → 40% EtOAc in hexanes), and provided 3.592 g (6.99 mmol, 90%) of the methyl ester 151d as a viscous clear oil. Rᵢ = 0.42 (hexanes/EtOAc, 2:1); [α]D²³⁺ = +29.6° (c 1.56, CHCl₃); IR (neat) ν 3500, 3070, 3050, 2980, 2950, 2930, 2890, 2850, 2100, 1755, 1590 cm⁻¹; ¹H NMR (CDCl₃) δ 7.72 (m, 4H), 7.41 (m, 6H), 4.40 (ddm, J= 3.9, 6.9 Hz, 1H), 4.22 (m, 2H), 3.52 (m, 6H), 1.57 (s, 3H), 1.31 (s, 3H), 1.05 (s, 9H); ¹³C NMR (CCl₃D) δ 170.3 (C), 133.3 (C), 133.0 (C), 110.6 (C), 19.6 (C), 136.0 (CH), 135.9 (CH), 130.0 (CH), 127.8 (CH), 127.7 (CH), 80.1 (CH), 75.4 (CH), 70.4 (CH), 64.6 (CH), 51.8 (CH), 62.3 (CH₂), 27.0 (CH₃), 26.4 (CH₃), 25.1 (CH₃); MS Cl m/z (rel. intensity) 514 (M⁺+1, 2), 487 (33), 486 (100), 429 (17), 428 (73.6), 408 (60), 379 (12), 378 (73), 358 (31), 350 (25), 300 (22), 291 (45), 240 (25), 220 (49), 199 (21), 144 (21), 135 (21), 59 (25); Found: C, 61.16; H, 7.15; N, 8.18. Found: C, 61.16; H, 7.15; N, 8.18.  

