Thematic Review Series: Sphingolipids

Biodiversity of sphingoid bases ("sphingosines") and related amino alcohols

Sarah T. Pruett,[1,*] **Anatoliy Bushnev,*** **Kerri Hagedorn,**[†] **Madhura Adiga,**[†] **Christopher A. Haynes,**[†] **M. Cameron Sullards,**[†] **Dennis C. Liotta,*** **and Alfred H. Merrill, Jr.**[2,†]

Department of Chemistry,* Emory University, Atlanta, GA 30322; and Schools of Biology, Chemistry, and Biochemistry and the Parker H. Petit Institute for Bioengineering and Bioscience,[†] Georgia Institute of Technology, Atlanta, GA 30332-0230

Abstract **"Sphingosin" was first described by J. L. W. Thudichum in 1884 and structurally characterized as 2*S*,3*R*,4*E*-2-aminooctadec-4-ene-1,3-diol in 1947 by Herb Carter, who also proposed the designation of "lipides derived from sphingosine as sphingolipides." This category of amino alcohols is now known to encompass hundreds of compounds that are referred to as sphingoid bases and sphingoid base-like compounds, which vary in chain length, number, position, and stereochemistry of double bonds, hydroxyl groups, and other functionalities. Some have especially intriguing features, such as the tail-to-tail combination of two sphingoid bases in the α,ω-sphingoids produced by sponges. Most of these compounds participate in cell structure and regulation, and some (such as the fumonisins) disrupt normal sphingolipid metabolism and cause plant and animal disease. Many of the naturally occurring and synthetic sphingoid bases are cytotoxic for cancer cells and pathogenic microorganisms or have other potentially useful bioactivities; hence, they offer promise as pharmaceutical leads.▪ This thematic review gives an overview of the biodiversity of the backbones of sphingolipids and the broader field of naturally occurring and synthetic sphingoid base-like compounds.—**Pruett, S. T., A. Bushnev, K. Hagedorn, M. Adiga, C. A. Haynes, M. C. Sullards, D. C. Liotta, and A. H. Merrill, Jr. **Biodiversity of sphingoid bases ("sphingosines") and related amino alcohols.** *J. Lipid Res.* 2008. **49:** 1621–1639.

Supplementary key words sphinganine • phytosphingosine • fumonisin • myriocin • long-chain base • anti-tumor • anti-fungal

Sphingolipids are composed of a structurally related family of backbones termed sphingoid bases, which are sometimes referred to as "long-chain bases" or "sphingosines"

after the original designation of the first isolated compound from brain as "sphingosin" by J. L. W. Thudichum in 1884 (1). Today, the term "sphingosine" is usually reserved for (2*S*,3*R*,4*E*)-2-aminooctadec-4-ene-1,3-diol (compound **6** in **Fig. 1**), which has important biological functions in cell signaling per se (2, 3) as well as after derivatization to the 1-phosphate (compound **9** in Fig. 1) (2, 4, 5), *N*-acylated metabolites (ceramides; compound **4** in Fig. 1) (2, 6, 7), and more complex phosphosphingolipids and glycosphingolipids with head groups attached to the hydroxyl on carbon 1. The structural diversity of the latter compounds is widely appreciated, with hundreds of head group variants for mammals alone, as was reviewed recently (8, 9) and addressed at a number of "omics" web sites, such as SphinGOMAP (www.sphingomap.org), the Japanese Lipid Bank (http://www.lipidbank.jp) and Glycoforum (http://www.glycoforum.gr.jp/), the Lipid Maps Consortium (www.lipidmaps.org), the Consortium for Functional Glycomics (http://www.functionalglycomics.org/fg/), and the Complex Carbohydrate Research Center at the University of Georgia (http://www.ccrc.uga.edu/~moremen/glycomics/).

Somewhat less well appreciated is that sphingoid bases also display considerable structural diversity, as was elegantly reviewed by K. A. Karlsson almost 40 years ago (10, 11). In remembrance of Herbert E. Carter, who first elucidated the structure of sphingosine **6** and dihydrosphingosine **2** (12) and "proposed to designate those lipides derived from sphingosine as sphingolipides" (13), this thematic review summarizes and updates points made previously regarding the structural diversity of sphingoid bases (10, 11) and expands the topic to include sphingoid bases and sphingoid base-like compounds that have been discov-

Abbreviations: SPT, serine palmitoyltransferase.
[1] Present address of S. T. Pruett: Yerkes National Primate Research Center, Emory University, Atlanta, GA 30329.
[2] To whom correspondence should be addressed.
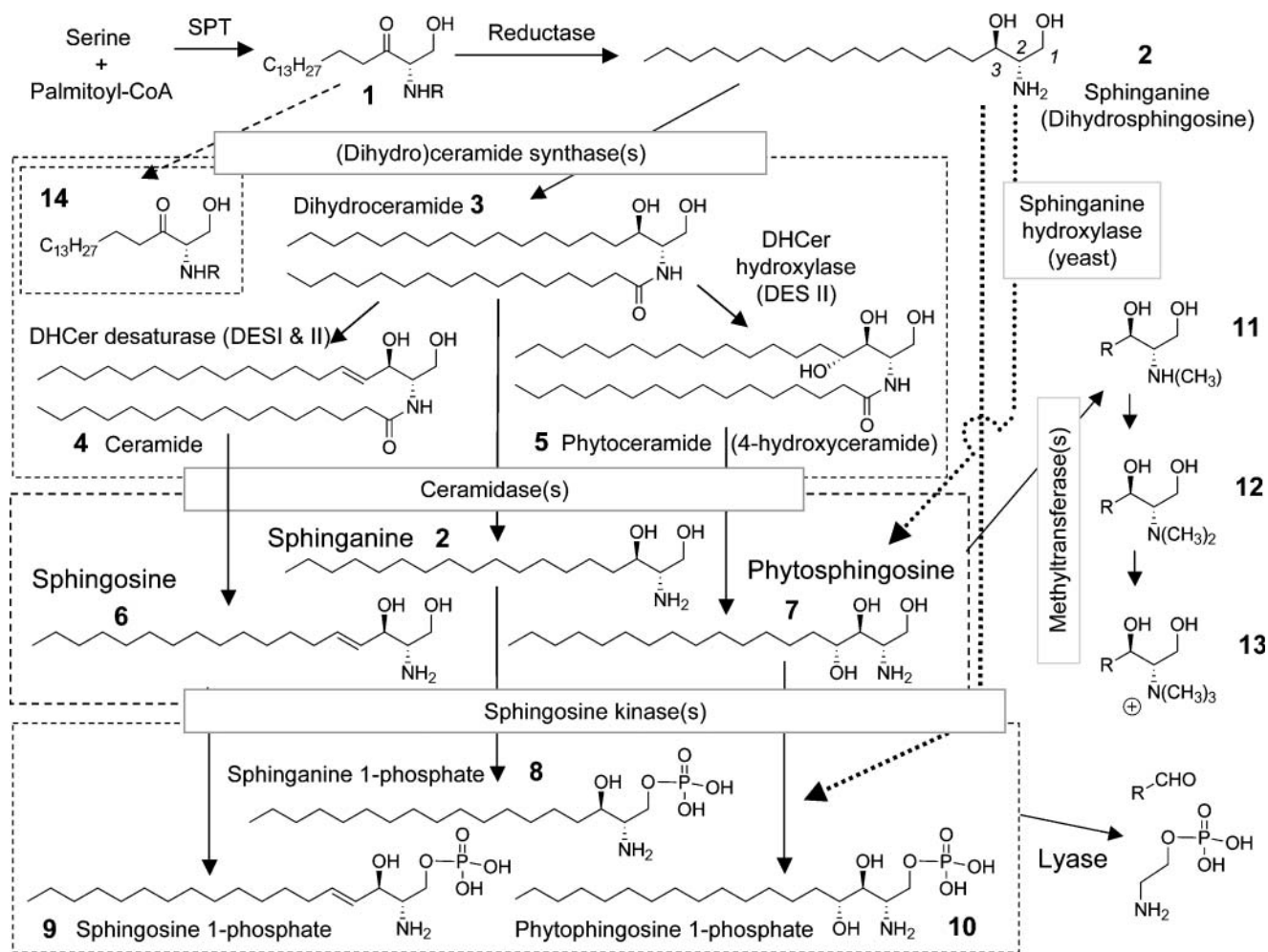 e-mail: al.merrill@biology.gatech.edu

Fig. 1. Biosynthesis and turnover of the three major categories of sphingoid bases in mammalian cells. DHR, dihydroceramide; SPT, serine palmitoyltransferase.

ered in intervening years. In addition to being fascinating for their biodiversity, some of these naturally occurring compounds (and synthetic analogs) are promising drug leads, while others cause disease, as exemplified by the fumonisins (14).

DIVERSITY IN THE SPHINGOID BASE BACKBONES OF SPHINGOLIPIDS

Within a few decades after the structure for sphingosine **6** had been determined (12) and sensitive methods for the analysis of sphingoid bases devised (15), there was evidence for >60 structural variations (10, 11). The 1997 International Union of Pure and Applied Chemists-International Union of Biochemists Joint Commission on Biochemical Nomenclature (16) proposed that "Sphingoids are long-chain aliphatic amino alcohols...represented by the compound originally called 'dihydrosphingosine' [(2S,3R)-2-amino-octadecane-1,3-diol]...[and]... imply a chain length of 18 carbon atoms." Dihydrosphingosine (compound **2** in Fig. 1; also called "sphinganine") is one of the major sphingoid bases found in many organisms as well as an early inter-

mediate in the de novo biosynthesis of sphingosine via desaturation of dihydroceramides (**3**) to produce ceramides (**4**) (17) and for the formation of "phytosphingosine" **7** (2S,3S,4R-2-aminooctadecane-1,3,4-triol) and what is colloquially referred to as "phytoceramide" (compound **5** in Fig. 1) via hydroxylation of sphinganine (18) or dihydroceramide (17). The alternative names (4E)-sphing-4-enine and (4E)-sphingenine are sometimes used to designate the specific location of the double bond of sphingosine.

The International Union of Pure and Applied Chemists-International Union of Biochemists Joint Commission (16) and others (19) have recommended naming chain length homologs by the root chemical name of the parent hydrocarbon (e.g., a 20 carbon sphinganine is called an icosasphinganine and one with 14 carbon atoms is called tetradecasphinganine), and the position and stereochemistry of substituents such as double bonds (with E/Z preferred over trans/cis), hydroxyl groups, methyl groups, etc., should be stated explicitly, if known. Examples of such compounds are shown in **Figs. 2 and 3**. A useful shorthand nomenclature is to give the number of hydroxyl groups ["d" for the two (di-) hydroxyls of sphingosine and sphinganine and "t" (tri-) for the additional hydroxyl in 4-
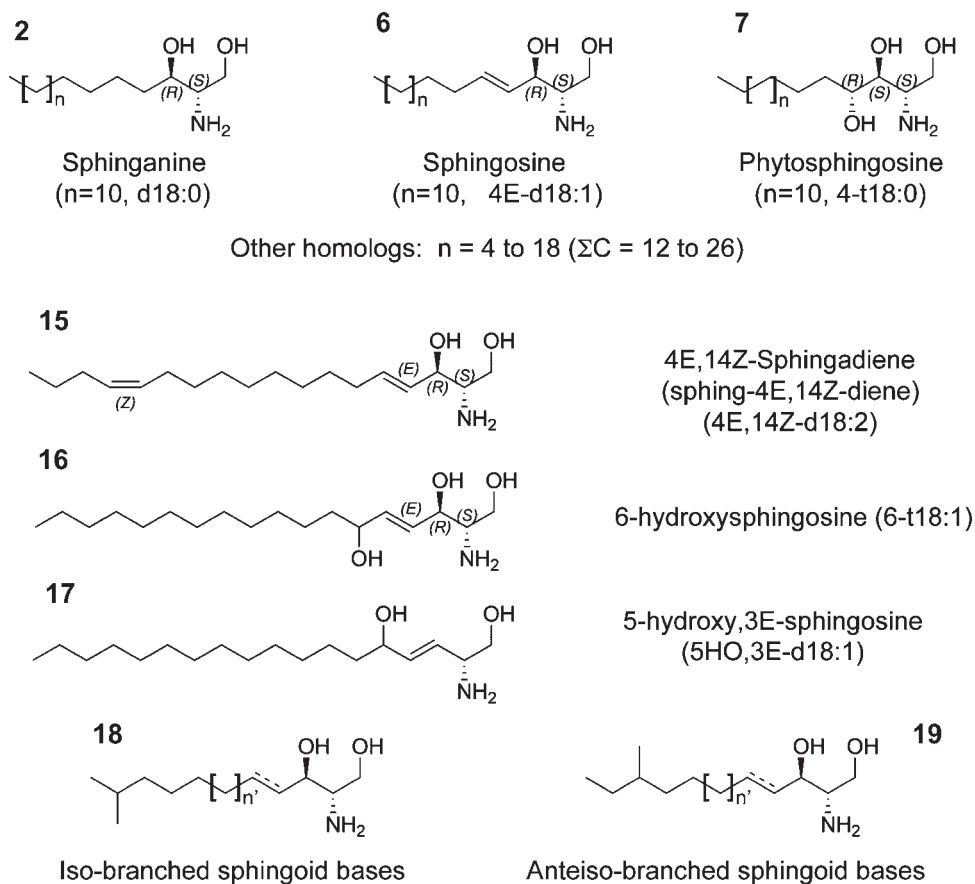
Fig. 2. Sphingoid bases of mammalian tissues.

hydroxysphinganine] followed by the number of carbon atoms in the backbone and the number of double bonds, with the location and configuration given as a prefix or suffix. Therefore, sphingosine is designated 4*E*-d18:1 (and sometimes d18:1$^{\Delta 4t}$), dihydrosphingosine is designated d18:0, and phytosphingosine (4-hydroxysphinganine) is designated t18:0.

Mammalian sphingoid bases

The predominance of 18 carbon sphingoid bases (d18:0, d18:1, and t18:0) in most mammalian sphingolipids is consistent with the preference of mammalian serine palmitoyltransferase (SPT) for saturated fatty acyl-CoAs with 16 ± 1 carbon atoms, combined with the abundance of palmitoyl-CoA (20, 21); nonetheless, small amounts of sphingoid bases with other chain lengths of 12 to 26 carbons have been reported (22, 23). The most common chain length variant is eisosasphingosine (2*S*,3*R*,4*E*-d20:1), which has been found in substantial amounts in gangliosides from brain (24) and human stomach and intestinal mucosa (25) and in sphingomyelin from rats bearing Morris hepatoma 7777 (26). Sphingoid bases with 16 carbon atoms are found in substantial proportions in bovine sphingolipids (e.g., 25–30% in milk sphingomyelin), which also have small amounts of other even and odd carbon chain length homologs (27, 28). Milk gangliosides appear to contain the unusual sphingoid bases 3-ethoxy-d15:0, 3-ethoxy-d17:0, and 9-methyl-3-ethoxy-d15:0 (29). Sphingomyelin and cere-

brosides in black epidermis from the Antarctic minke whale also have a high proportion (∼25%) of 16 carbon sphingoid bases (30).

Variation in the number and position of double bonds and hydroxyl groups also occurs. Plasma, brain, and human aorta contain a 4*E*,14*Z*-diene **15** (31, 32), and 6-hydroxysphingosine **16** is present in skin sphingolipids (23, 33, 34). An unusual sphingosine with the double bond between carbons 3 and 4 (5-hydroxy,3*E*-sphingosine; compound **17** in Fig. 3) has been found in acid-hydrolyzed brain extracts (35). While it is possible that **17** is a by-product of the acid hydrolysis (36, 37) (as will be discussed below for Fig. 8), it is nonetheless interesting that the *N*-octanoyl derivatives of both the 5*R* and 5*S* stereoisomers of **17** have been reported to be more potent than ceramide in inhibition of the proliferation of a human breast cancer cell line (MCF-7 cells) (38). This is surprising because the 4,5-*trans*-(*E*) double bond is usually necessary for ceramide signaling (39).