(2S,3R,4S,5R)-5-Azido-4-butyldiphenylsilyloxy-hexanoicmethylester-2,3,4-triol (152d). To a 30 mL of 1% I₂ in MeOH (i.e. 1.0 g of I₂ per 100 mL of MeOH). This solution was placed in an oil bath at 45 °C with stirring. After 46 h no more acetonide remained by TLC and the reaction was quenched with 80 mL of saturated sodium thiosulfate (Na₂S₂O₃). This solution was extracted with EtOAc (X3), the organic extracts were combined, dried with MgSO₄, filtered, and the volatile organics removed under vacuum. This procedure provided the crude product as a viscous oil which was purified by gradient elution (25% EtOAc in Hexanes → 65% EtOAc in hexanes) chromatography. (Note: Four inches of silica gel are sufficient and necessary. This product is prone to silyl migration and lactonization on prolonged exposure to silica gel.) This provided 1.278 g (2.699 mmol, 80% yield) of the desired vicinal diol (152d). Note: If the reaction is performed at higher temperatures two known products (lactones 154d and 155d) appear. The physical data for the lactones follows this experimental. Rᵢ = 0.19 (hexanes/EtOAc, 1:1); [α]D²³⁺ = +13.4° (c 1.00, CHCl₃); mp = 78 - 82 °C; IR (KBr) ν 3470, 3430, 3350, 3075, 3050, 3020, 2900, 2930, 2890, 2855, 2095, 1755, 1590, 1425, 1100, 1080, 810 cm⁻¹; ¹H NMR (DMSO-d₆) δ 7.69 (m, 4H), 7.42 (m, 6H), 5.62 (d, J = 5.9 Hz, -OH), 5.31 (d, J = 5.8 Hz, -OH), 4.91 (t, J = 5.2 Hz, -OH), 4.27 (t, J = 6.1 Hz, 1H), 3.83 (t, J = 4.3 Hz, 1H), 3.76 (q, J = 5.4 Hz, 1H), 3.66 (q, J = 5.7 Hz, 1H), 3.52 (s, 3H), 3.37 (t, J = 5.6 Hz, 2H), 0.98 (s, 9H); ¹³C NMR (CCl₃D) δ 172.9 (C), 132.8 (C), 19.5 (C), 136.0 (CH), 130.2 (CH), 127.9 (CH), 75.4 (CH), 71.8 (CH), 71.6 (CH), 64.5 (CH), 52.5 (CH), 61.4 (CH₂), 27.0 (CH₃); MS Cl m/z (rel. intensity) 514 (M⁺+1, 2); Anal. calcd for C₂₃H₃₁N₃O₆Si: C, 58.33; H, 6.59; N, 8.87; Found: C, 64.47; H, 7.49; N 9.30.
(2S,3S,4S)-2-hydroxy-3-(tert-Butyldiphenylsilyl)oxy-4-((1R')-1'-azido-2'-hydroxyethyl)-γ-butyrolactone (154d). The procedure used to make (2S,3R,4S,5R)-5-Azido-4-butyldiphenylsiloxy-hexanoicmethylster-2,3,4-triol (152d) also provided this lactone. \( \text{RF} = 0.45 \) (hexanes/ EtOAc, 1:1); \([\alpha]_p^{24} = -45.3^\circ \) (c 1.03, CHCl₃); mp = 104-105 °C; IR (KBr) ν 3350, 3070, 3045, 2940, 2890, 2850, 2105, 1790, 1775, 700 cm\(^{-1}\); \(^1\)H NMR (CDCl₃) δ 7.66 (m, 4H), 7.46 (m, 6H), 4.57 (m, 1H, note with a drop of D₂O present the multiplet is simplified to a d, J = 6.0 Hz, 1H), 4.34 (d, J = 5.9 Hz, 1H), 3.99 (d, J = 2.1 Hz, 1H), 3.61 (dd, J = 7.7, 11.2 Hz, 1H), 3.44 (dd, 5.4, 11.2 Hz, 1H), 2.92 (br s, 1H, -OH), 2.69 (m, 1H, note with a drop of D₂O present the multiplet is simplified to a heptet, J = 2.2, 5.4, 7.6 Hz, 1H), 1.64 (br s, 1H, -OH), 1.09 (s, 9H), (Note: If the \(^1\)H NMR solvent is DMSO-d₆ or acetone-d₆ the lactone (155d) immediately appears.); \(^{13}\)C NMR (CCl₃D) δ 174.8 (C), 132.5 (C), 131.4 (C), 19.2 (C), 135.7 (CH), 130.8 (CH), 130.6 (CH), 128.3 (CH), 83.2 (CH), 71.7 (CH), 68.2 (CH), 62.15 (CH), 62.24 (CH₂), 26.9 (CH₃); MS Cl m/z (rel. intensity) 514 (M⁺+1, 2); Anal. calcd for C₂₂H₂₇N₃O₅Si: C, 59.84; H, 6.16; N, 9.52. Found: C, 59.46; H, 6.15; N 9.34.

(2S,3S,4S)-2-(tert-Butyldiphenylsilyl)oxy-3-hydroxy-4-((1R')-1'-azido-2'-hydroxyethyl)-γ-butyrolactone(155d) second lactone. The procedure used to make (2S,3R,4S,5R)-5-Azido-4-butyldiphenylsiloxy-hexanoicmethylster-2,3,4-triol (152d) also provided this lactone. \( \text{RF} = 0.28 \) (hexanes/ EtOAc, 1:1); \([\alpha]_p^{24} = -77.2^\circ \) (c 1.00, CHCl₃); mp = 121-123 °C; IR (KBr) ν 3350, 3070, 3045, 2960, 2935, 2890, 2860, 2105, 1785, 700 cm\(^{-1}\); \(^1\)H NMR (CDCl₃) δ 7.82 (m, 2H), 7.69 (m, 2H), 7.47 (m, 6H), 4.65 (d, J = 5.5 Hz, 1H), 4.44 (d, J = 2.4 Hz, 1H), 3.81 (d, J = 6.4 Hz, 1H), 3.78 (dd, J = 0.8, 5.5 Hz, 1H), 3.61 (ddd, J = 2.5, 6.4 Hz, 1H), 2.98 (br s, -OH), 2.05 (br s, -OH), 1.16 (s, 9H), (Note: If the \(^1\)H NMR solvent is DMSO-d₆ the lactone (154d) immediately appears.); \(^{13}\)C NMR (CCl₃D) δ 173.3 (C), 132.2 (C), 131.0 (C), 19.3 (C), 135.9 (CH), 135.5 (CH), 130.6 (CH), 130.5 (CH), 128.1 (CH), 128.0 (CH), 82.1 (CH), 70.2 (CH), 69.5 (CH), 62.3 (CH), 62.8 (CH₂), 26.8 (CH₃); MS Cl m/z (rel. intensity) 442 (M⁺+1, 2.5), 385 (11), 384 (43), 365 (22), 364 (100), 328 (26), 310 (11), 309 (18), 308 (83), 290 (26), 263 (17), 249 (20), 222 (17), 221 (24), 202 (25), 200 (21), 199 (34), 163 (17), 60 (52); Anal. calcd for C₂₂H₂₇N₃O₅Si: C, 59.84; H, 6.16; N, 9.52. Found: C, 59.83; H, 6.23; N 9.42.

Series III compound leading to \( \delta \)-three-sphingosine

(3R)-Azido-(2S)-tert-butyldiphenylsiloxy-γ-butyrolactol (156d). To a rb flask was added vicinal diol (152d) (1.278 g, 2.699 mmol), 80 mL of a 3:1 mixture of MeOH/H₂O, and NaN₃O₄ (1.154 g, 5.397 mmol, 2.00 equiv). The reaction was excluded from light and stirred. The
starting material was consumed after 3 h. Note: A white precipitate accumulates as the reaction proceeds, it is not the product. Work-up: CH$_2$Cl$_2$ (80 mL) was added to the reaction and the resulting solution was passed through a plug of silica gel over celite. To the filtrate was added H$_2$O (80 mL). This solution was placed in a separatory funnel and the organic layer removed. The aqueous layer was extracted with CH$_2$Cl$_2$ (X3), the organic extracts were combined, dried with MgSO$_4$, filtered, and volatiles removed to provide an oil. Gradient elution (25% EtOAc in hexanes → 50% EtOAc/hexanes) chromatography provided 778.1 mg (2.029 mmol, 75%) of lactol (156d) as a viscous clear oil. In addition 134.4 mg (0.350 mmol, 13%) of the silyl migrated product (157d) was isolated. Note the two possible anomeric lactols are evident in the $^1$H NMR and $^{13}$C NMR, complicating the spectrum. The two anomers exist in an approximate ratio of 1 to 2 (conc. of 10 - 20 mg in 0.5 mL of CDCl$_3$). H and H' are used to indicate the separate lactol resonances, as established by TOCSY NMR. $R_f$= 0.28 (hexanes/EtOAc, 4:1); $\alpha_{D}^{25} = -2.22^\circ$ (c 1.35, CHCl$_3$); IR (neat) $\nu$ 3435, 3080, 3055, 2980, 2950, 2895, 2860, 2100, 1590, 1425, 1110, 695 cm$^{-1}$; $^1$H NMR (CDCl$_3$) $\delta$ 7.66 (m, 4H), 7.45 (m, 6H), 5.28 (dd, J = 4.0, 9.8 Hz, 1H'), 5.25 (d, J = 7.9 Hz, 1H), 4.28 (dd, J = 5.2, 9.8 Hz, 1H), 4.18 (dd, J = 4.6, 9.8 Hz, 1H', d, J = 1.5 Hz, 1H), 4.12 (dd, J = 2.2, 4.0 Hz, 1H'), 4.01 (dd, J = 2.3, 9.8 Hz, 1H), 3.96 (d, J = 9.8 Hz, -OH'), 3.76 (dt, J = 1.7, 5.0 Hz, 1H), 3.66 (dd, J = 0.5, 2.1, 9.8 Hz, 1H'), 3.62 (p, J = 2.1, 4.6 Hz, 1H'), 2.63 (d, J = 7.9 Hz, -OH), 1.13 (s, 9H), 1.09 (s, 9H); $^{13}$C NMR (CCl$_3$D) $\delta$ 132.7 (C), 132.4 (C), 132.0 (C), 131.4 (C), 19.1 (C), 19.0 (C), 135.6 (CH), 135.58 (CH), 130.6 (CH), 130.5 (CH), 130.24 (CH), 130.19 (CH), 128.2 (CH), 128.1 (CH), 128.0 (CH), 127.9 (CH), 103.5 (CH), 97.0 (CH), 81.1 (CH), 76.5 (CH), 66.2 (CH), 65.9 (CH), 71.2 (CH$_2$), 68.2 (CH$_2$), 26.9 (CH$_3$), 26.8 (CH$_3$).