Sphingoid bases with branched side chains (such as the iso-**18** and anteiso-**19** configurations shown in Fig. 2) have been reported in sphingolipids from bovine milk and kidney (40), atherosclerotic human aorta (32), and pig harderian gland (41, 42) (which is not present in all mammals, including humans). Branched-chain sphingoid bases might become associated with mammalian tissues by microorganisms that are part of normal or pathogenic microflora, as illustrated by an iso-d15:0 sphingoid base that is found in *Porphyromonas gingivalis* from diseased dental tissues (43).
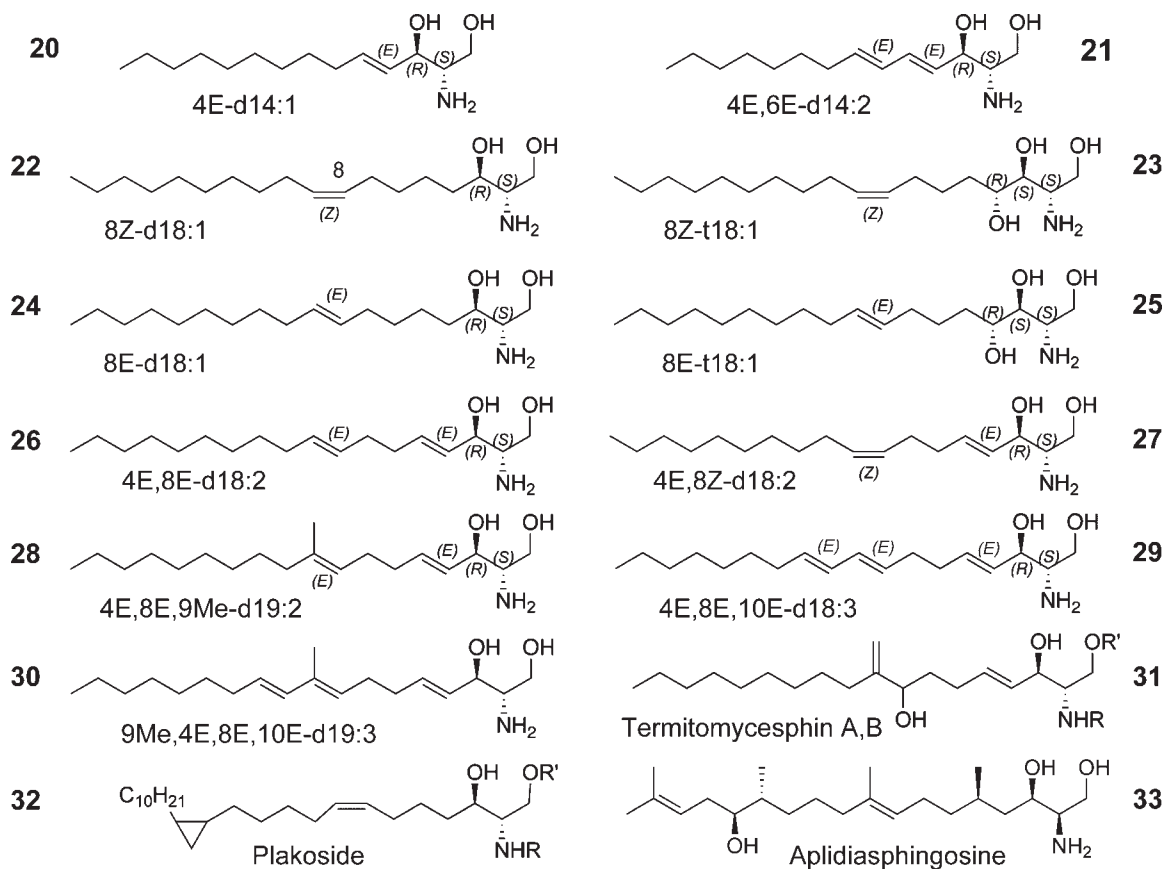
Fig. 3. Sphingoid bases found in diverse organisms other than mammals.

Interestingly, it appears that the poor absorption of "non-mammalian" sphingoid bases, such as the plant 4,8-diene, is due to the efflux of these compounds via P-glycoprotein in the apical membranes of enterocytes (44, 45), which raises the possibility that if this system is not working properly, there might be uptake of such compounds into mammalian tissues.

Small amounts of *N*- and *O*-methyl-sphingoid bases are sometimes found in mammalian sphingolipids and are thought mostly to be artifacts of the extraction and handling (36, 37) (as will be discussed below); however, a sphingosine *N*-methyltransferase activity has been found in mouse brain (46), and recent studies of mice treated with safingol, the L-*threo* stereoisomer of sphinganine, have found that it undergoes significant *N*-methylation (*N*-methyl, *N,N*-dimethyl, and *N,N,N*-trimethyl; compounds **11–13** in Fig. 1) and that under these conditions, there is also methylation of endogenous sphingosine and sphinganine (47), which suggests that the methyltransferases are inducible. The endogenous formation of *N,N*-dimethyl-sphingosine is interesting because this compound inhibits protein kinase C (48) and sphingosine kinase (49) as well as affects multiple cellular processes (50) and potently induces apoptosis in cancer cell lines (51).

Sphingoid bases of sphingolipids from other species

Fungi, plants, insects, and aquatic organisms extend the structural and compositional variation even further, as illustrated in Fig. 3. Insects have primarily 14 and 16 carbon sphingoid bases (52, 53) such as 4*E*-d14:1 (**20** in Fig. 3) and the conjugated diene 4*E*,6*E*-d14:2 **21** found in *Drosophila* (54). Nematodes have both iso-branched (4*E*,15-methyl-d17:1) and anteiso-branched (4*E*,14-methyl-d17:1) sphingoid bases (compare **18** and **19** in Fig. 3) (55, 56) in several categories of novel glycosphingolipids, including phosphocholine-containing glycosphingolipids that have been found in the parasitic nematodes *Onchocerca volvulus* (57) and *Ascaris suum* (58), with the latter also containing sulfatides (which is not common in invertebrates) (58). A 15-carbon atom (unbranched) phytosphingosine (in amide linkage with a 21:0 iso-branched α-hydroxy fatty acid) has been found in urine of the female hairy crab, *Erimacrus isenbeckii*, and serves as a sex pheromone to elicit precopulatory behavior in males (59).

Recent studies of a group of viruses (Coccolithovirus) that infect the marine calcifying microalga *Emiliania huxleyi* have revealed that the viral genome contains a cluster of putative sphingolipid biosynthetic genes, including a SPT (Fig. 1) that utilizes myristoyl-CoA when expressed in yeast (60). This might cause an infected host to produce a 16 carbon chain length sphingoid base, which is interesting because at least one virus (picornavirus) has a capsid protein with a hydrophobic pocket that has been suggested to bind sphingosine (61).

Other types of structural variation include the location of the double bond(s), as shown for compounds **22** and **24**

in Fig. 3, where the double bond is at the 8,9 position versus 4,5 for sphingosine **6**. Double bonds are also seen in the phytosphingosine-type compounds **23** and **25** that are common backbones of plants (62), which also have 4,8-dienes (**25**–**27**), but interestingly, very little of the prevalent species of mammals (sphingosine, 4*E*-d18:1), with only a single 4*E* double bond. Plant 4,8-dienes sometimes have branching methyl groups (or hydroxyls at other positions) (62); however, branched sphingoid bases such as 4*E*,8*E*,9-methyl-d19:2 (**28** in Fig. 3) and 4*E*,8*Z*,9-methyl-d19:2 (data not shown) are considered to be more typical of fungal sphingolipids (63, 64), including human pathogens such as *Cryptococcus neoformans* (64, 65). It appears that fungi produce different types of backbones for incorporation into different categories of more complex sphingolipids, based on studies of the mycelial forms of *Histoplasma capsulatum*, which found compound **28** in glucosylceramides but phytosphingosine **7** as the major backbone of the glycosylinositol phosphorylceramides (66). Fungi are sources for a wide variety of unique sphingoid bases, such as the compounds named termitomycesphins (**31** in Fig. 3) from the Chinese mushroom *Termitomyces albuminosus* (67). Other interesting examples will be elaborated upon in discussion of **Table 1** and **Figs. 4 and 5**.

Sphingoid bases with three double bonds, such as (4*E*, 8*E*,10*E*)-2-amino-4,8,10-octadecatriene-1,3-diol (4*E*,8*E*,10*E*-d18:3; **29** in Fig. 3), are found in the spermatozoa of the starfish, *Asterias amurensis* (68), and the branched version, 2-amino-9-methyl-4,8,10-octadecatriene-1,3-diol (**30** in Fig. 3), has been identified in squid nerve sphingomyelin (69). Sponges are another source of sphingoid bases with interesting features, such as the cyclopropane ring in the alkyl side chain of plakosides (**32** in Fig. 3), a family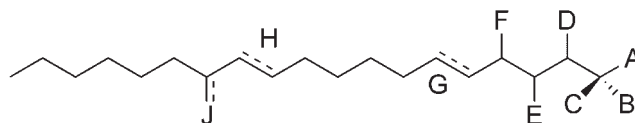 of immunosuppressive prenylated galactosphingolipids produced by *Plakortis simplex* (70). Sphingoid bases with a terpenoid alkyl chain, the aplidiasphingosines (compound **33** in Fig. 4; 1,2-amino-5,9,13,17-tetramethyl-8,16-octadecadiene-1,3,14-triol), have been isolated from the marine tunicate *Aplidium* species (71, 72) and noted to have antimicrobial and antitumorial activity (71, 73).

Many of the species in the genus *Sphingomonas*, which are Gram-positive bacteria with glycosphingolipids instead of lipopolysaccharide in the outer membrane, have sphingoid bases with a cyclopropane ring, such as the 13,14-cyclopropane-eicosasphinganine produced by *Sphingomonas adhaesiva* (74). Because the SPT of *Sphingomonaspaucimobilis* is a cytoplasmic homodimer instead of the membrane-bound heterodimer found in most other organisms, it has been possible to elucidate the crystal structure of the holo form of *S. paucimobilis* SPT at 1.3 A resolution (75) and to conduct in-depth spectroscopic studies of the catalytic mechanism of this pyridoxal 5′-phosphate-dependent enzyme (76) and comparative studies of the three novel SPT genes from *Sphingobacterium multivorum*, *Sphingobacterium spiritivorum*, and *Bdellovibrio stolpii* (77).

3-Keto sphingoid bases

The first product of de novo sphingoid base biosynthesis, 3-ketosphinganine (**1** in Fig. 1), is often not detected in organisms and tissues, because under most circumstances it is rapidly reduced to sphinganine (78); nonetheless, rat liver mitochondria have been reported to contain *N*-acylated and *O*-glycosylated derivatives of 3-keto bases (79), and 3-ketodihydroceramide (compound **14** in Fig. 1) has been detected in cells when SPT activity is very high (2); therefore, it appears that when this keto intermediate is not reduced rapidly enough, it is acylated by the next enzyme of the pathway. This might have biological consequences,

TABLE 1. Sphingoid base-like inhibitors of serine palmitoyltransferase

| Compound Name	J	H	G	F	E	D	C	B	A	Ref.
Myriocin (**36** in Fig. 4)	O		=	H	OH (*S*)	OH (*S*)	COOH	NH_2	CH_2OH	(76, 84–86)
Sphingofungin A	OH		=	OH (*S*)	OH (*R*)	OH (*R*)	COOH	$NHCNHNH_2$	H	(76, 86, 91)
Sphingofungin B	OH		=	OH (*S*)	OH (*R*)	OH (*R*)	COOH	NH_2	H	(76, 86, 91)
Sphingofungin C	OH		=	*O*-Acetyl (*S*)	OH (*R*)	OH (*R*)	COOH	NH_2	H	(76, 86, 91)
Sphingofungin D	OH		=	OH (*S*)	OH (*R*)	OH (*R*)	COOH	HN-Acetyl	H	(76, 86, 91)
Sphingofungin E	O		=	OH (*S*)	OH (*R*)	OH (*R*)	NH_2	COOH	CH_2OH	(76, 86, 91)
Sphingofungin F	O		=	OH (*S*)	OH (*R*)	OH (*R*)	COOH	NH_2	CH_3	(76, 86, 91)
Sulfamisterin (**37** in Fig. 4)	O			H	H	OSO_3Na (*R*)	COOH	NH_2	CH_2OH	(89)
Mycestericin A	O	=		H	OH (*R*)	OH (*S*)	COOH	NH_2	CH_2OH	(85)
Mycestericin B	OH			H	OH (*R*)	OH	COOH	NH_2	CH_2OH	(85)
Mycestericin C	O			H	OH (*S*)	OH (*S*)	H	H	H	(85)
Mycestericin D	O		=	H	H	OH (*S*)	NH_2	COOH	CH_2OH	(85)
Mycestericin E						OH (*R*)				
Mycestericin F	O		=	H	H	OH (*S*)	NH_2	COOH	CH_2OH	(85)
Mycestericin G						OH (*R*)				
Malonofungin	O		*O*-Ac (*S*)		OH (*R*)	OH (*R*)	NH_2	COOH	COOH	(93)
Fumifungin	OH		OH	O-Ac		OH	NH_2, COOH, and H (stereochemistry not specified)			(94)
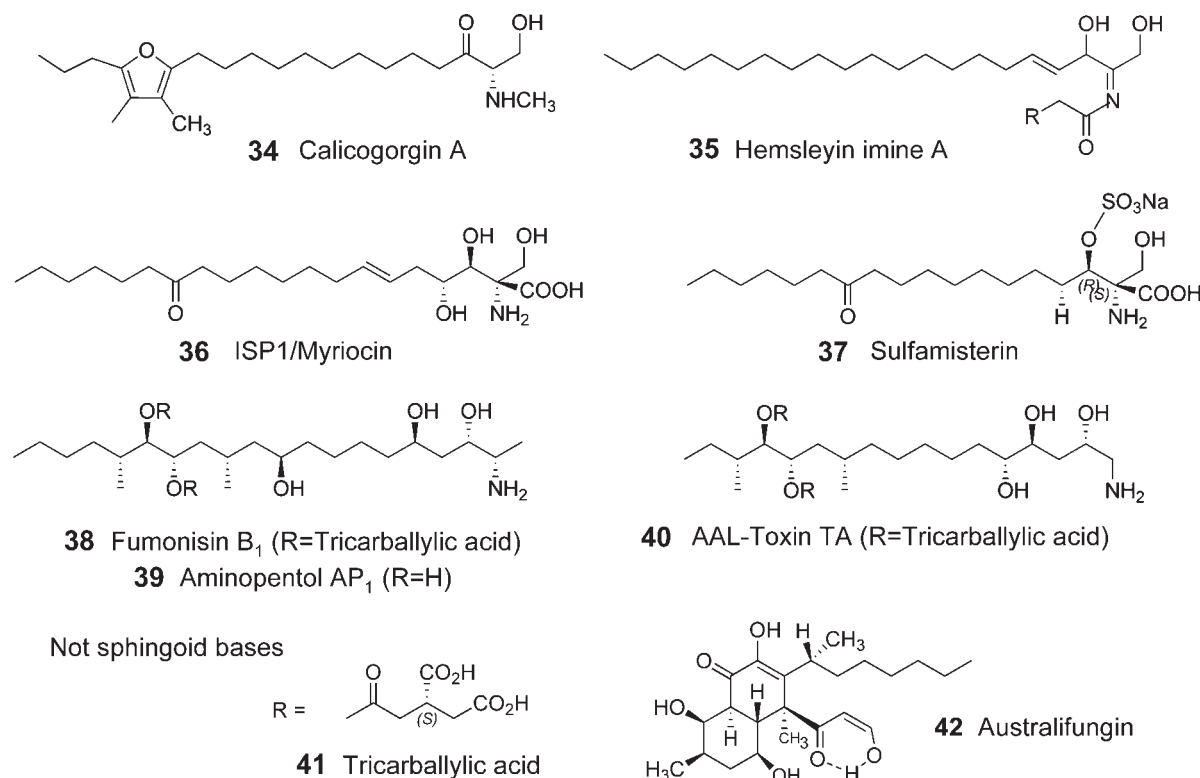
Fig. 4. Sphingoid base-like compounds that mimic metabolites and/or inhibit early steps of sphingolipid metabolism. In the category "not sphingoid bases" are tricarballylic acid, which is the "R" group found on fumonisins and AAL toxins, and australifungin, which is shown as an example of a ceramide synthase inhibitor that is not a sphingoid base-like compound.

because short-chain 3-ketoceramides have been found to be strong inducers of apoptosis in human leukemia HL-60 cells (80). A family of 3-keto sphingoid base-like compounds, the calicogorgins (**34** in Fig. 4), have been found in marine invertebrates and are repellent and lethal against the snail *Drupella. fragum* (81, 82). Another type of oxidized backbone, an imine, is found in hemsleyin imine A (2-octadecanoylimino-heneicosan-1,3-diol; compound **35**), which was isolated (83) from the rhizomes of *Hemsleya macrocarpa* var. *clavata*, a perennial plant found in northwestern China that is used as folk medicine for the treatment of bronchitis, bacillary dysentery, and tuberculosis.

SPHINGOID BASES AND SPHINGOID BASE-LIKE COMPOUNDS THAT DISRUPT SPHINGOLIPID METABOLISM

SPT inhibitors

Several categories of fungi produce SPT inhibitors that are sphingoid base-like compounds, as summarized in Table 1 and illustrated for a few examples in Fig. 4: compound **36** in Fig. 4, which has been named ISP-1 (from *Isaria sinclairii*) (76, 84, 86), myriocin (isolated from *Myrioccocum albomyces*) (87), and thermozycidin (from *Mycelia sterilia*) (88); sulfamisterin from *Pycnidiella* species (**37** in Fig. 4) (89), a sulfated, 18 carbon myriocin-like analog (minus the 4-hydroxyl group); sphingofungins produced by *Aspergillus* and *Paecilomyces* (76, 86, 90–92) and other compounds with

similar structural features (Table 1), the mycestericins (from *Mycelia sterilia*) (85), malonofungin (from *Phaeramularia fusimaculans*) (93), and fumifungin from *Aspergillus fumigatus* (94). Some of these have not only been found to be potent inhibitors of SPT but also to have immunosuppressive activity, although inhibition of this enzyme is not obligatory for immunosuppression by some of the compounds in this structural series (95), because a more specific immunosuppressive agent (FTY720) has been found that is not an SPT inhibitor. Many also have antifungal activity, as do other categories of SPT inhibitors that are not sphingoid base analogs, such as lipoxamycin (96) and viridiofungins (97).

ISP-1/myriocin **36** has been studied most extensively because it is highly potent, with an IC_{50} in the nanomolar range (84), and commercially available. Spectroscopic studies suggest that ISP-1/myriocin inhibits activity by forming an external aldimine with pyridoxal 5'-phosphate in the active site of SPT, as does the substrate serine (76). Indeed, all of the SPT inhibitors share this resemblance to serine (Table 1). It is noteworthy that the sulfate group of sulfomisterin (37) only reduces the potency of sulfamisterin by ~10-fold versus ISP-1/myriocin when assayed in a cell-free system, which implies that this enzyme can accommodate both the extra size and charge at this position (89).

There is growing interest in the pharmaceutical potential of SPT inhibition because ISP-1/myriocin interferes with the production of cytotoxic ceramide in response to a wide variety of stresses (98), suppresses virus infectivity (99, 100), alters brain amines (101), and suppresses athero-
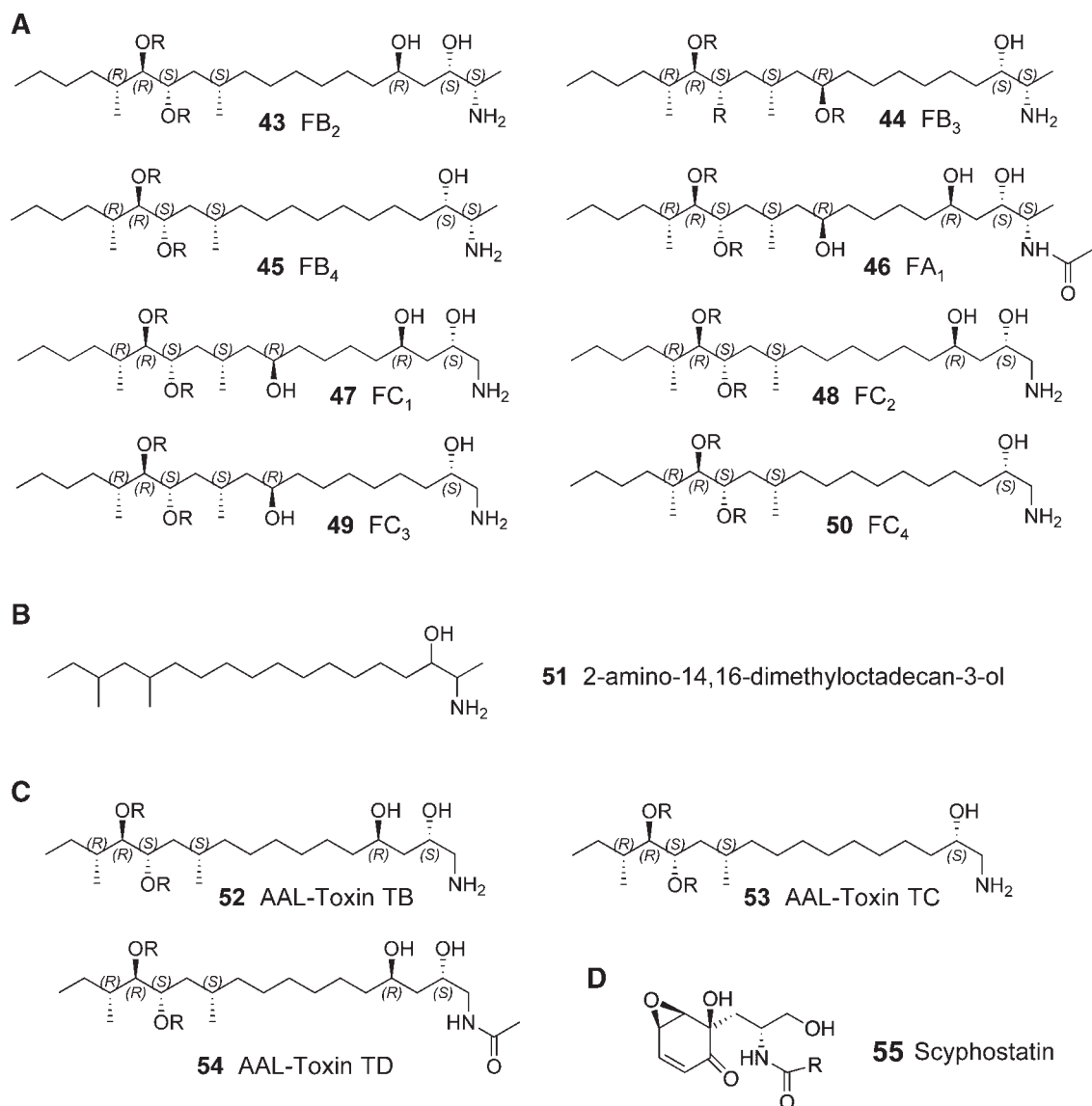
Fig. 5. Structures of the major members of the fumonisin (A) and AAL toxin (C) families. Also shown are a compound with a fumonisin-like backbone but without side chain hydroxyls (B) and the sphingomyelinase inhibitor scyphostatin (D).

sclerotic lesions (102–104). It has been noted that the *Isaria* (=*Cordyceps*) group of fungi also includes *Cordyceps sinensis* Sacc., which has been used in Chinese traditional medicine as a drug (Chinese name, *dong chong xia cao*) for eternal youth (85, 105).

Ceramide synthase inhibitors

Fungi produce diverse sphingoid base-like compounds (typified by compounds **38–40** in Fig. 4) and nonsphingoid base-like compounds (compound **42** in Fig. 4) that inhibit ceramide synthases, the enzymes that are responsible for the acylation of sphingoid bases (106, 107) (Fig. 1). The first to be discovered (14) were the fumonisins (represented by fumonisin B_1 **38** in Fig. 4 and additional family members **43–50** in Fig. 5), which are mycotoxins produced by *Fusarium verticolloides*. This fungus commonly infests maize and causes diseases of plants (108) and animals (including humans) (109, 110) that consume these compounds

as food contaminants (111). The traditional method of preparation of corn (maize) for tortillas involves treating the maize with lime (nixtamalization) followed by extensive washing, which removes a substantial portion of the fumonisins and hydrolyzes the tricarballylic acid side chains (compound **41** in Fig. 4) that are ester linked to side chain hydroxyls (as "R" on compound **38** and **43–50**). The compounds that are produced are usually referred to as "aminopentols" numbered according to the parent fumonisin (such as AP_1 **39** from FB_1 **38** in Fig. 4) (112, 113). Removal of the tricarballylic acid(s) diminishes the potency of the inhibition of ceramide synthase (which has been suggested to be due to the interaction of the polyanionic side chains with the binding site for the fatty acyl-CoA cosubstrate) (109) but also converts the compound into a ceramide synthase substrate that is acylated to produce cytotoxic *N*-acylaminopentols (114–116); therefore, it appears that nixtamalization will be most ef-

fective at detoxifying contaminated maize by washing away of the fumonisins rather than just the removal of the tricarballylic side chains.

The discovery of the fumonisins in South Africa in 1988 is a classic story of scientific sleuthing by W. S. A. Marasas (117) and coworkers, beginning with the isolation of *F. verticillioides* (originally named *F. moniliforme*) from moldy corn implicated in a field outbreak of equine leukoencephalomalacia in South Africa in 1970; observation that this fungus was also prevalent in moldy corn consumed by people in high-incidence areas of esophageal cancer in the Transkei region of South Africa; demonstration that feeding culture material from *F. verticillioides* strain MRC 826 to horses recapitulated the equine disease and also caused porcine pulmonary edema syndrome in pigs and liver cancer in rats; isolation and structural elucidation of the fumonisins; and demonstration that purified fumonisins cause these symptoms.

Soon after the structure of fumonisin B$_1$ was elucidated (118), the similarity to sphinganine led to the discovery of its inhibition of ceramide synthase (14). This was significant not only because it provided a specific target for this mycotoxin but also because it was the first case in which a defect in sphingolipid biosynthesis causes disease, in contrast to the many diseases that were then known to be caused by genetic defects in sphingolipid turnover (e.g., Niemann-Pick, Gaucher's disease, etc.) (119). It is not surprising that fumonisins have a wide range of pathologic effects, since inhibition of ceramide synthase (Fig. 1) causes not only the depletion of ceramides and complex sphingolipids but also the buildup of highly bioactive compounds (sphinganine, sphinganine 1-phosphate, *N*-acetyl-sphinganine, and other sphingoid bases depending on the time of treatment and dosage) (109, 110). Fumonisin also causes a "mystery" compound to appear (120), which was recently found to be 1-deoxy-sphinganine (N. C. Zitomer, personal communication). In addition to the tissue damage (e.g., equine leukoencephalomalacia, porcine pulmonary edema, hepatotoxicity, and renal toxicity) and carcinogenicity generally associated with fumonisin consumption (117), one might suspect that changes in so many bioactive species could adversely affect health in more ways than are currently appreciated, and recent laboratory (121–124) and epidemiologic (125) studies have implicated fumonisins in birth defects.

Figure 5 shows the types of structural variants found in fumonisins and a related family of mycotoxins, the AAL toxins (represented by AAL toxin TA **40** in Fig. 4), which are produced by *Alternaria alternata* and cause disease in plants (108, 126, 127). All are sphingoid base-like amino alcohols, but the B series fumonisins (**38**, **39**, and **43–45**) are 1-deoxy-sphingoid bases, the C series fumonisins (**47–50**) and the AAL toxins (**40** and **52–54**) lack the 1-hydroxymethyl group, and the A series fumonisin (represented by FA$_1$ **46**) and AAL-toxin TD (**54**) are *N*-acetylated. Another metabolite, 2-amino-14,16-dimethyloctadecan-3-ol (compound **51** in Fig. 5), has been isolated from a strain of *Fusarium avenaceum* cultured on rice and found to be cytotoxic for the rat hepatoma cell line H4IIE-W and a porcine

epithelial kidney cell line at micromolar concentrations (128). This compound is more "sphinganine-like" because it does not have the side chain hydroxyls; therefore, it should undergo substantial acylation like other 1-deoxy-sphinganines (114). The authors comment that *F. avenaceum* has the potential to produce the metabolite under field conditions that might occur in northern Europe (128), meaning in regions that have heretofore been found to have little fumonisin contamination because *F. verticillioides* requires a warmer climate.

The broad category of toxins that mimic sphingolipids has been referred to as sphinganine (or sphingosine/ceramide) analog toxins (129, 130). This categorization has been employed to compare not only the structure and function of these compounds but also their biosynthesis (131), because sphingoid base analogs minus the 1-hydroxyl (compounds **38**, **39**, **43–45**, and **51**) versus the 1-hydroxymethyl (compounds **40**, **47–50**, and **52–54**) groups are biosynthesized from alanine versus glycine, respectively, instead of serine, which is the source of the 1-hydroxyl and 2-amino groups of sphingoid bases (Fig. 1).

Other steps of sphingolipid metabolism

Naturally occurring inhibitors of other steps of sphingolipid metabolism have been found (132), such as rustmicin, which inhibits inositol phosphoceramide synthase (133); however, with the exception of scyphostatin (compound **55** in Fig. 5), a neutral sphingomyelinase inhibitor from *Trichopeziza mollissima* (134), most do not fit in the category of sphingoid bases or sphingoid base-like compounds.

"Simpler" 1-deoxy-sphingoid bases

In addition to the relatively complex 1-deoxy-sphingoid bases discussed above, plants and marine organisms have a wide variety of simpler compounds, even a species that is identical to sphinganine but lacking the 1-hydroxyl. This compound (spisulosine **56** in **Fig. 6**) has been isolated from the clam *Spisula polynyma* (Stimpson's surf clam or Atlantic surf clam; syn. *Mactromeris polynyma*) (135). It is also referred to as "ES-285" (the "285" refers to its molecular weight) as an investigational marine anticancer drug (136) because it inhibits the proliferation of numerous cancer cell lines. The mechanisms for its effects are not known, but it disrupts actin stress fibers through the inactivation of Rho (135) and has been suggested to induce cell death in the prostate cancer cell lines PC-3 and LNCaP via stimulation of the de novo synthesis of ceramide and protein kinase Cζ activation (137). Spisulosine has also been found to activate caspase 3 and 12 and to modify the phosphorylation of p53, but it did not affect JNK, Erks, or Akt; therefore, it has been suggested to trigger an atypical cell death program compared with other sphingosine-dependent apoptosis pathways (138).

In other studies, 1-deoxy-sphingoid bases have been shown to be more cytotoxic than sphingosine against HT29 cells (139). This category of compound is also known to be acylated by ceramide synthase (114); hence, it is possible that 1-deoxyceramides may play a role in the biological effects.
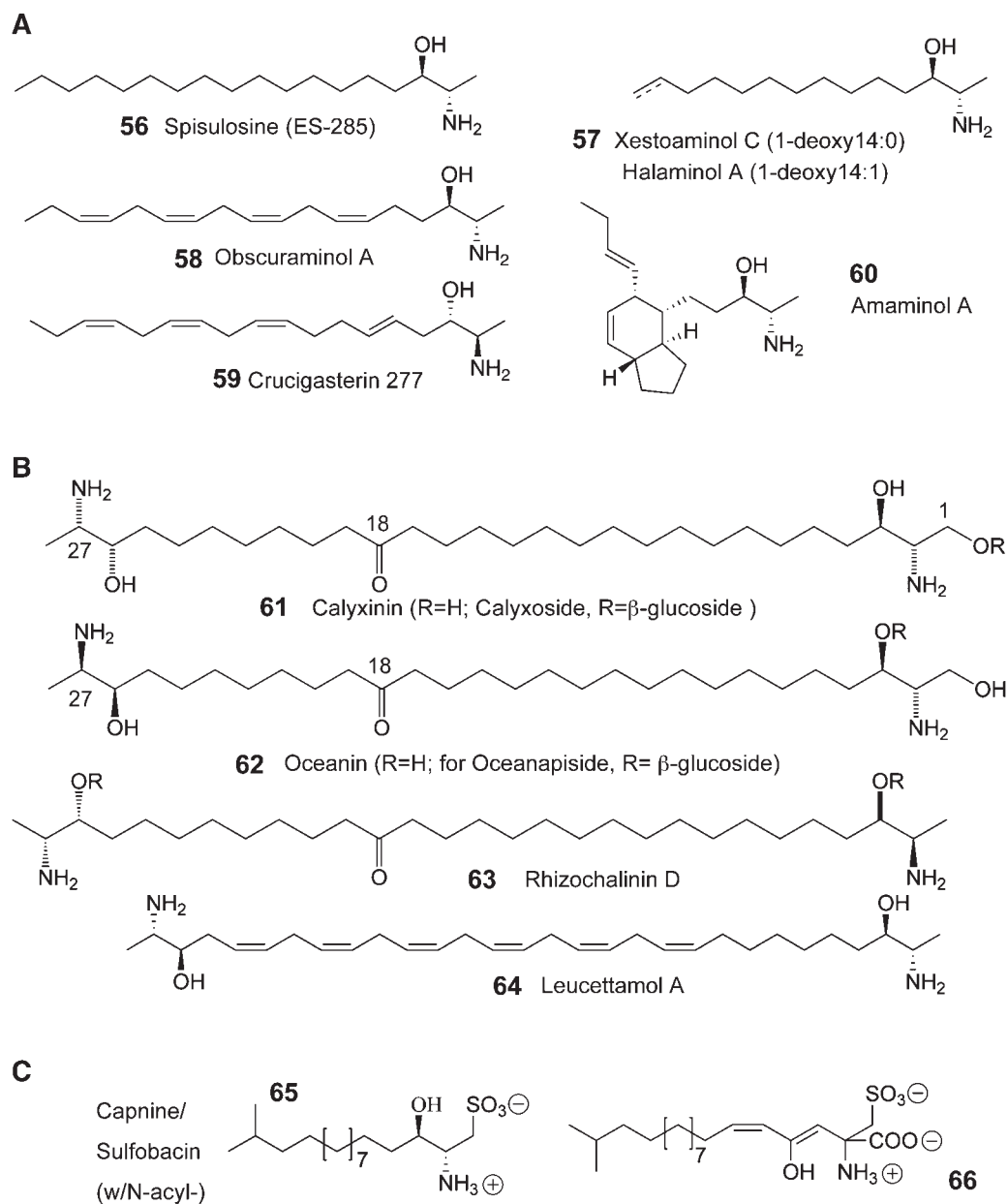
Fig. 6. Novel sphingoid base-like compounds. A: 1-Deoxy compounds. B: α,ω-Bifunctional sphingoid bases. C: Sphingoid base-like compounds with sulfonic acid.

Other spisulosine-related compounds include the shorter chain length xestoaminols (represented by xestoaminol C, 1-deoxy14:0, from *Xestospongia* species and halaminol A, 1-deoxy14:1, from *Haliclona n.* species; **57** in Fig. 6) (140), and the polyunsaturated obscuraminols (represented by obscuraminol A **58** in Fig. 6) from *Pseudodistoma obscurum* (141). The obscuraminols were isolated from a chloroform extract of this sponge that was cytotoxic for mouse lymphoma P-388, human lung carcinoma A-549, and human colon carcinoma HT-29 tumor cell lines, but the isolated compounds were only mildly cytotoxic. Crucigasterins (represented by crucigasterin 277; **59** in Fig. 6), from *Pseudodistoma crucigaster* (142), are similar to these compounds but have 2*R*,3*S* rather than 2*S*,3*R* stereochemistry (135, 137, 141). Crucigasterins show antimicrobial activity against

Bacillus subtilis and are cytotoxic against mouse lymphocytic leukemia L1210 cells (142).

There are also 1-deoxy-sphingoid base-like compounds with five and six member rings as part of the side chain (amaminol A, isolated from tunicates of family Polyclinidae; compound **60** in Fig. 6) (143). This type of compound might be formed by cyclization of a polyene such as **58** or **59**.

α,ω-SPHINGOIDS: "TWO-HEADED" SPHINGOID BASE-LIKE COMPOUNDS

A fascinating series of compounds, which have been referred to as "α,ω-bifunctionalized amino alcohols" (144),

so we have called them "α,ω-sphingoid base-like compounds," resemble "two-headed" sphingoid bases (i.e., two sphingoid bases connected tail to tail; compounds **61**–**64** in Fig. 6). Calyxinin **61**, the aglycone of a sphingoid base 1-β-glucoside named calyxoside from sponges of the genus *Calyx* in the family *Oceanapiidae*, resembles sphinganine (numbering from position 1) on one end and a 1-deoxy-sphinganine at the other end (with the opposite, *threo*, stereochemistry) (145). It has been proposed that calyxoside is probably not formed from the union of the tails of two identical smaller lipids because the ketone is not at the expected position (unless the ketone is created later) (144, 145).