**(3R)-Azido-1-tert-butyldiphenylsilyloxy-(2S)-hydroxy-γ-butyrolactol (157d).** The procedure used to make **(3R)-Azido-(2S)-2-tert-butyldiphenylsilyloxy-γ-butyrolactol (156d)** also provided 134.4 mg (0.350 mmol, 13%) of this anomeric silyloxy protected lactol (157d). $R_f$= 0.45 (hexanes/EtOAc, 4:1); $\alpha_{D}^{25} = -86.2^\circ$ (c 1.30, CHCl$_3$); IR (neat) $\nu$ 3505, 3070, 3050, 2955, 2930, 2895, 2855, 2100, 1590, 1425, 1110, 695 cm$^{-1}$; $^1$H NMR (CDCl$_3$) $\delta$ 7.67 (m, 4H), 7.43 (m, 6H), 5.41 (d, J = 3.7 Hz, 1H), 4.20 (dd, J = 5.7, 9.7 Hz, 1H), 4.13 (m, 2H), 3.66 (dd, J = 4.4, 9.0 Hz, 1H), 2.91 (d, J = 7.5 Hz, -OH), 1.12 (s, 9H); $^{13}$C NMR (CCl$_3$D) $\delta$ 132.5 (C), 132.4 (C), 19.2 (C), 135.6 (CH), 135.5 (CH), 130.2 (CH), 130.1 (CH), 127.9 (CH), 127.8 (CH), 97.2 (CH), 77.2 (CH), 66.1 (CH), 68.6 (CH$_2$), 26.8 (CH$_3$);
Series III compound leading to d-threeo-sphingosine

(2R,3R,4E)-2-Azido-4-octadecen-1,3-diol (148d/149d). To a flame dried 2b flask under Ar was added n-tetradecyltriphenylphosphonium bromide (358.8 mg, 0.665 mmol, 3.50 equiv). The phosphonium salt was put under vacuum (0.075 ± 0.025 mm Hg) for 1 h and subsequently flooded with Ar. THF (2.50 mL) was added* and the solution was placed in an ice-bath. After stirring for 15 min, sodium amylate† (0.27 M, 2.5 mL, 3.5 equiv) was added§ dropwise over 15 sec. The solution immediately became an orange color. This solution stirred for 15 min and then the ice-bath was removed. After 30 min at rt the ylide solution was cooled to 0 °C again. The lactol (72.9 mg, 0.190 mmol, 1.0 equiv) in THF (1.5 mL) was added dropwise over 1 min. After 0.5 h the ice-bath was removed. After 1.5 h†† of total reaction time the reaction was cooled to -55 °C and then quenched with saturated NH₄Cl (10 mL). This solution was extracted with EtOAc (X3), the organic extracts were combined, dried with MgSO₄, filtered, and the volatile organics removed to yield the crude product as a viscous dark oil. Gradient elution chromatography (100% hexanes → hexanes/EtOAc, 7:3) provided 32 mg of impure sphingosine adduct (##). Further attempts at purification were futile. This crude product had THF (2.0 mL) added to it followed by n-BuNF (40 mg). After 5 min the reaction was filtered and the THF evaporated to provide an oil. Column chromatography of this oil, using gradient elution (100% hexanes → hexanes/EtOAc, 7:3), provided 8.3 mg (0.025 mmol, 13% yield) of azidosphingosine (##).