Oceanin (**62** in Fig. 6) is the aglycone of oceanapiside from *Oceanapia phillipensis* (146–148). While oceanin is very similar to calyxinin with the same stereochemistry as a sphingoid base for carbons 2 and 3 (*S,R-erythro*) as well as a *threo* stereochemistry on the ω-amino alcohol, the latter is opposite for these compounds (i.e., oceanin is 26*R*,27*R* versus 26*S*,27*S* for calyxinin). They differ also in the position of *O*-glucosylation for oceanapiside (the hydroxyl at carbon 3) and calyxoside (carbon 1) and were once thought to have the ketone in different positions (11 vs. 18); however, this has been revised and the structures shown in Fig. 6 reflect the most recent report (148). The carbohydrate plays a major role in some of the biological activities of oceanapiside, which has been reported to inhibit mycothiol-/*S*/-conjugate amidase, a mycobacterial detoxification enzyme, 50- to 100-fold more potently than the aglycone (149). In contrast, the carbohydrate has little, and perhaps a slightly negative, effect on the antifungal activity of oceanapiside versus oceanin against *Candida glabrata*, with minimum inhibitory concentrations of 10 and 3 μg/ml, respectively (144).

Two other α,ω-sphingoid base-like compounds are shown in Fig. 6: rhizochalin D (63), from *Rhizochalina incrustata*, which is a 1-deoxy-sphingoid base (both amino alcohols with *threo* stereochemistry) on both ends and glycosylated at carbon 3 (142, 144, 148); and leucettamol A (64), which has both 1-deoxy amino alcohols with *erythro* stereochemistry and polyunsaturation in the alkyl chain. This compound represents a large family of polyunsaturated "leucettamols" found in *Leucetta* species, which differ in the numbers and locations of double bonds in the alkyl chain (142, 150, 151). Another compound not shown, BRS1, is a 30 carbon bis-amino,bis-hydroxy polyunsaturated lipid from an Australian calcareous sponge (*Calcarea*) (152). Each of these has been found to have some form of biological activity, such as antibacterial activity against *Staphylococcus aureus* and cytotoxicity for Ehrlich carcinoma cells for rhizochalin (153) and inhibition of phorbol ester binding to protein kinase C for BRS1 (which is a known feature of sphingosine) (152).

One can envision that the purpose of the two sphingoid base (-like) moieties is to allow these compounds to interact simultaneously with two binding sites on a single target or perhaps to interact with more than one target to effect their biological response (144). This might include interaction with two binding sites on a dimeric enzyme of sphin-golipid metabolism (or signaling) or perhaps the active sites of two enzymes in close proximity in a multienzyme complex. It is also possible that the length of these compounds enables them to span a membrane bilayer for structural or signaling purposes.

Capnines, sulfobacins, and other 1-sulfonosphingoids

Gliding bacteria of the genus *Cytophaga* synthesize sulfonolipids that contain capnine (1-deoxy,15-methyl-hexadecasphinganine-1-sulfonic acid; compound **65** in Fig. 6) (154), which appears to be the same as the backbone for the sulfobacins that have been isolated from the culture broth of *Chryseobacterium* species (*Flavobacterium* species) *NR 2993* and for which the stereochemistry has been assigned (2*R*,3*R*) (155). Studies of capnine biosynthesis (156, 157) using isotopically labeled precursors have suggested that the biosynthesis of capnine occurs by the condensation of 13-methylmyristoyl-CoA with cysteic acid in a reaction analogous to the condensation of palmitoyl-CoA with serine for the biosynthesis of sphingolipids. A cysteine auxotroph of *Cytophaga johnsonae* was able to incorporate sulfur from sulfate into cysteate and sulfonolipid, which further indicates that cysteine per se is not an obligatory intermediate of capnine biosynthesis (158). Sulfobacins A and B (which vary in the *N*-acyl fatty acid) have been found to be von Willebrand factor receptor antagonists (159). Another sulfonolipid (compound **66** in Fig. 6) that has features similar to the SPT inhibitors (particularly sulfamisterin **37**) has been found in the halophilic bacterium *Salinibacter ruber* (160).

Heteroyclic sphingoid base-like compounds

There are a large number of sphingoid base-like compounds in which the amino group is part of a heterocyclic ring, as illustrated in **Fig. 7**. The simplest heterocycle is the aziridine (azacyclopropene) ring found in 4E-(*R*)-dysidazirine **67** (161) and (*S*)-antazirine **68** (162), which were isolated from the marine sponge *Dysidea fragilis* (Dysideidae). These do not formally qualify as sphingoid bases because they are not amino alcohols; however, it is easy to envision how the aziridine ring might be formed via a 2-amino,3-keto intermediate similar to that formed in de novo sphingolipid biosynthesis (e.g., compound **1** in Fig. 1).

Penaresidin A and B (**69** in Fig. 7) (163–165) and penazetidine A **70** (166) are azetidines produced by *Penares* sponges. Penaresidins have been reported to activate AT-Pases (164), and penazetidine A is an inhibitor of protein kinase C (166, 167). Both have shown cytotoxicity against a variety of cell types, and analogs of penaresidin B with a simple alkyl chain have been found to be considerably (i.e., up to 10-fold, or an IC$_{50}$ of ∼1 μM) more cytotoxic against lung (A549) and colon (HT29) cancer cell lines and showed antibacterial activity against Gram-positive bacteria (*Bacillus subtilis*, *Micrococcus luteus*, and *Staphylococcus aureus*) and, in one case, against Gram-negative *Escherichia coli* (165).

Pramanicin (compound **71** in Fig. 7) (168) is an interesting sphingoid base-like pyrrole biosynthesized from ser-
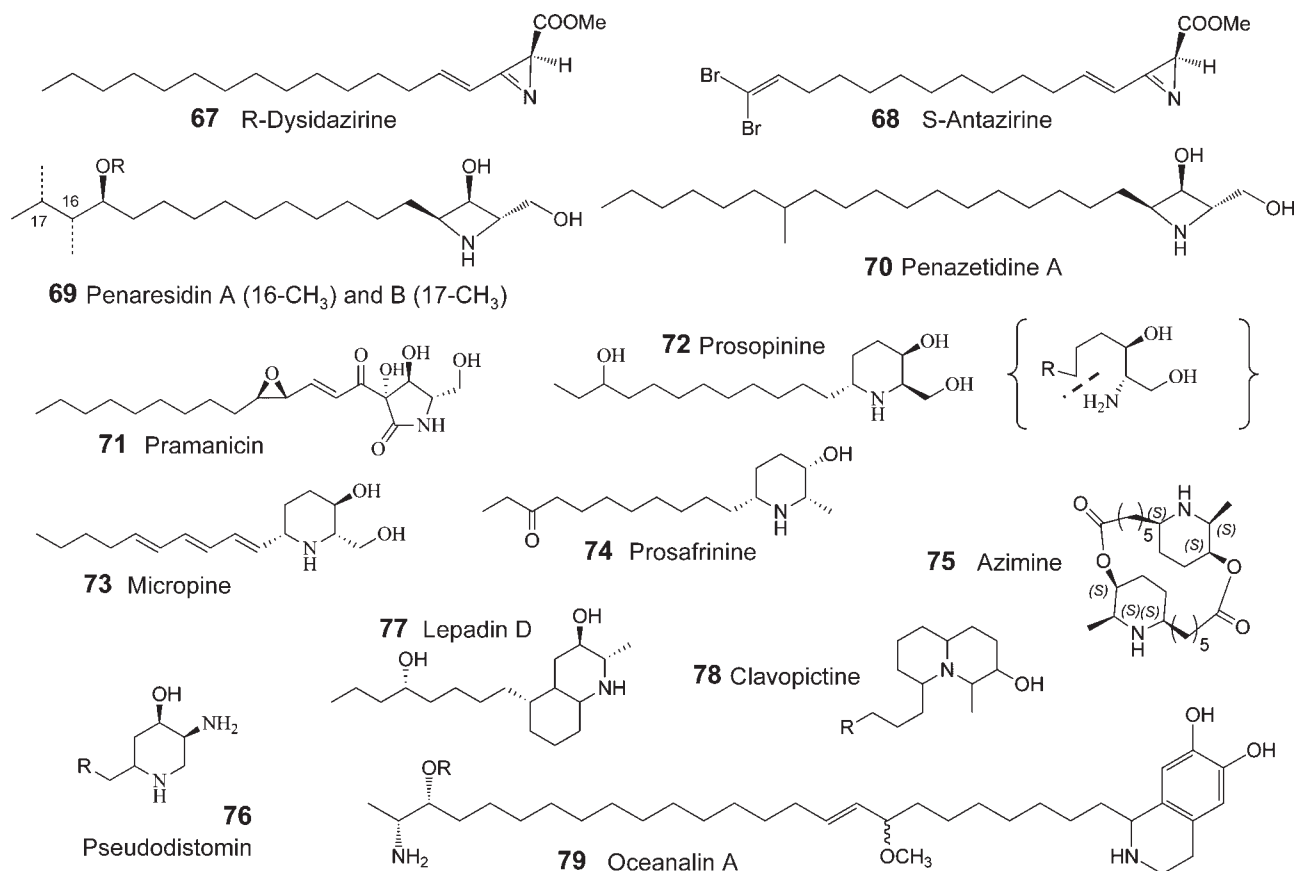
Fig. 7. Sphingoid base-like compounds with heterocyclic rings.

ine by *Stagonospora* species ATCC 74235 that, in addition to having a highly polar, five-member heterocyclic head group, has an aliphatic side chain with both a vinyl ketone and an epoxide. This compound is active against a number of fungi, including *Cryptococcus neoformans*. Pramanicin has also been found to disturb the vasorelaxation of dog carotid artery by selectively acting on the endothelial cells, causing relaxation through the endothelium-dependent nitric oxide pathway to activate endothelial nitric oxide synthase (169), and the epoxide is required for the optimal effects (170). It also activates caspases and induces apoptosis in Jurkat leukemia cells (171).

There are a very large number of six-member ring heterocyclic compounds that appear to be derived from sphingoid bases (e.g., compounds **72–76** in Fig. 7). While these might not be as readily recognizable as sphingoid bases, disconnection of the heterocyclic ring (as displayed in the brackets beside compound **72** in Fig. 7) reveals an acyclic species that is essentially a sphingoid base. Prosopinine **72** and its isomer prosophylline (data not shown), which have been isolated from the spiny shrub *Prosopis*, are antibacterial and anesthetic (172, 173). Micropine **73** (from *Microcos phillippensis*) has a side chain with three conjugated double bonds and has shown antimicrobial activity against several bacteria, including *Pseudomonas auriginosa* and *S. aureus* (172).

Prosafrinine (compound **74** in Fig. 7), which has been isolated from *Prosopis africana* leaves (174), represents a

cyclized 1-deoxy-sphingoid base (by the same retrosynthetic logic shown in the brackets beside **72**). A wide variety of diastereomers and chain length variants of prosafrinine have been found in *Cassia spectablis* (175, 176) and *Cassia leptophylla* (175, 177–179). Azimine (compound **75** in Fig. 7) and carpaine (data not shown) are complex cyclic dimers produced by the plant *Azima tetracantha* (180, 181).

The pseudodistomins (represented by **76** in Fig. 7) are diamine analogs that have been isolated from the sponges *Pseudodistoma kanoko* and *Pseudodistoma amegalarva*, and subspecies in this family differ in the stereochemistry of the amino alcohol and the alkyl chains (147, 150, 182). Some have been found to be cytotoxic against murine lymphoma L1210 cells (151), which might make them interesting antitumor candidates (183), but others cause DNA damage in cell culture (184).

There are also heterocyclic sphingoid base-like compounds that have more than one ring, such as the lepadins (represented by leparin D **77** in Fig. 7) and clavopictines (represented by **78**) from *Prosteceraeus villatus*, *Clavelina lepadiformis*, and *Aplidium tabascum* and pictamine (data not shown) from *Clavelina picta* (174, 185–188). It has been noted that pictamine and the lepadins can also be envisioned to contain an acetylcholine mimetic in their backbone, and this might account for their biological activity blocking nicotinic acetylcholine receptors (174, 185, 186, 188). Oceanalin A (compound **79** in Fig. 7) is an α,ω-bifunctionalized sphingoid base-tetrahydroisoquinoline

from the sponge *Oceanapia* species that in its glycoside form (R = galactose) has in vitro antifungal activity against *C. glabrata* and has been suggested to block sphingolipid biosynthesis by inhibiting ceramide synthase (189).

COMPOUNDS PRODUCED FROM THE REACTION OF SPHINGOID BASES AND SPHINGOLIPIDS UNDER OTHER "PHYSIOLOGIC" CONDITIONS

In addition to the naturally occurring and synthetic compounds described above, there are also numerous conditions that structurally modify sphingoid bases, beginning with the well-known metabolic pathways (acylation, phosphorylation, and head group addition) to which *N*-methylation was recently added (47) (Fig. 1). There are also "lyso" derivatives that are thought to be formed by first biosynthesizing a complex sphingolipid (i.e., with amide-linked fatty acid and head group, such as sphingomyelin) followed by removal of the fatty acid (examples being lysosphingomyelin = sphingosylphosphocholine, and psychosine, which is a monohexosylsphingosine such as galactosylsphingosine).

A very interesting category of highly reactive products, sphingoid base chloramines (represented by compound **80** in **Fig. 8**), are created upon the reaction of the free amine with hypochlorous acid and hypochlorite, which are produced in some biological systems by myeloperoxidase, a heme-containing enzyme that neutrophils use to kill bacteria (190). As shown in the reaction pathway diagram in Fig. 8A, the intermediate chloramine eliminates HCl and undergoes chain cleavage to produce 2-hexadecenal (the same catabolic product that is formed by sphingoid base turnover enzymatically; Fig. 1) and 1-cyanomethanophosphocholine (if the sphingoid base is sphingosylphosphorylcholine, as shown in this example) by the likely mechanism in Fig. 8. Fatty aldehydes are also highly reactive compounds and have been associated with the pathogenesis of Sjögren-Larsson syndrome, an inherited neurocutaneous disorder caused by mutations in the enzyme that catalyzes the oxidation of fatty aldehydes to fatty acids (191). Fatty aldehydes are also encountered as natural components of food (and food additives) (192) and as insect pheromones (193, 194).

The reactions shown in Fig. 8B have long been known [dating back to studies by Herb Carter (36)] to occur during acid hydrolysis of sphingolipids (37). In addition, acyl chain migration can occur in ceramides under acidic conditions, as shown in Fig. 8C (195). Because biochemistry often capitalizes on the intrinsic chemical reactivity of compounds, one can envision how these chemical interconversions might occur in a biological context.
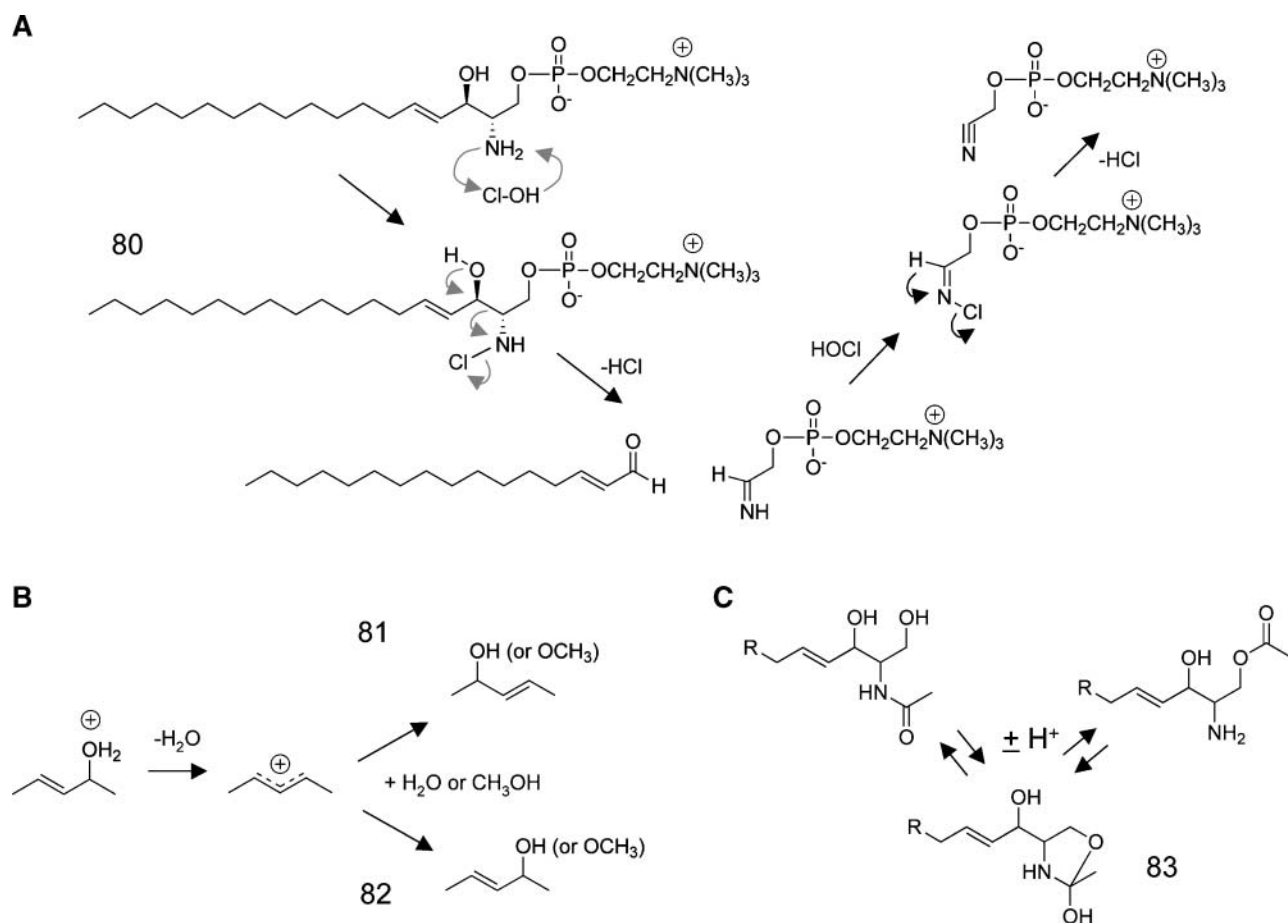

Fig. 8. Common chemical reactions that modify sphingoid bases. A: The formation and decomposition of sphingoid base chloramines due to myeloperoxidase generated reactive chlorination species. B, C: Reactions of sphingoid bases and ceramides under acidic conditions.