* Note: The phosphonium salt completely dissolves in the THF, but the solution is not transparent.
§ You must heat the sodium amylate to ±65-70 °C, then remove it via syringe, and add it quickly to the cooled solution of the phosphonium salt. If the syringe cools to rt the amylate will crash out of solution and jamm the syringe.
†† Note: After 15 min (45 min total reaction time) at rt the solution turned a non translucent brown color.

(2R,3R,4Z & 4E)-2-Azido-3-tert-butyldiphenylsilyloxy-4-octadecen-1,3-diol (159d). The ¹H NMR of the impure silyloxy azidosphingosine indicates a 9:1 cis/trans ratio of geometric isomers. Note: When the reaction is run at rt a 6:4 cis/trans ratio is observed. One proton will be defined as the sum of equivalent cis and trans proton integration areas. The major impurity is tert-butyldiphenylsilylalol. ¹H NMR (CDCl₃) δ 7.67 (m, 4H), 7.42 (m, 6H), 5.72 (m, 0.1H, trans), 5.55 (dt, J= 7.3, 11.0 Hz, 0.9H, cis), 5.38 (m, 1H), 4.43 (pentet, J= 4.8 ave., 0.9H, cis), 4.13 (q, J= 5.6 Hz, 0.1H, trans), 3.8 (m, 4H), 3.42 (m, 2H), 1.99 (m, 2H), 1.26 (s, 22H), 0.89 (t, J= 6.9 Hz, 3H).
(2R,3R,4Z)-2-Azido-4-octadecen-1,3-diol (148d). The cis isomer was produced in 12.0% yield.  

$R_f=0$; $^1H$ NMR (CDCl$_3$) $\delta$ 5.66 (dm, $J=7.7$, 11.0 Hz, 1H), 5.46 (ddt, $J=1.5$, 10.8, 10.8 Hz, 1H), 4.52 (m, 1H), 3.81 (m, 1H), 3.67 (m, 1H), 3.46 (ddd, $J=4.1$, 6.4, 6.4 Hz, 1H), 2.08 (m, 2H), 1.90 (m, 2H), 1.24 (s, >20H), 0.90 (t, $J=7.0$ Hz, 1H).
VI. SELECTED SPECTRA

On the following pages selected spectra are provided.
XL:120100
AUX:000
AUX:000
DATE 9-3-93
SP 61.925
SY 67.9330000
SY 3300000
SY 16384
SY 2192
SW 47441.379
H/P: 2.105
PM 0.0
PR 0.0
AB 0.238
AG 200
NS 1220
TF 297
FW 21500
CZ 4416.000
UP 90.00
LB 0.000
BB 0.000
CX 25.000
CY 0.0
FL 240.0120
F2 9.9750
HZ/CM 679.215
PPM/CM 9.999
SN 4992.51
AU PROG:
EZAPT.AU
DATE 14-8-92
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SY 67.9300000
C1  3300.000
S1  16384
TD  8192
SW 17241.379
HZ/PT 2.105
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AQ 0.0
RG 0.238
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TE 297
FW 21600
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DP 5H 00
LB 1.000
GB 0.0
CZ 25.00
CY 0.0
F1 240.012P
F2 -9.975P
HZ/CM 673.215
PPM/CM 9.999
SR -4988.30

**138b**
TN-III-86-DETERMINATION OF CIS/TRANS AZIDOSPHINGOSINE RATIO

KEM.001
DATE 26-4-84

SF 670.133
SY 120.1380.000
01 4415.000
Si 16384
TD 16384
SW 2994.012
HZ/PT .365

PM 4.9
RD 0.0
AQ 2.736
RS 20
NS 16
TE 297

FW 3805
D2 4415.000
DP 63L PO

LB 0.0
GB 0.0
CX 25.00

F1 0.09PM
F2 -0.498PM
H2/CH 91.823
PPM/DM 340
SR 3065.11

D-erythro-azidosphingosine

148a
A 1:1 mixture of lactones 154d and 155d results when either is placed DMSO-d$_6$. 