Synthetic analogs based on sphingoid bases

Due to the numerous associations between sphingo-lipids and disease, sphingoid bases, sphingoid base-like compounds, and derivatives of such compounds offer promise as therapeutic agents (196, 197), especially as antibacterial (198), antifungal (199), and anticancer (196, 197, 200) drugs. In addition to mimicking endogenous sphingolipids to activate or inhibit cellular targets of pharmaceutical interest, analogs might also be useful as modulators of endogenous sphingolipid metabolism to achieve this goal (201).

Figure 9 summarizes some of the sphingoid bases and analogs that have been developed as potential pharmaceutical leads and/or as tools to study the functions of sphingolipids. Safingol (L-*threo*-sphinganine, **84**) is currently being evaluated in phase I human clinical trials because it has modest host toxicity (202) and displayed antitumor activity in preclinical studies when used in combination with another agent, such as mitomycin C (203) or fenretinide (204, 205). Safingol is acylated by mammalian ceramide synthase(s) (114) and was recently found to undergo substantial N-methylation (to N-methyl, N,N-dimethyl, and N,N,N-trimethyl derivatives) (47), which is interesting because N,N-dimethylsphingosine is cytotoxic for many cancer cell lines (51, 206, 207) (and presumably, the same may be the case for N,N-dimethylsafingol, **85**). The formation of N-methyl derivatives in vivo is also interesting because administration of N,N,N-trimethylsphingosine (86) intra-

venously at the onset of ischemia has been found to reduce myocardial infarct size and improvement in cardiac function (208).

Synthetic 1-deoxy-sphinganines have been developed as potentially useful alternatives to natural sphingoid bases because they cannot be phosphorylated and degraded via sphingosine 1-phosphate lyase (209). One category (which has been given the name "enigmols," represented by compound **87** in Fig. 9) shifts the 1-hydroxyl to carbon 5 to maintain the relative hydrophilicity of the parent sphinganine and thereby facilitate cellular delivery (209). Depending on the stereochemistry, 1-deoxy-sphingoid bases are also less rapidly acylated to 1-deoxy-dihydroceramide analogs (114). It is possible that such analogs affect the same cellular target(s) as 1-deoxy-sphinganine (spisulosine, ES-285, compound **56**).

Other unusual sphingoid bases have been tested as the backbones for ceramide analogs; for example, (2S,3R)-(4E,6E)-2-amino-octadecadiene-1,3-diol (**88** in Fig. 9) coupled with an eight carbon chain length fatty acid has been found to be more cytotoxic than ceramide for MCF-7 cells, which the authors suggest may be due in part to its causing a prolonged elevation of intracellular ceramide (210). A cationic, water-soluble derivative of safingol (L-*threo*-C6-pyridinium-ceramide-bromide; **89** in Fig. 9) has also been shown to inhibit the growth of various human head and neck squamous cell carcinoma cell lines alone or in combination with gemcitabine (211). N-Oleoyl-serinol (**90**) has been uti-
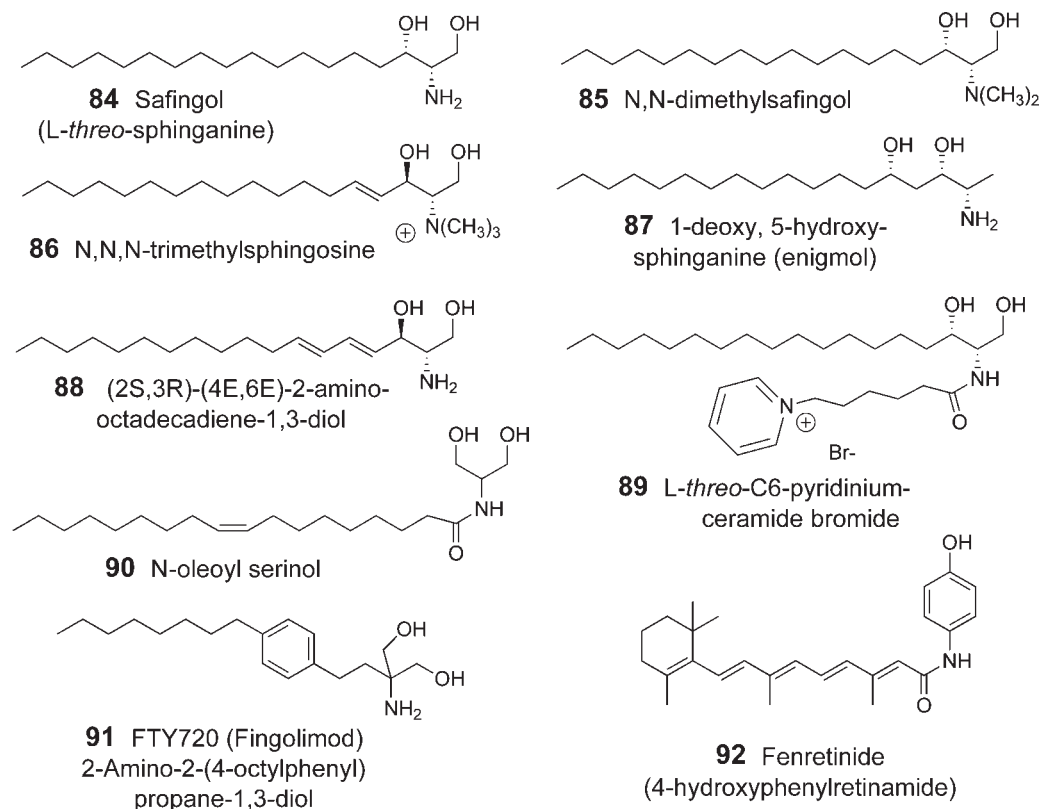

Fig. 9. Examples of compounds of interest as sphingoid base/ceramide analog pharmaceutical leads.

lized to modulate the path of development of embryonic stem cells as well as to suppress the formation of stem cell-derived tumors (teratomas), which is regarded to be a significant obstacle to stem cell therapy (212).

One of the most studied sphingoid base-like compounds is the drug FTY720 (also referred to as "fingolimod"; compound **91** in Fig. 9), which was developed in the course of looking for a less toxic form of the SPT inhibitor ISP-1/myriocin (85, 101, 213). FTY720 does not inhibit this enzyme, but it is phosphorylated by sphingosine kinase to yield an agonist for sphingosine 1-phosphate receptors that also behaves as an antagonist by desensitizing the sphingosine 1-phosphate receptor, resulting in immunosuppression (214). Although FTY720's mechanisms of action are not fully understood, it appears to reduce the number of circulating lymphocytes by inhibiting lymphocyte egress from peripheral lymph nodes, hence, tissue-damaging T-cells cannot recirculate and infiltrate sites of inflammation. FTY720 effectively prevents transplant rejection, is being evaluated in human clinical trials for safety and tolerability in renal transplantation, and has shown promising results in phase II trials for multiple sclerosis (95, 213).

Fenretinide (4-hydroxyphenylretinamide; compound **92**) is another ceramide-like analog that has low toxicity and promising efficacy in some human clinical trials (e.g., risk reduction of second breast cancer in premenopausal women) (215) but not others, such as for advanced renal carcinoma (216) or the prevention of tumor recurrence in patients with transitional cell carcinoma of the bladder (217). Its mechanism of action is thought to involve the induction of de novo sphingolipid biosynthesis (204) and was initially thought to induce tumor cell death via ceramide; however, it has been shown instead to elevate dihydroceramide and autophagic cell death (2).

PERSPECTIVES ON SPHINGOID BASE LIPIDOMICS

This review has given an overview of the amazing biodiversity of sphingoid bases and sphingoid base-like compounds. This complexity presents a large conceptual challenge (i.e., what are the biochemical functions of these biomolecules?) and an equally serious analytical challenge, since one will ultimately need to identify which of these are present in a given biological system, then quantify all of the pertinent subspecies.

In principle, sphingoid bases are relatively easy to analyze by liquid chromatography, electrospray tandem mass spectrometry in positive ion mode (218). However, when studying these compounds in a living organism, it is not sufficient to analyze only the sphingoid bases per se but also all of the potential downstream metabolites, which multiplies the complexity of the analysis. An excellent in-depth review of methods for the analysis of complex (glyco)sphingolipids has been published (63), and methods for more narrow subclasses, such as all of the backbone metabolites and immediate products (such as sphingomyelins, ceramide phosphates, glucosylceramides, etc.) are

also available (63, 218, 219). Nonetheless, much more sophisticated technologies will be needed to analyze a sphingolipidome of this complexity.

"Sphingolipidomic" analysis is becoming increasingly vital for studies of cell signaling to know, for example, the relative amounts of proapoptotic versus antiapoptotic ceramide and sphingosine 1-phosphate (220). In addition, sphingolipids can serve as biomarkers for disease (221, 222) and even as a biological signature, as illustrated by a study of the physiological status and bacterial diversity of estuarine microbial mats (223), which used the presence of sphingoid bases (d18:0, d19:0, and d21:1) and hydroxy fatty acids to predict the presence of organisms in the *Bacteroides* genus, because they are known to have sphingolipids (224).

The biodiversity of the sphingoid bases and sphingoid base-like compounds will continue to amaze, challenge, and amuse scientists for many years to come, just as Thudichum, Carter, and other giants of the early days of sphingolipid research experienced as they gave birth to this field.▊

REFERENCES

1. Thudichum, J. L. W. 1884. A Treatise on the Chemical Constitution of Brain. Bailliere, Tindall, and Cox, London.
2. Zheng, W., J. Kollmeyer, H. Symolon, A. Momin, E. Munter, E. Wang, S. Kelly, J. C. Allegood, Y. Liu, Q. Peng, et al. 2006. Ceramides and other bioactive sphingolipid backbones in health and disease: lipidomic analysis, metabolism and roles in membrane structure, dynamics, signaling and autophagy. *Biochim. Biophys. Acta.* **1758:** 1864–1884.
3. Dickson, R. C. 2008. Thematic review series: sphingolipids. New insights into sphingolipid metabolism and function in budding yeast. *J. Lipid Res.* **49:** 909–921.
4. Spiegel, S., and S. Milstien. 2003. Sphingosine-1-phosphate: an enigmatic signalling lipid. *Nat. Rev. Mol. Cell Biol.* **4:** 397–407.
5. Alvarez, S. E., S. Milstien, and S. Spiegel. 2007. Autocrine and paracrine roles of sphingosine-1-phosphate. *Trends Endocrinol. Metab.* **18:** 300–307.
6. Hannun, Y. A., and L. M. Obeid. 2002. The ceramide-centric universe of lipid-mediated cell regulation: stress encounters of the lipid kind. *J. Biol. Chem.* **277:** 25847–25850.
7. Kitatani, K., J. Idkowiak-Baldys, and Y. A. Hannun. 2008. The sphingolipid salvage pathway in ceramide metabolism and signaling. *Cell. Signal.* **20:** 1010–1018.
8. Merrill, A. H., Jr., M. D. Wang, M. Park, and M. C. Sullards. 2007. (Glyco)sphingolipidology: an amazing challenge and opportunity for systems biology. *Trends Biochem. Sci.* **32:** 457–468.
9. Yu, R. K., M. Yamagisawa, and T. Ariga. 2008. Glycosphingolipid structures. *In* Comprehensive Glycoscience. J. P. Kamerling, editor. Elsevier, Oxford, UK. in press.
10. Karlsson, K. A. 1970. On the chemistry and occurrence of sphingolipid long-chain bases. *Chem. Phys. Lipids.* **5:** 6–43.
11. Karlsson, K. A. 1970. Sphingolipid long chain bases. *Lipids.* **5:** 878–891.
12. Carter, H. E., F. J. Glick, W. P. Norris, and G. E. Phillips. 1947. Biochemistry of the sphingolipids. III. Structure of sphingosine. *J. Biol. Chem.* **170:** 285–294.
13. Carter, H. E., W. J. Haines, W. E. Ledyard, and W. P. Norris. 1947. Biochemistry of the sphingolipids. I. Preparation of sphingolipides from beef brain and spinal cord. *J. Biol. Chem.* **169:** 77–82.
14. Wang, E., W. P. Norred, C. W. Bacon, R. T. Riley, and A. H. Merrill, Jr. 1991. Inhibition of sphingolipid biosynthesis by fumonisins. Implications for diseases associated with *Fusarium moniliforme. J. Biol. Chem.* **266:** 14486–14490.
15. Sweeley, C. C., and E. A. Moscatelli. 1959. Qualitative microanalysis and estimation of sphingolipid bases. *J. Lipid Res.* **1:** 40–47.
16. Chester, M. A. 1998. IUPAC-IUB Joint Commission on Biochem-

ical Nomenclature (JCBN). Nomenclature of glycolipids—recommendations 1997. *Eur. J. Biochem.* **257:** 293–298.

17. Ternes, P., S. Franke, U. Zahringer, P. Sperling, and E. Heinz. 2002. Identification and characterization of a sphingolipid delta 4-desaturase family. *J. Biol. Chem.* **277:** 25512–25518.

18. Omae, F., M. Miyazaki, A. Enomoto, M. Suzuki, Y. Suzuki, and A. Suzuki. 2004. DES2 protein is responsible for phytoceramide biosynthesis in the mouse small intestine. *Biochem. J.* **379:** 687–695.

19. Fahy, E., S. Subramaniam, H. A. Brown, C. K. Glass, A. H. Merrill, Jr., R. C. Murphy, C. R. Raetz, D. W. Russell, Y. Seyama, W. Shaw, et al. 2005. A comprehensive classification system for lipids. *J. Lipid Res.* **46:** 839–861.

20. Merrill, A. H., Jr., and R. D. Williams. 1984. Utilization of different fatty acyl-CoA thioesters by serine palmitoyltransferase from rat brain. *J. Lipid Res.* **25:** 185–188.

21. Haynes, C. A., J. C. Allegood, K. Sims, E. W. Wang, M. C. Sullards, and A. H. Merrill, Jr. 2008. Quantitation of fatty acyl-coenzyme As in mammalian cells by liquid chromatography-electrospray ionization tandem mass spectrometry. *J. Lipid Res.* **49:** 1113–1125.

22. Farwanah, H., B. Pierstorff, C. E. Schmelzer, K. Raith, R. H. Neubert, T. Kolter, and K. Sandhoff. 2007. Separation and mass spectrometric characterization of covalently bound skin ceramides using LC/APCI-MS and Nano-ESI-MS/MS. *J. Chromatogr: B Analyt. Technol. Biomed. Life Sci.* **852:** 562–570.

23. Stewart, M. E., and D. T. Downing. 1995. Free sphingosines of human skin include 6-hydroxysphingosine and unusually long-chain dihydrosphingosines. *J. Invest. Dermatol.* **105:** 613–618.

24. Sonnino, S., and V. Chigorno. 2000. Ganglioside molecular species containing C18- and C20-sphingosine in mammalian nervous tissues and neuronal cell cultures. *Biochim. Biophys. Acta.* **1469:** 63–77.

25. Keranen, A. 1976. Fatty acids and long-chain bases of gangliosides of human gastrointestinal mucosa. *Chem. Phys. Lipids.* **17:** 14–21.

26. Merrill, A. H., Jr., E. Wang, and P. W. Wertz. 1986. Differences in the long chain (sphingoid) base composition of sphingomyelin from rats bearing Morris hepatoma 7777. *Lipids.* **21:** 529–530.

27. Byrdwell, W. C., and R. H. Perry. 2007. Liquid chromatography with dual parallel mass spectrometry and ^{31}P nuclear magnetic resonance spectroscopy for analysis of sphingomyelin and dihydrosphingomyelin. II. Bovine milk sphingolipids. *J. Chromatogr. A.* **1146:** 164–185.

28. Karlsson, A. A., P. Michelsen, and G. Odham. 1998. Molecular species of sphingomyelin: determination by high-performance liquid chromatography/mass spectrometry with electrospray and high-performance liquid chromatography/tandem mass spectrometry with atmospheric pressure chemical ionization. *J. Mass Spectrom.* **33:** 1192–1198.

29. Martin, M. J., S. Martin-Sosa, and P. Hueso. 2001. Bovine milk gangliosides: changes in ceramide moiety with stage of lactation. *Lipids.* **36:** 291–298.

30. Yunoki, K., H. Ishikawa, Y. Fukui, and M. Ohnishi. 2008. Chemical properties of epidermal lipids, especially sphingolipids, of the Antarctic minke whale. *Lipids.* **43:** 151–159.

31. Renkonen, O., and E. L. Hirvisalo. 1969. Structure of plasma sphingadienine. *J. Lipid Res.* **10:** 687–693.

32. Panganamala, R. V., J. C. Geer, and D. G. Cornwell. 1969. Long-chain bases in the sphingolipids of atherosclerotic human aorta. *J. Lipid Res.* **10:** 445–455.

33. Stewart, M. E., and D. T. Downing. 1999. A new 6-hydroxy-4-sphingenine-containing ceramide in human skin. *J. Lipid Res.* **40:** 1434–1439.

34. Chun, J., H. S. Byun, and R. Bittman. 2003. First asymmetric synthesis of 6-hydroxy-4-sphingenine-containing ceramides. Use of chiral propargylic alcohols to prepare a lipid found in human skin. *J. Org. Chem.* **68:** 348–354.

35. Kadowaki, H., E. G. Bremer, J. E. Evans, F. B. Jungalwala, and R. H. McCluer. 1983. Acetonitrile-hydrochloric acid hydrolysis of gangliosides for high performance liquid chromatographic analysis of their long chain bases. *J. Lipid Res.* **24:** 1389–1397.

36. Carter, H. E., O. Nalbandov, and P. A. Tavormina. 1951. Biochemistry of the sphingolipides. VI. The o-methyl ethers of sphingosine. *J. Biol. Chem.* **192:** 197–207.

37. Kisic, A., M. Tsuda, R. J. Kulmacz, W. K. Wilson, and G. J. Schroepfer, Jr. 1995. Sphingolipid bases: a revisitation of the O-methyl derivatives of sphingosine. Isolation and characterization of diacetate derivatives, with revised ^{13}C nuclear magnetic resonance assignments for D-erythro-sphingosine. *J. Lipid Res.* **36:** 787–803.

38. Chun, J., H. S. Byun, G. Arthur, and R. Bittman. 2003. Synthesis and growth inhibitory activity of chiral 5-hydroxy-2-N-acyl-(3E)-

39. Bielawska, A., H. M. Crane, D. Liotta, L. M. Obeid, and Y. A. Hannun. 1993. Selectivity of ceramide-mediated biology. Lack of activity of erythro-dihydroceramide. *J. Biol. Chem.* **268:** 26226–26232.

40. Morrison, W. R. 1973. Long-chain bases in the sphingolipids of bovine milk and kidney, rumen bacteria, rumen protozoa, hay and concentrate. *Biochim. Biophys. Acta.* **316:** 98–107.

41. Yasugi, E., T. Kasama, M. Shibahara, and Y. Seyama. 1990. Composition of long-chain bases in sphingomyelin of the guinea pig harderian gland. *Biochem. Cell Biol.* **68:** 154–160.

42. Yasugi, E., T. Kasama, and Y. Seyama. 1991. Composition of long chain bases in ceramide of the guinea pig harderian gland. *J Biochem.* **110:** 202–206.

43. Nichols, F. C., and K. Rojanasomsith. 2006. Porphyromonas gingivalis lipids and diseased dental tissues. *Oral Microbiol. Immunol.* **21:** 84–92.

44. Sugawara, T., M. Kinoshita, M. Ohnishi, J. Nagata, and M. Saito. 2003. Digestion of maize sphingolipids in rats and uptake of sphingadienine by Caco-2 cells. *J. Nutr.* **133:** 2777–2782.

45. Sugawara, T., M. Kinoshita, M. Ohnishi, T. Tsuzuki, T. Miyazawa, J. Nagata, T. Hirata, and M. Saito. 2004. Efflux of sphingoid bases by P-glycoprotein in human intestinal Caco-2 cells. *Biosci. Biotechnol. Biochem.* **68:** 2541–2546.

46. Igarashi, Y., and S. Hakomori. 1989. Enzymatic synthesis of N,N-dimethyl-sphingosine: demonstration of the sphingosine:N-methyl-transferase in mouse brain. *Biochem. Biophys. Res. Commun.* **164:** 1411–1416.

47. Morales, P. R., D. L. Dillehay, S. J. Moody, D. C. Pallas, S. Pruett, J. C. Allgood, H. Symolon, and A. H. Merrill, Jr. 2007. Safingol toxicology after oral administration to TRAMP mice: demonstration of safingol uptake and metabolism by N-acylation and N-methylation. *Drug Chem. Toxicol.* **30:** 197–216.

48. Merrill, A. H., Jr., S. Nimkar, D. Menaldino, Y. A. Hannun, C. Loomis, R. M. Bell, S. R. Tyagi, J. D. Lambeth, V. L. Stevens, R. Hunter, et al. 1989. Structural requirements for long-chain (sphingoid) base inhibition of protein kinase C in vitro and for the cellular effects of these compounds. *Biochemistry.* **28:** 3138–3145.

49. Melendez, A. J., E. Carlos-Dias, M. Gosink, J. M. Allen, and L. Takacs. 2000. Human sphingosine kinase: molecular cloning, functional characterization and tissue distribution. *Gene.* **251:** 19–26.

50. Kim, H. L., and D. S. Im. 2008. N,N-Dimethyl-D-erythro-sphingosine increases intracellular Ca^{2+} concentration via Na^+-Ca^{2+}-exchanger in HCT116 human colon cancer cells. *Arch. Pharm. Res.* **31:** 54–59.

51. Sweeney, E. A., C. Sakakura, T. Shirahama, A. Masamune, H. Ohta, S. Hakomori, and Y. Igarashi. 1996. Sphingosine and its methylated derivative N,N-dimethylsphingosine (DMS) induce apoptosis in a variety of human cancer cell lines. *Int. J. Cancer.* **66:** 358–366.

52. Wiegandt, H. 1992. Insect glycolipids. *Biochim. Biophys. Acta.* **1123:** 117–126.

53. Fyrst, H., D. R. Herr, G. L. Harris, and J. D. Saba. 2004. Characterization of free endogenous C14 and C16 sphingoid bases from *Drosophila melanogaster*. *J. Lipid Res.* **45:** 54–62.

54. Fyrst, H., X. Zhang, D. R. Herr, H. S. Byun, R. Bittman, V. H. Phan, G. L. Harris, and J. D. Saba. 2008. Identification and characterization by electrospray mass spectrometry of endogenous Drosophila sphingadienes. *J. Lipid Res.* **49:** 597–606.

55. Gerdt, S., R. D. Dennis, G. Borgonie, R. Schnabel, and R. Geyer. 1999. Isolation, characterization and immunolocalization of phosphorylcholine-substituted glycolipids in developmental stages of *Caenorhabditis elegans*. *Eur. J. Biochem.* **266:** 952–963.

56. Chitwood, D. J., W. R. Lusby, M. J. Thompson, J. P. Kochansky, and O. W. Howarth. 1995. The glycosylceramides of the nematode *Caenorhabditis elegans* contain an unusual, branched-chain sphingoid base. *Lipids.* **30:** 567–573.

57. Wuhrer, M., S. Rickhoff, R. D. Dennis, G. Lochnit, P. T. Soboslay, S. Baumeister, and R. Geyer. 2000. Phosphocholine-containing, zwitterionic glycosphingolipids of adult *Onchocerca volvulus* as highly conserved antigenic structures of parasitic nematodes. *Biochem. J.* **348:** 417–423.

58. Lochnit, G., S. Nispel, R. D. Dennis, and R. Geyer. 1998. Structural analysis and immunohistochemical localization of two acidic glycosphingolipids from the porcine, parasitic nematode, *Ascaris suum*. *Glycobiology.* **8:** 891–899.

59. Asai, N., N. Fusetani, and S. Matsunaga. 2001. Sex pheromones of the hair crab *Erimacrus isenbeckii*. II. Synthesis of ceramides. *J. Nat. Prod.* **64:** 1210–1215.

60. Han, G., K. Gable, L. Yan, M. J. Allen, W. H. Wilson, P. Moitra, J. M. Harmon, and T. M. Dunn. 2006. Expression of a novel marine viral single-chain serine palmitoyltransferase and construction of yeast and mammalian single-chain chimera. *J. Biol. Chem.* **281:** 39935–39942.

61. Zhao, R., D. C. Pevear, M. J. Kremer, V. L. Giranda, J. A. Kofron, R. J. Kuhn, and M. G. Rossmann. 1996. Human rhinovirus 3 at 3.0 A resolution. *Structure.* **4:** 1205–1220.

62. Sperling, P., and E. Heinz. 2003. Plant sphingolipids: structural diversity, biosynthesis, first genes and functions. *Biochim. Biophys. Acta.* **1632:** 1–15.

63. Levery, S. B. 2005. Glycosphingolipid structural analysis and glycosphingolipidomics. *Methods Enzymol.* **405:** 300–369.

64. Barreto-Bergter, E., M. R. Pinto, and M. L. Rodrigues. 2004. Structure and biological functions of fungal cerebrosides. *An. Acad. Bras. Cienc.* **76:** 67–84.

65. Rhome, R., T. McQuiston, A. Bielawska, M. Hennig, M. Drago, G. Morace, C. Luberto, and M. Del Poeta. 2007. Biosynthesis and immunogenicity of glucosylceramide in *Cryptococcus neoformans* and other human pathogens. *Eukaryot. Cell.* **6:** 1715–1726.

66. Toledo, M. S., S. B. Levery, E. Suzuki, A. H. Straus, and H. K. Takahashi. 2001. Characterization of cerebrosides from the thermally dimorphic mycopathogen *Histoplasma capsulatum*: expression of 2-hydroxy fatty N-acyl(E)-Delta(3)-unsaturation correlates with the yeast-mycelium phase transition. *Glycobiology.* **11:** 113–124.

67. Qi, J., M. Ojika, and Y. Sakagami. 2001. Neuritogenic cerebrosides from an edible Chinese mushroom. II. Structures of two additional termitomycesphins and activity enhancement of an inactive cerebroside by hydroxylation. *Bioorg. Med. Chem.* **9:** 2171–2177.

68. Irie, A., H. Kubo, and M. Hoshi. 1990. Glucosylceramide having a novel tri-unsaturated long-chain base from the spermatozoa of the starfish, *Asterias amurensis*. *J Biochem.* **107:** 578–586.

69. Ohashi, Y., T. Tanaka, S. Akashi, S. Morimoto, Y. Kishimoto, and Y. Nagai. 2000. Squid nerve sphingomyelin containing an unusual sphingoid base. *J. Lipid Res.* **41:** 1118–1124.

70. Seki, M., and K. Mori. 2001. Synthesis of a prenylated and immunosuppressive marine galactosphingolipid with cyclopropane-containing alkyl chains: (2S,3R,11S,12R,2‴R,5‴Z,11‴S,12‴R)-plakoside A and its (2S,3R,11R,12S,2‴R,5‴Z,11‴R,12‴S) isomer. *Eur. J. Org. Chem.* **2001:** 3797–3809.

71. Carter, G. T., and K. L. Rinehart. 1978. Aplidiasphingosine, an antimicrobial and antitumor terpenoid from an Aplidium sp. (marine tunicate). *J. Am. Chem. Soc.* **100:** 7441–7442.

72. Umemura, T., and K. Mori. 1987. Synthesis of both 2,3-erythro-and 2,3-threo-isomers of aplidiasphingosine, a marine terpenoid. *Agric. Biol. Chem.* **51:** 217–224.

73. Molinski, T. F. 2000. Antifungal compounds from marine organisms. *Current Medicinal Chemistry Anti-Infective Agents.* **3:** 197–220.

74. Kawahara, K., B. Lindner, Y. Isshiki, K. Jakob, Y. A. Knirel, and U. Zahringer. 2001. Structural analysis of a new glycosphingolipid from the lipopolysaccharide-lacking bacterium *Sphingomonas adhaesiva*. *Carbohydr. Res.* **333:** 87–93.

75. Yard, B. A., L. G. Carter, K. A. Johnson, I. M. Overton, M. Dorward, H. Liu, S. A. McMahon, M. Oke, D. Puech, G. J. Barton, et al. 2007. The structure of serine palmitoyltransferase: gateway to sphingolipid biosynthesis. *J. Mol. Biol.* **370:** 870–886.

76. Ikushiro, H., H. Hayashi, and H. Kagamiyama. 2004. Reactions of serine palmitoyltransferase with serine and molecular mechanisms of the actions of serine derivatives as inhibitors. *Biochemistry.* **43:** 1082–1092.

77. Ikushiro, H., M. M. Islam, H. Tojo, and H. Hayashi. 2007. Molecular characterization of membrane-associated soluble serine palmitoyltransferases from *Sphingobacterium multivorum* and *Bdellovibrio stolpii*. *J. Bacteriol.* **189:** 5749–5761.

78. Merrill, A. H., Jr. 2002. De novo sphingolipid biosynthesis: a necessary, but dangerous, pathway. *J. Biol. Chem.* **277:** 25843–25846.

79. Ardail, D., I. Popa, K. Alcantara, A. Pons, J. P. Zanetta, P. Louisot, L. Thomas, and J. Portoukalian. 2001. Occurrence of ceramides and neutral glycolipids with unusual long-chain base composition in purified rat liver mitochondria. *FEBS Lett.* **488:** 160–164.

80. Azuma, H., S. Ijichi, M. Kataoka, A. Masuda, T. Izumi, T. Yoshimoto, and T. Tachibana. 2007. Short-chain 3-ketoceramides, strong apoptosis inducers against human leukemia HL-60 cells. *Bioorg. Med. Chem.* **15:** 2860–2867.

81. MaGee, D. I., J. D. Leach, and T. C. Mallais. 1997. Studies on the synthesis of highly substituted furans: the synthesis of calicogorgins A and C. *Tetrahedron Lett.* **38:** 1289–1292.

82. Ochi, M., K. Yamada, H. Kawakami, A. Tatsukawa, and H. Kotsuki. 1992. Calicogorgins A, B, and C, three bioactive sphinganine derivatives from the gorgonian *Calicogorgia* sp. *Tetrahedron Lett.* **33:** 7531–7534.

83. Lin, Y. P., J. Yan, and M. H. Qiu. 2006. Novel imine from *Hemsleya macrocarpa* var. *clavata*. *Lipids.* **41:** 97–99.

84. Miyake, Y., Y. Kozutsumi, S. Nakamura, T. Fujita, and T. Kawasaki. 1995. Serine palmitoyltransferase is the primary target of a sphingosine-like immunosuppressant, ISP-1/myriocin. *Biochem. Biophys. Res. Commun.* **211:** 396–403.

85. Fujita, T., R. Hirose, M. Yoneta, S. Sasaki, K. Inoue, M. Kiuchi, S. Hirase, K. Chiba, H. Sakamoto, and M. Arita. 1996. Potent immunosuppressants, 2-alkyl-2-aminopropane-1,3-diols. *J. Med. Chem.* **39:** 4451–4459.

86. Hanada, K., M. Nishijima, T. Fujita, and S. Kobayashi. 2000. Specificity of inhibitors of serine palmitoyltransferase (SPT), a key enzyme in sphingolipid biosynthesis, in intact cells: a novel evaluation system using an SPT-defective mammalian cell mutant. *Biochem. Pharmacol.* **59:** 1211–1216.

87. Kluepfel, D., J. Bagli, H. Baker, M. P. Charest, and A. Kudelski. 1972. Myriocin, a new antifungal antibiotic from *Myriococcum albomyces*. *J. Antibiot. (Tokyo).* **25:** 109–115.

88. Aragozzini, F., P. L. Manachini, R. Craveri, B. Rindone, and C. Scolastico. 1972. Structure of thermozymocidin. *Experientia.* **28:** 881–882.

89. Yamaji-Hasegawa, A., A. Takahashi, Y. Tetsuka, Y. Senoh, and T. Kobayashi. 2005. Fungal metabolite sulfamisterin suppresses sphingolipid synthesis through inhibition of serine palmitoyltransferase. *Biochemistry.* **44:** 268–277.

90. Zweerink, M. M., A. M. Edison, G. B. Wells, W. Pinto, and R. L. Lester. 1992. Characterization of a novel, potent, and specific inhibitor of serine palmitoyltransferase. *J. Biol. Chem.* **267:** 25032–25038.

91. Kobayashi, S., T. Hayashi, S. Iwamoto, T. Furuta, and M. Matsumura. 1996. Asymmetric synthesis of antifungal agents sphingofungins using catalytic asymmetric aldol reactions. *Synlett.* **1996:** 672–674.

92. Horn, W. S., J. L. Smith, G. F. Bills, S. L. Raghoobar, G. L. Helms, M. B. Kurtz, J. A. Marrinan, B. R. Frommer, R. A. Thornton, and S. M. Mandala. 1992. Sphingofungins E and F: novel serinepalmitoyl transferase inhibitors from *Paecilomyces variotii*. *J. Antibiot. (Tokyo).* **45:** 1692–1696.

93. Berova, N., J. Breinholt, G. W. Jensen, A. Kjaer, L. C. Lo, K. Nakanishi, R. I. Nielsen, C. E. Olsen, C. Pedersen, and C. E. Stidsen. 1994. Malonofungin: an antifungal animomalonic acid from *Phaeramularia fusimaculans*. *Acta Chem. Scand.* **48:** 240–251.

94. Mukhopadhyay, T., K. Roy, L. Coutinho, R. H. Rupp, B. N. Ganguli, and H. W. Fehlhaber. 1987. Fumifungin, a new antifungal antibiotic from *Aspergillus fumigatus* Fresenius 1863. *J. Antibiot. (Tokyo).* **40:** 1050–1052.

95. Napoli, K. L. 2000. The FTY720 story. *Ther. Drug Monit.* **22:** 47–51.

96. Mandala, S. M., B. R. Frommer, R. A. Thornton, M. B. Kurtz, N. M. Young, M. A. Cabello, O. Genilloud, J. M. Liesch, J. L. Smith, and W. S. Horn. 1994. Inhibition of serine palmitoyl-transferase activity by lipoxamycin. *J. Antibiot. (Tokyo).* **47:** 376–379.