\[
\text{154d} \quad \text{155d}
\]
TN-111-149-SPOT ABOVE PRODUCT

[Chemical structure diagram]

N3

OTBDPS

OH

155d

[Chemical spectrum graph]
TH-III-170-allyl migrated product

PERIODIC 399.985 MHz
SPECTRAL WIDTH 5000.0 Hz
ACQUISITION TIME 3.344 sec
RELAXATION DELAY 1.000 sec
PULSE WIDTH 3.0 usec
AMBIENT TEMPERATURE
NO. REPSITIONS 32
DUAL PERIOD ACQUISITION
DATA PROCESSING
FT SIZE 65536
TOTAL ACQUISITION TIME 2 minutes
May 6 95
Virginia Tech NMR Facility
NMR OBSERVE
PULSE SEQUENCE #1
REGIME C1S
FREQUENCY 100.578 MHz
SPECTRAL WIDTH 20000.0 Hz
ACQUISITION TIME 300 sec
RELAXATION DEP 1.000 sec
PULSE WIDTH 7.3 usec
FIRST PHASE WIDTH 0.0 usec
AMBIENT TEMPERATURE
NO. REPETITIONS 25
DECOUPLED H1
HIGH POWER AN
DECOUPLER GATED ON DURING ACQUISITION
DOUBLE PRECISION ACQUISITION
DATA PROCESSING
LINE BROADENING 1.0 Hz
FT SIZE 5000
TOTAL ACQUISITION TIME 8 minutes
May 24
Virginia Tech NMR Facility

OTBDPS

HO
N3
CH3

159d
VI. APPENDIX

Table 1. Biological Activity Chart

<table>
<thead>
<tr>
<th>Activity</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Tumor antigens</strong></td>
</tr>
<tr>
<td>GM₂ expression on human melanoma cells</td>
</tr>
<tr>
<td>GD₂ expression upon progression of melanoma</td>
</tr>
<tr>
<td>Antibodies to GD₃ have antitumor activity</td>
</tr>
<tr>
<td>TerC glycolipid marker for teratocarcinoma</td>
</tr>
<tr>
<td>Lactosylceramide marker for colon carcinoma</td>
</tr>
<tr>
<td>A sialylated ganglioside as marker for chronic myelogenous leukemia</td>
</tr>
<tr>
<td>Glycolipid alteration in ras-transfected NIH-3T3 cells</td>
</tr>
<tr>
<td>GD₁₈ expressed by rat hepatoma cells</td>
</tr>
<tr>
<td><strong>Markers of cell differentiation</strong></td>
</tr>
<tr>
<td>Increased sphingomyelin upon differentiation of hairy cell leukemia</td>
</tr>
<tr>
<td>with phorbol esters</td>
</tr>
<tr>
<td>Increased sphingomyelin in 3T3-L1 cells and in polymorphonuclear</td>
</tr>
<tr>
<td>leukocytes treated with dexamethasone</td>
</tr>
<tr>
<td>GM₃ elevation in differentiated HL-60 cells and U937 cells; turnover</td>
</tr>
<tr>
<td>of GM₃ sialic acid residues during fibroblast growth</td>
</tr>
<tr>
<td>SSEA-1 glycolipid in embryo development</td>
</tr>
<tr>
<td>GM₁ and GD₁₈ elevation on differentiation of teratocarcinoma</td>
</tr>
<tr>
<td>GM₃ elevation in differentiating intestinal epithelium</td>
</tr>
<tr>
<td>I antigen in erythrocyte differentiation</td>
</tr>
<tr>
<td>GD₃ elevation in muscle cell differentiation</td>
</tr>
<tr>
<td>Ganglioside elevation in neuroblastoma</td>
</tr>
<tr>
<td>GM₁ elevation in lymphoid cell differentiation</td>
</tr>
<tr>
<td><strong>Role in membrane fluidity</strong></td>
</tr>
<tr>
<td>Gangliosides on outer leaflet of bilayer confer rigidity</td>
</tr>
<tr>
<td>Correlation of sphingomyelin content with fluidity</td>
</tr>
<tr>
<td><strong>Modulation of cell proliferation</strong></td>
</tr>
<tr>
<td>Nerve growth factor-like activity of GQ₁₈</td>
</tr>
<tr>
<td>Neuritogenic and neuronutrophic activities of gangliosides</td>
</tr>
<tr>
<td>GM₁ inhibition of Swiss 3T3 cell growth</td>
</tr>
</tbody>
</table>