97. Mandala, S. M., R. A. Thornton, B. R. Frommer, S. Dreikorn, and M. B. Kurtz. 1997. Viridiofungins, novel inhibitors of sphingolipid synthesis. *J. Antibiot. (Tokyo).* **50:** 339–343.

98. Petrache, I., V. Natarajan, L. Zhen, T. R. Medler, A. T. Richter, C. Cho, W. C. Hubbard, E. V. Berdyshev, and R. M. Tuder. 2005. Ceramide upregulation causes pulmonary cell apoptosis and emphysema-like disease in mice. *Nat. Med.* **11:** 491–498.

99. Amemiya, F., S. Maekawa, Y. Itakura, A. Kanayama, A. Matsui, S. Takano, T. Yamaguchi, J. Itakura, T. Kitamura, T. Inoue, et al. 2008. Targeting lipid metabolism in the treatment of hepatitis C virus infection. *J. Infect. Dis.* **197:** 361–370.

100. Umehara, T., M. Sudoh, F. Yasui, C. Matsuda, Y. Hayashi, K. Chayama, and M. Kohara. 2006. Serine palmitoyltransferase inhibitor suppresses HCV replication in a mouse model. *Biochem. Biophys. Res. Commun.* **346:** 67–73.

101. Osuchowski, M. F., V. J. Johnson, Q. He, and R. P. Sharma. 2004. Myriocin, a serine palmitoyltransferase inhibitor, alters regional brain neurotransmitter levels without concurrent inhibition of the brain sphingolipid biosynthesis in mice. *Toxicol. Lett.* **147:** 87–94.

102. Tabas, I. 2004. Sphingolipids and atherosclerosis: a mechanistic connection? A therapeutic opportunity? *Circulation.* **110:** 3400–3401.

103. Hojjati, M. R., Z. Li, H. Zhou, S. Tang, C. Huan, E. Ooi, S. Lu, and X-C. Jiang. 2005. Effect of myriocin on plasma sphingolipid me-

tabolism and atherosclerosis in apoE-deficient mice. *J. Biol. Chem.* **280:** 10284–10289.

104. Glaros, E. N., W. S. Kim, C. M. Quinn, W. Jessup, K. A. Rye, and B. Garner. 2008. Myriocin slows the progression of established atherosclerotic lesions in apolipoprotein E gene knockout mice. *J. Lipid Res.* **49:** 324–331.

105. Riley, R. T., and R. D. Plattner. 2000. Fermentation, partial purification, and use of serine palmitoyltransferase inhibitors from *Isaria* (=*Cordyceps*) *sinclairii*. *Methods Enzymol.* **311:** 348–361.

106. Pewzner-Jung, Y., S. Ben-Dor, and A. H. Futerman. 2006. When do Lasses (longevity assurance genes) become CerS (ceramide synthases)? Insights into the regulation of ceramide synthesis. *J. Biol. Chem.* **281:** 25001–25005.

107. Lahiri, S., H. Lee, J. Mesicek, Z. Fuks, A. Haimovitz-Friedman, R. N. Kolesnick, and A. H. Futerman. 2007. Kinetic characterization of mammalian ceramide synthases: determination of K(m) values towards sphinganine. *FEBS Lett.* **581:** 5289–5294.

108. Abbas, H. K., S. O. Duke, A. H. Merrill, Jr., E. Wang, and W. T. Shier. 1998. Phytotoxicity of australifungin, AAL-toxins and fumonisin B$_1$ to *Lemna pausicostata*. *Phytochemistry.* **47:** 1509–1514.

109. Merrill, A. H., Jr., M. C. Sullards, E. Wang, K. A. Voss, and R. T. Riley. 2001. Sphingolipid metabolism: roles in signal transduction and disruption by fumonisins. *Environ. Health Perspect.* **109 (Suppl. 2):** 283–289.

110. Riley, R. T., E. Enongene, K. A. Voss, W. P. Norred, F. I. Meredith, R. P. Sharma, J. Spitsbergen, D. E. Williams, D. B. Carlson, and A. H. Merrill, Jr. 2001. Sphingolipid perturbations as mechanisms for fumonisin carcinogenesis. *Environ. Health Perspect.* **109 (Suppl. 2):** 301–308.

111. Trucksess, M. W., and P. M. Scott. 2008. Mycotoxins in botanicals and dried fruits: a review. *Food Addit. Contam.* **25:** 181–192.

112. Murphy, P. A., S. Hendrich, E. C. Hopmans, C. C. Hauck, Z. Lu, G. Buseman, and G. Munkvold. 1996. Effect of processing on fumonisin content of corn. *Adv. Exp. Med. Biol.* **392:** 323–334.

113. Palencia, E., O. Torres, W. Hagler, F. I. Meredith, L. D. Williams, and R. T. Riley. 2003. Total fumonisins are reduced in tortillas using the traditional nixtamalization method of Mayan communities. *J. Nutr.* **133:** 3200–3203.

114. Humpf, H. U., E. M. Schmelz, F. I. Meredith, H. Vesper, T. R. Vales, E. Wang, D. S. Menaldino, D. C. Liotta, and A. H. Merrill, Jr. 1998. Acylation of naturally occurring and synthetic 1-deoxysphinganines by ceramide synthase. Formation of N-palmitoyl-aminopentol produces a toxic metabolite of hydrolyzed fumonisin, AP1, and a new category of ceramide synthase inhibitor. *J. Biol. Chem.* **273:** 19060–19064.

115. Seiferlein, M., H. U. Humpf, K. A. Voss, M. C. Sullards, J. C. Allegood, E. Wang, and A. H. Merrill, Jr. 2007. Hydrolyzed fumonisins HFB1 and HFB2 are acylated in vitro and in vivo by ceramide synthase to form cytotoxic N-acyl-metabolites. *Mol. Nutr. Food Res.* **51:** 1120–1130.

116. Abou-Karam, M., H. K. Abbas, and W. T. Shier. 2004. N-Fatty acylation of hydrolyzed fumonisin B1, but not of intact fumonisin B1, strongly enhances in vitro mammalian toxicity. *J. Toxicol. Toxin Rev.* **23:** 123–151.

117. Marasas, W. F. 2001. Discovery and occurrence of the fumonisins: a historical perspective. *Environ. Health Perspect.* **109 (Suppl. 2):** 239–243.

118. Gelderblom, W. C., K. Jaskiewicz, W. F. Marasas, P. G. Thiel, R. M. Horak, R. Vleggaar, and N. P. Kriek. 1988. Fumonisins—novel mycotoxins with cancer-promoting activity produced by *Fusarium moniliforme*. *Appl. Environ. Microbiol.* **54:** 1806–1811.

119. Kolter, T., and K. Sandhoff. 2006. Sphingolipid metabolism diseases. *Biochim. Biophys. Acta.* **1758:** 2057–2079.

120. Riley, R. T., K. A. Voss, W. P. Norred, R. P. Sharma, E. Wang, and A. H. Merrill. 1998. Fumonisins: mechanism of mycotoxicity. *Rev. Med. Vet. (Toulouse).* **149:** 617–626.

121. Stevens, V. L., and J. Tang. 1997. Fumonisin B1-induced sphingolipid depletion inhibits vitamin uptake via the glycosylphosphatidylinositol-anchored folate receptor. *J. Biol. Chem.* **272:** 18020–18025.

122. Marasas, W. F., R. T. Riley, K. A. Hendricks, V. L. Stevens, T. W. Sadler, J. Gelineau-van Waes, S. A. Missmer, J. Cabrera, O. Torres, W. C. Gelderblom, et al. 2004. Fumonisins disrupt sphingolipid metabolism, folate transport, and neural tube development in embryo culture and in vivo: a potential risk factor for human neural tube defects among populations consuming fumonisin-contaminated maize. *J. Nutr.* **134:** 711–716.

123. Gelineau-van Waes, J., L. Starr, J. Maddox, F. Aleman, K. A. Voss, J. Wilberding, and R. T. Riley. 2005. Maternal fumonisin exposure and risk for neural tube defects: mechanisms in an in vivo mouse model. *Birth Defects Res. A Clin. Mol. Teratol.* **73:** 487–497.

124. Sadler, T. W., A. H. Merrill, V. L. Stevens, M. C. Sullards, E. Wang, and P. Wang. 2002. Prevention of fumonisin B1-induced neural tube defects by folic acid. *Teratology.* **66:** 169–176.

125. Missmer, S. A., L. Suarez, M. Felkner, E. Wang, A. H. Merrill, Jr., K. J. Rothman, and K. A. Hendricks. 2006. Exposure to fumonisins and the occurrence of neural tube defects along the Texas-Mexico border. *Environ. Health Perspect.* **114:** 237–241.

126. Winter, C. K., D. G. Gilchrist, M. B. Dickman, and C. Jones. 1996. Chemistry and biological activity of AAL toxins. *Adv. Exp. Med. Biol.* **392:** 307–316.

127. Spassieva, S. D., J. E. Markham, and J. Hille. 2002. The plant disease resistance gene *Asc-1* prevents disruption of sphingolipid metabolism during AAL-toxin-induced programmed cell death. *Plant J.* **32:** 561–572.

128. Uhlig, S., D. Petersen, A. Flaoyen, and A. Wilkins. 2005. 2-Amino-14,16-dimethyloctadecan-3-ol, a new sphingosine analogue toxin in the fungal genus *Fusarium*. *Toxicon.* **46:** 513–522.

129. Wang, W., C. Jones, J. Ciacci-Zanella, T. Holt, D. G. Gilchrist, and M. B. Dickman. 1996. Fumonisins and *Alternaria alternata lycopersici* toxins: sphinganine analog mycotoxins induce apoptosis in monkey kidney cells. *Proc. Natl. Acad. Sci. USA.* **93:** 3461–3465.

130. Shier, W. T., and A. C. Shier. 2000. Sphingosine- and ceramide-analog toxins—an update. *J. Toxicol. Toxin Rev.* **19:** 189–246.

131. Du, L., X. Zhu, R. Gerber, J. Huffman, L. Lou, J. Jorgenson, F. Yu, K. Zaleta-Rivera, and Q. Wang. 2008. Biosynthesis of sphinganine-analog mycotoxins. *J. Ind. Microbiol. Biotechnol.* **35:** 455–464.

132. Mandala, S. M., and G. H. Harris. 2000. Isolation and characterization of novel inhibitors of sphingolipid synthesis: australifungin, viridiofungins, rustmicin, and khafrefungin. *Methods Enzymol.* **311:** 335–348.

133. Mandala, S. M., R. A. Thornton, J. Milligan, M. Rosenbach, M. Garcia-Calvo, H. G. Bull, G. Harris, G. K. Abruzzo, A. M. Flattery, C. J. Gill, et al. 1998. Rustmicin, a potent antifungal agent, inhibits sphingolipid synthesis at inositol phosphoceramide synthase. *J. Biol. Chem.* **273:** 14942–14949.

134. Nara, F., M. Tanaka, T. Hosoya, K. Suzuki-Konagai, and T. Ogita. 1999. Scyphostatin, a neutral sphingomyelinase inhibitor from a discomycete, *Trichopeziza mollissima*: taxonomy of the producing organism, fermentation, isolation, and physico-chemical properties. *J. Antibiot. (Tokyo).* **52:** 525–530.

135. Cuadros, R., E. M. de Garcini, F. Wandosell, G. Faircloth, J. M. Fernandez-Sousa, and J. Avila. 2000. The marine compound spisulosine, an inhibitor of cell proliferation, promotes the disassembly of actin stress fibers. *Cancer Lett.* **152:** 23–29.

136. Jimeno, J. M. 2002. A clinical armamentarium of marine-derived anti-cancer compounds. *Anticancer Drugs.* **13 (Suppl. 1):** 15–19.

137. Sanchez, A. M., S. Malagarie-Cazenave, N. Olea, D. Vara, C. Cuevas, and I. Diaz-Laviada. 2008. Spisulosine (ES-285) induces prostate tumor PC-3 and LNCaP cell death by de novo synthesis of ceramide and PKCzeta activation. *Eur. J. Pharmacol.* **584:** 237–245.

138. Salcedo, M., C. Cuevas, J. L. Alonso, G. Otero, G. Faircloth, J. M. Fernandez-Sousa, J. Avila, and F. Wandosell. 2007. The marine sphingolipid-derived compound ES 285 triggers an atypical cell death pathway. *Apoptosis.* **12:** 395–409.

139. Desai, K., M. C. Sullards, J. Allegood, E. Wang, E. M. Schmelz, M. Hartl, H-U. Humpf, D. C. Liotta, Q. Peng, and A. H. Merrill, Jr. 2002. Fumonisins and fumonisin analogs as inhibitors of ceramide synthase and inducers of apoptosis. *Biochim. Biophys. Acta.* **1585:** 188–192.

140. Ichihashi, M., and K. Mori. 2003. Determination of the absolute configuration of (+)-xestoaminol C [(2S, 3R)-2-amino-3-tetradecanol], a metabolite of Fiji sponge, *Xestospongia* sp., by the synthesis of its N,O-diacetyl derivative. *Biosci. Biotechnol. Biochem.* **67:** 329–333.

141. Garrido, L., E. Zubia, M. J. Ortega, S. Naranjo, and J. Salva. 2001. Obscuraminols, new unsaturated amino alcohols from the tunicate *Pseudodistoma obscurum*: structure and absolute configuration. *Tetrahedron.* **57:** 4579–4588.

142. Jares-Erijman, E. A., C. P. Bapat, A. Lithgow-Bertelloni, K. L. Rinehart, and R. Sakai. 1993. Crucigasterins, new polyunsaturated amino alcohols from the Mediterranean tunicate *Pseudodistoma crucigaster*. *J. Org. Chem.* **58:** 5732–5737.

143. Sata, N. U., and N. Fusetani. 2000. Amaminols A and B, new bicyclic amino alcohols from an unidentified tunicate of the family Polyclinidae. *Tetrahedron Lett.* **41:** 489–492.

144. Nicholas, G. M., R. Li, J. B. MacMillan, and T. F. Molinski. 2002. Antifungal activity of bifunctional sphingolipids. Intramolecular

synergism within long-chain α,ω-bis-aminoalcohols. *Bioorg. Med. Chem. Lett.* **12:** 2159–2162.

145. Zhou, B-N., M. P. Mattern, R. K. Johnson, and D. G. I. Kingston. 2001. Structure and stereochemistry of a novel bioactive sphingolipid from a *Calyx* sp. *Tetrahedron.* **57:** 9549–9554.

146. Nicholas, G. M., T. W. Hong, T. F. Molinski, M. L. Lerch, M. T. Cancilla, and C. B. Lebrilla. 1999. Oceanapiside, an antifungal bis-alpha,omega-amino alcohol glycoside from the marine sponge *Oceanapia phillipensis. J. Nat. Prod.* **62:** 1678–1681.

147. Nicholas, G. M., and T. F. Molinski. 2000. Enantiodivergent biosynthesis of the dimeric sphingolipid oceanapiside from the marine sponge *Oceanapia phillipensis.* Determination of remote stereochemistry. *J. Am. Chem. Soc.* **122:** 4011–4019.

148. Makarieva, T. N., P. S. Dmitrenok, A. M. Zakharenko, V. A. Denisenko, A. G. Guzii, R. Li, C. K. Skepper, T. F. Molinski, and V. A. Stonik. 2007. Rhizochalins C and D from the sponge *Rhizochalina incrustata.* A rare threo-sphingolipid and a facile method for determination of the carbonyl position in alpha,omega-bifunctionalized keto-sphingolipids. *J. Nat. Prod.* **70:** 1991–1998.

149. Nicholas, G. M., L. L. Eckman, G. L. Newton, R. C. Fahey, S. Ray, and C. A. Bewley. 2003. Inhibition and kinetics of *Mycobacterium tuberculosis* and *Mycobacterium smegmatis* mycothiol-S-conjugate amidase by natural product inhibitors. *Bioorg. Med. Chem.* **11:** 601–608.

150. Crews, P., D. P. Clark, and K. Tenney. 2003. Variation in the alkaloids among Indo-Pacific *Leucetta* sponges. *J. Nat. Prod.* **66:** 177–182.

151. Kobayashi, J. I., K. Naitoh, Y. Doi, K. Deki, and M. Ishibashi. 1995. Pseudodistomin C, a new piperidine alkaloid with unusual absolute configuration from the Okinawan tunicate *Pseudodistoma kanoko. J. Org. Chem.* **60:** 6941–6945.

152. Willis, R. H., and D. J. de Vries. 1997. BRS1, A C30 bis-amino, bis-hydroxy polyunsaturated lipid from an Australian calcareous sponge that inhibits protein kinase C. *Toxicon.* **35:** 1125–1129.

153. Makarieva, T. N., A. G. Guzii, V. A. Denisenko, P. S. Dmitrenok, E. A. Santalova, E. V. Pokanevich, T. F. Molinski, and V. A. Stonik. 2005. Rhizochalin A, a novel two-headed sphingolipid from the sponge *Rhizochalina incrustata. J. Nat. Prod.* **68:** 255–257.

154. Godchaux, W., 3rd, and E. R. Leadbetter. 1984. Sulfonolipids of gliding bacteria. Structure of the N-acylaminosulfonates. *J. Biol. Chem.* **259:** 2982–2990.

155. Kamiyama, T., T. Umino, Y. Itezono, Y. Nakamura, T. Satoh, and K. Yokose. 1995. Sulfobacins A and B, novel von Willebrand factor receptor antagonists. II. Structural elucidation. *J. Antibiot. (Tokyo).* **48:** 929–936.