143
<table>
<thead>
<tr>
<th>GM&lt;sub&gt;3&lt;/sub&gt; inhibition of growth factor-induced mitogenesis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inhibition of lymphocyte proliferation</td>
</tr>
<tr>
<td>Stimulation of astroglial and neuroblastoma proliferation by</td>
</tr>
<tr>
<td>exogenous gangliosides</td>
</tr>
</tbody>
</table>

**Modulation of protein phosphorylation**

<table>
<thead>
<tr>
<th>GQ&lt;sub&gt;1b&lt;/sub&gt;-dependent protein kinase activity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ganglioside-inhibited protein kinase from pig brain</td>
</tr>
<tr>
<td>GM&lt;sub&gt;3&lt;/sub&gt; inhibition of EGF-dependent tyrosine phosphorylation</td>
</tr>
<tr>
<td>of EGF receptor</td>
</tr>
<tr>
<td>Ganglioside-induced phosphorylation of proteins in myelin</td>
</tr>
<tr>
<td>Inhibition of protein kinase C activity by gangliosides</td>
</tr>
<tr>
<td>GT&lt;sub&gt;1b&lt;/sub&gt;- and GD&lt;sub&gt;13&lt;/sub&gt;-activated protein kinase</td>
</tr>
<tr>
<td>Ca&lt;sup&gt;2+&lt;/sup&gt;- and ganglioside-dependent protein kinase</td>
</tr>
<tr>
<td>GM&lt;sub&gt;3&lt;/sub&gt; inhibition of tyrosine phosphorylation of the platelet-derived growth factor receptor</td>
</tr>
<tr>
<td>de-N-acetyl GM&lt;sub&gt;3&lt;/sub&gt; stimulation of tyrosine kinase activity of EGF receptor</td>
</tr>
</tbody>
</table>

**Cell Contact response**

<table>
<thead>
<tr>
<th>Inhibition of cell contact</th>
</tr>
</thead>
<tbody>
<tr>
<td>Globoside modulation of neuromuscular junction formation</td>
</tr>
<tr>
<td>GM&lt;sub&gt;2&lt;/sub&gt; modulation of retinal adhesion</td>
</tr>
<tr>
<td>Induction of GD&lt;sub&gt;1b&lt;/sub&gt; and GT&lt;sub&gt;1b&lt;/sub&gt; during neuroglial interaction</td>
</tr>
</tbody>
</table>

**Gangliosides as receptors and receptor cofactors**

| GM<sub>1</sub> binds B subunit of cholera toxin and mediates its mitogenic effect |
| G<sub>1b</sub> gangliosides as receptors for tetanotoxin |
| Binding of gangliosides to fibronectin                       |
| Binding of laminin, thrombospondin, and von Willebrand factor to sulfated glycolipids |

**Immune recognition**

<table>
<thead>
<tr>
<th>Blood group antigens</th>
</tr>
</thead>
<tbody>
<tr>
<td>Autoimmune antigens</td>
</tr>
<tr>
<td>Tumor antigens</td>
</tr>
<tr>
<td>Differentiation antigens</td>
</tr>
<tr>
<td>Lymphocyte markers</td>
</tr>
</tbody>
</table>

**Miscellaneous**
<table>
<thead>
<tr>
<th>Topic</th>
</tr>
</thead>
<tbody>
<tr>
<td>Embryo inversion by a complex glycolipid</td>
</tr>
<tr>
<td>Cerebrosides with antiulcerogenic activity</td>
</tr>
<tr>
<td>Stimulation of fruiting of Schizophyllum commune by plant cerebrosides</td>
</tr>
<tr>
<td>Modulation of sodium transport by complex gangliosides</td>
</tr>
<tr>
<td>&quot;Ganglioside syndrome&quot; in rabbits intensively immunized with GM₁ and GD₁a</td>
</tr>
<tr>
<td>Glycolipid changes with the cell cycle</td>
</tr>
</tbody>
</table>
VII. REFERENCES

(12) What follows, represents the experience acquired by Carter et. al. in processing 600 pounds of brain and 2000 pounds of spinal cord: The brain or spinal cord matter is ground into a soft paste and then extracted with acetone overnight. The acetone is decanted, and more acetone is added to the paste. A total of six extractions in this manner are performed.
The residue remaining after evaporation of the combined acetone extracts gave on average 9 to 12 kilos for spinal cord and 8.5 to 12 kilos for brain, of a light brown, gummy solid. Next the glycerophosphatides were extracted using ether, in which the sphingolipids are sparing soluble. The residue left behind was air dried, giving (5.2 to 6.0 kilos for spinal cord and 6.0 to 7.5 kilos for brain) a light brown friable material, which was moderately stable on exposure to air and could be stored almost indefinitely for use in the final step. This material was extracted 3 times with boiling ethanol for 10 - 15 minutes. These ethanol extracts were combined and allowed to cool overnight in an ice box. The precipitate was filtered, washed once with acetone and air dried to give a white powdery material (2.0 g to 2.5 g for spinal cord and 1.3 to 1.6 g for brain).

(15) Courbe, J. P. *1834*, *56* (series 2), 160.
(29) Seydel, P. V. Dissertation, Eidgenoessische Technische Hochschule, Zuerich, 1941.


(35) Thudichum, J. L. W., Die chemische Konstitution des Gehirns des Menschen und der Tiere, Pietzcker, F., Tubingen, 1901.

(36) For more recent historical information see a)K. Prostenik, M. Chem. Phys. Lipids 1970, 5, 1. b) The quote is found on the page before the preface of volume five of Chem. Phys. Lipids, the volume is dedicated to Herbert E. Carter, the subject matter of the articles pertain to the Chemistry and Metabolism of Sphingolipids, see Sweeney, C. C. Chem. Phys. Lipids 1970, 5, 1-300.


(49) This table was copied in whole from Hannun, Y. A.; Bell, R. M. *Science* 1989, 243, 500 and all references are cited therein. This is an excellent reference article concerning the biological functions of sphingolipids, biological effects of lysosphingolipids, mechanism of action of sphingosine and lysosphingolipids, sphingosine and lysosphingolipids as pathobiological molecules, lysosphingolipids and structural analogs as pharmacological agents, and possible physiological functions of the lysosphingolipids.

(50) For a review see Mueller, J. F. *Vitamins Hormones* 1964, 22, 787.


(56) For a broader and more detailed account of this material see *Chem Phys. Lipids* 1970, *vol.* 5. The entire issue was dedicated to Herbert E. Carter, who devoted a large share of his studies to the chemistry of sphingolipids, the papers contained within concern the chemistry and metabolism of sphingolipids.


(100) Tueting, D. R.; Echavarre, A. M.; Stille, J. K. Tetrahedron 1989, 45, 979.


(114) Post-doctoral researcher Jacques Rouden, notebook JHR-II-152.


IV. VITA

Thomas Christopher Nugent was born November 04, 1967 to Mr. and Mrs. Thomas Christopher Nugent in London, England. He graduated from Tottenville High School, Staten Island, New York in the spring of 1986 and enrolled at Virginia Polytechnic Institute and State University the following fall. In December, 1990 he earned a Bachelor of Science degree in chemistry. In fall, 1991 he began his graduate career under the direction of Dr. Tomas Hudlicky. His accomplishments while at Virginia Polytechnic Institute and State University are listed below:

Publications:


† - undergraduate researcher.

Presentations:


2) "Sphingosines and Their Derivatives from Monohalogented Benzenes," ACS
SE Regional Meeting," Johnson City, October 1993, Thomas Nugent and Tomas Hudlicky.


\[\text{Signature: Thomas Nugent}\]