156. White, R. H. 1984. Biosynthesis of the sulfonolipid 2-amino-3-hydroxy-15-methylhexadecane-1-sulfonic acid in the gliding bacterium *Cytophaga johnsonae. J. Bacteriol.* **159:** 42–46.

157. Abbanat, D. R., W. Godchaux, 3rd, G. Polychroniou, and E. R. Leadbetter. 1985. Biosynthesis of a sulfonolipid in gliding bacteria. *Biochem. Biophys. Res. Commun.* **130:** 873–878.

158. Gilmore, D. F., W. Godchaux, 3rd, and E. R. Leadbetter. 1989. Cysteine is not an obligatory intermediate in the biosynthesis of cysteate by *Cytophaga johnsonae. Biochem. Biophys. Res. Commun.* **160:** 535–539.

159. Kamiyama, T., T. Umino, T. Satoh, S. Sawairi, M. Shirane, S. Ohshima, and K. Yokose. 1995. Sulfobacins A and B, novel von Willebrand factor receptor antagonists. I. Production, isolation, characterization and biological activities. *J. Antibiot. (Tokyo).* **48:** 924–928.

160. Corcelli, A., V. M. Lattanzio, G. Mascolo, F. Babudri, A. Oren, and M. Kates. 2004. Novel sulfonolipid in the extremely halophilic bacterium *Salinibacter ruber. Appl. Environ. Microbiol.* **70:** 6678–6685.

161. Molinski, T. F., and C. M. Ireland. 1988. Dysidazirine, a cytotoxic azacyclopropene from the marine sponge *Dysidea fragilis. J. Org. Chem.* **53:** 2103–2105.

162. Salmon, C. E., D. H. Williams, and D. J. Faulkner. 1995. New azacyclopropene derivatives from *Dysidea fragilis* collected in Pohnpei. *J. Nat. Prod.* **58:** 1463–1466.

163. Kobayashi, J. I., M. Tsuda, J.-F. Cheng, M. Ishibashi, H. Takikawa, and K. Mori. 1996. Absolute stereochemistry of penaresidins A and B. *Tetrahedron Lett.* **37:** 6775–6776.

164. Kobayashi, J. I., J.-F. Cheng, M. Ishibashi, M. R. Wälchli, S. Yamamura, and Y. Ohizumi. 1991. Penaresidin A and B, two novel azetidine alkaloids with potent actomyosin ATPase-activating activity from the Okinawan marine sponge *Penares* sp. *J. Chem. Soc. Perkin Trans. 1.* 1135–1137.

165. Ohshita, K., H. Ishiyama, Y. Takahashi, J. Ito, Y. Mikami, and J. Kobayashi. 2007. Synthesis of penaresidin derivatives and its biological activity. *Bioorg. Med. Chem.* **15:** 4910–4916.

166. Alvi, K. A., M. Jaspars, and P. Crews. 1994. Penazetidine A, an alkaloid inhibitor of protein kinase C. *Bioorg. Med. Chem. Lett.* **4:** 2447–2450.

167. Li, Z. M., B. Wang, F. G. Tao, and G. Q. Lin. 2004. Studies on the synthesis of penazetidine A, an alkaloid inhibitor of protein kinase C. *Chinese Chemical Letters.* **15:** 138–140.

168. Harrison, P. H. M., D. W. Hughs, and R. W. Riddoch. 1998. The biosynthesis of pramanicin: origin of the carbon skeleton. *Chem. Commun.* **1998:** 273–274.

169. Kwan, C-Y., P. H. M. Harrison, P. A. Duspara, and E. E. Daniel. 2001. Vasorelaxant effects of pramanicin, an anti-fungal agent: selective action on endothelial cells. *Jpn. J. Pharmacol.* **85:** 234–240.

170. Kwan, C. Y., W. B. Zhang, J. Miller, P. H. Harrison, S. Kassan, and D. Liscombe. 2003. The epoxy group of pramanicin is required for the optimal endothelium-dependent relaxation of rat aorta. *J. Pharmacol. Sci.* **92:** 203–208.

171. Kutuk, O., A. Pedrech, P. Harrison, and H. Basaga. 2005. Pramanicin induces apoptosis in Jurkat leukemia cells: a role for JNK, p38 and caspase activation. *Apoptosis.* **10:** 597–609.

172. Aguinaldo, A. M., and R. W. Read. 1990. A major piperidine alkaloid from *Microcos philippinensis. Phytochemistry.* **29:** 2309–2313.

173. Cook, G. R., L. G. Beholz, and J. R. Stille. 1994. Aza-annulation as a route to hydroxylated alkaloid lipids. The synthesis of (+)-prosopinine. *Tetrahedron Lett.* **35:** 1669–1672.

174. Tsuneki, H., Y. You, N. Toyooka, T. Sasaoka, H. Nemoto, J. A. Dani, and I. Kimura. 2005. Marine alkaloids (−)-pictamine and (−)-lepadin B block neuronal nicotinic acetylcholine receptors. *Biol. Pharm. Bull.* **28:** 611–614.

175. Bolzani, V. D. S., A. A. L. Gunatilaka, and D. G. I. Kingston. 1995. Bioactive and other piperidine alkaloids from *Cassia leptophylla. Tetrahedron.* **51:** 5929–5934.

176. Vegas, C., Jr., V. D. S. Bolzani, M. Furlan, E. J. Barreiro, M. C. M. Young, D. Tomazela, and M. N. Eberlin. 2004. Further bioactive piperidine alkaloids from the flowers and green fruits of *Cassia spectabilis. J. Nat. Prod.* **67:** 908–910.

177. Sansores-Perasa, P., M. Rosado-Vallado, W. Brito-Loeza, G. J. Mena-Rejon, and L. Quijano. 2000. Cassine, an antimicrobial alkaloid from *Senna racemosa. Fitoterpia.* **71:** 690–692.

178. Makabe, H., L. K. Kong, and M. Hirota. 2003. Total synthesis of (−)-cassine. *Org. Lett.* **5:** 27–29.

179. Christofidis, I., A. Welter, and J. Jadot. 1977. Spectalinine and iso-6-carnavaline, two unprecedented piperidine alkaloids from the seeds of *Cassia spectabilis. Tetrahedron.* **33:** 3005–3006.

180. Rall, G. J. H., T. M. Smalberger, and H. L. de Waal. 1967. Dimeric piperidine alkaloids from *Azima tetracantha* Lam.: azimine, azcarpine and carpaine. *Tetrahedron Lett.* **36:** 3465–3469.

181. Randl, S., and S. Blechert. 2004. Concise total synthesis of (+)-carpamic acid. *Tetrahedron Lett.* **45:** 1167–1169.

182. Jayatilake, G. S., B. J. Baker, and J. B. McClintock. 1997. Rhapsamine, a cytotoxin from the Antarctic sponge *Leucetta leptorhapsis. Tetrahedron Lett.* **38:** 7505–7510.

183. Ishibashi, M., Y. Ohizumi, T. Sasaki, H. Nakamura, Y. Hirata, and J. i. Kobayashi. 1987. Pseudodistomins A and B, novel antineoplastic piperidine alkaloids with calmodulin antagonistic activity from the Okinawan tunicate *Pseudodistoma kanoko. J. Org. Chem.* **52:** 450–453.

184. Freyer, A. J., A. D. Patil, L. Killmer, N. Troupe, M. Mentzer, B. Carte, L. Faucette, and R. K. Johnson. 1997. Three new pseudodistomins, piperidine alkaloids from the ascidian *Pseudodistoma megalarva. J. Nat. Prod.* **60:** 986–990.

185. Davis, R. A., A. R. Carroll, and R. J. Quinn. 2002. Lepadins F-H, new *cis*-decahydroquinoline alkaloids from the Australian ascidian *Aplidium tabascum. J. Nat. Prod.* **65:** 454–457.

186. Steffan, B. 1991. Lepadin A, a decahydroquinoline alkaloid from the tunicate *Clavelina lepadiformis. Tetrahedron.* **47:** 8729–8732.

187. Toyooka, N. 2001. Synthesis and its application to the synthesis of biologically active natural products of new and versatile chiral building blocks. *Yakugaku Zasshi.* **121:** 467–479.

188. Kong, F., and D. J. Faulkner. 1991. Pictamine, a quinolizidine alkaloid from the tunicate *Clavelina picta. Tetrahedron Lett.* **32:** 3667–3668.

189. Makarieva, T. N., V. A. Denisenko, P. S. Dmitrenok, A. G. Guzii, E. A. Santalova, V. A. Stonik, J. B. Macmillan, and T. F. Molinski. 2005. Oceanalin A, a hybrid alpha,omega-bifunctionalized sphingoid tetrahydroisoquinoline beta-glycoside from the marine sponge *Oceanapia* sp. *Org. Lett.* **7:** 2897–2900.

190. Brahmbhatt, V. V., F. F. Hsu, J. L. Kao, E. C. Frank, and D. A. Ford.

2007. Novel carbonyl and nitrile products from reactive chlorinating species attack of lysosphingolipid. *Chem. Phys. Lipids.* **145:** 72–84.

191. Rizzo, W. B. 2007. Sjogren-Larsson syndrome: molecular genetics and biochemical pathogenesis of fatty aldehyde dehydrogenase deficiency. *Mol. Genet. Metab.* **90:** 1–9.

192. Dannenberger, D., S. Lorenz, G. Nuernberg, N. Scollan, K. Ender, and K. Nuernberg. 2006. Analysis of fatty aldehyde composition, including 12-methyltridecanal, in plasmalogens from longissimus muscle of concentrate- and pasture-fed bulls. *J. Agric. Food Chem.* **54:** 182–188.

193. Kalinova, B., M. Hoskovec, I. Liblikas, C. R. Unelius, and B. S. Hansson. 2001. Detection of sex pheromone components in *Manduca sexta* (L.). *Chem. Senses.* **26:** 1175–1186.

194. Linn, C. E., Jr., M. J. Domingue, C. J. Musto, T. C. Baker, and W. L. Roelofs. 2007. Support for (Z)-11-hexadecanal as a pheromone antagonist in *Ostrinia nubilalis*: flight tunnel and single sensillum studies with a New York population. *J. Chem. Ecol.* **33:** 909–921.

195. Van Overloop, H., G. Van der Hoeven, and P. P. Van Veldhoven. 2005. N-Acyl migration in ceramides. *J. Lipid Res.* **46:** 812–816.

196. Fox, T. E., C. M. Finnegan, R. Blumenthal, and M. Kester. 2006. The clinical potential of sphingolipid-based therapeutics. *Cell. Mol. Life Sci.* **63:** 1017–1023.

197. Zeidan, Y. H., and Y. A. Hannun. 2007. Translational aspects of sphingolipid metabolism. *Trends Mol. Med.* **13:** 327–336.

198. McQuiston, T. J., C. Haller, and M. Del Poeta. 2006. Sphingolipids as targets for microbial infections. *Mini Rev. Med. Chem.* **6:** 671–680.

199. Thevissen, K., I. E. Francois, A. M. Aerts, and B. P. Cammue. 2005. Fungal sphingolipids as targets for the development of selective antifungal therapeutics. *Curr. Drug Targets.* **6:** 923–928.

200. Curfman, C. L., K. Kirkland, and A. H. Merrill. 2006. Recent anticancer agents targeting sphingolipid pathways. *Expert Opinion on Therapeutic Patents.* **16:** 1129–1147.

201. Delgado, A., J. Casas, A. Llebaria, J. L. Abad, and G. Fabrias. 2007. Chemical tools to investigate sphingolipid metabolism and functions. *ChemMedChem.* **2:** 580–606.

202. Schwartz, G. K., D. Ward, L. Saltz, E. S. Casper, T. Spiess, E. Mullen, J. Woodworth, R. Venuti, P. Zervos, A. M. Storniolo, et al. 1997. A pilot clinical/pharmacological study of the protein kinase C-specific inhibitor safingol alone and in combination with doxorubicin. *Clin. Cancer Res.* **3:** 537–543.

203. Schwartz, G. K., A. Haimovitz-Friedman, S. K. Dhupar, D. Ehleiter, P. Maslak, L. Lai, F. Loganzo, Jr., D. P. Kelsen, Z. Fuks, and A. P. Albino. 1995. Potentiation of apoptosis by treatment with the protein kinase C-specific inhibitor safingol in mitomycin C-treated gastric cancer cells. *J. Natl. Cancer Inst.* **87:** 1394–1399.

204. Maurer, B. J., L. Melton, C. Billups, M. C. Cabot, and C. P. Reynolds. 2000. Synergistic cytotoxicity in solid tumor cell lines between N-(4-hydroxyphenyl)retinamide and modulators of ceramide metabolism. *J. Natl. Cancer Inst.* **92:** 1897–1909.

205. Wang, H., B. J. Maurer, C. P. Reynolds, and M. C. Cabot. 2001. N-(4-Hydroxyphenyl)retinamide elevates ceramide in neuroblastoma cell lines by coordinate activation of serine palmitoyltransferase and ceramide synthase. *Cancer Res.* **61:** 5102–5105.

206. Shirahama, T., E. A. Sweeney, C. Sakakura, A. K. Singhal, K. Nishiyama, S. Akiyama, S. Hakomori, and Y. Igarashi. 1997. In vitro and in vivo induction of apoptosis by sphingosine and N,N-dimethylsphingosine in human epidermoid carcinoma KB-3-1 and its multidrug-resistant cells. *Clin. Cancer Res.* **3:** 257–264.

207. Park, H. W., J. Y. Song, K. S. Kim, Y. Han, C. W. Kim, S. Y. Yi, and Y. S. Yun. 2004. Enhancement of radiosensitivity by combined ceramide and dimethylsphingosine treatment in lung cancer cells. *Exp. Mol. Med.* **36:** 411–419.

208. Gundewar, S., and D. J. Lefer. 2008. Sphingolipid therapy in myocardial ischemia-reperfusion injury. *Biochim. Biophys. Acta.* **1780:** 571–576.

209. Menaldino, D. S., A. Bushnev, A. Sun, D. C. Liotta, H. Symolon, K. Desai, D. L. Dillehay, Q. Peng, E. Wang, J. Allegood, et al. 2003. Sphingoid bases and de novo ceramide synthesis: enzymes involved, pharmacology and mechanisms of action. *Pharmacol. Res.* **47:** 373–381.

210. Struckhoff, A. P., R. Bittman, M. E. Burow, S. Clejan, S. Elliott, T. Hammond, Y. Tang, and B. S. Beckman. 2004. Novel ceramide analogs as potential chemotherapeutic agents in breast cancer. *J. Pharmacol. Exp. Ther.* **309:** 523–532.

211. Senkal, C. E., S. Ponnusamy, M. J. Rossi, K. Sundararaj, Z. Szulc, J. Bielawski, A. Bielawska, M. Meyer, B. Cobanoglu, S. Koybasi, et al. 2006. Potent antitumor activity of a novel cationic pyridinium-ceramide alone or in combination with gemcitabine against human head and neck squamous cell carcinomas in vitro and in vivo. *J. Pharmacol. Exp. Ther.* **317:** 1188–1199.

212. Bieberich, E., J. Silva, G. Wang, K. Krishnamurthy, and B. G. Condie. 2004. Selective apoptosis of pluripotent mouse and human stem cells by novel ceramide analogues prevents teratoma formation and enriches for neural precursors in ES cell-derived neural transplants. *J. Cell Biol.* **167:** 723–734.

213. Hiestand, P. C., M. Rausch, D. P. Meier, and C. A. Foster. 2008. Ascomycete derivative to MS therapeutic: S1P receptor modulator FTY720. *Prog. Drug Res.* **66:** 361, 363–381.

214. Huwiler, A., and J. Pfeilschifter. 2008. New players on the center stage: sphingosine 1-phosphate and its receptors as drug targets. *Biochem. Pharmacol.* **75:** 1893–1900.

215. Veronesi, U., L. Mariani, A. Decensi, F. Formelli, T. Camerini, R. Miceli, M. G. Di Mauro, A. Costa, E. Marubini, M. B. Sporn, et al. 2006. Fifteen-year results of a randomized phase III trial of fenretinide to prevent second breast cancer. *Ann. Oncol.* **17:** 1065–1071.

216. Vaishampayan, U., L. K. Heilbrun, R. E. Parchment, V. Jain, J. Zwiebel, R. R. Boinpally, P. LoRusso, and M. Hussain. 2005. Phase II trial of fenretinide in advanced renal carcinoma. *Invest. New Drugs.* **23:** 179–185.

217. Sabichi, A. L., S. P. Lerner, E. N. Atkinson, H. B. Grossman, N. P. Caraway, C. P. Dinney, D. F. Penson, S. Matin, A. Kamat, L. L. Pisters, et al. 2008. Phase III prevention trial of fenretinide in patients with resected non-muscle-invasive bladder cancer. *Clin. Cancer Res.* **14:** 224–229.

218. Sullards, M. C., J. C. Allegood, S. Kelly, E. Wang, C. A. Haynes, H. Park, Y. Chen, and A. H. Merrill, Jr. 2007. Structure-specific, quantitative methods for analysis of sphingolipids by liquid chromatography-tandem mass spectrometry: "inside-out" sphingolipidomics. *Methods Enzymol.* **432:** 83–115.

219. Han, X., and R. W. Gross. 2003. Global analyses of cellular lipidomes directly from crude extracts of biological samples by ESI mass spectrometry: a bridge to lipidomics. *J. Lipid Res.* **44:** 1071–1079.

220. Hait, N. C., C. A. Oskeritzian, S. W. Paugh, S. Milstien, and S. Spiegel. 2006. Sphingosine kinases, sphingosine 1-phosphate, apoptosis and diseases. *Biochim. Biophys. Acta.* **1758:** 2016–2026.

221. Riley, R. T., N. H. An, J. L. Showker, H. S. Yoo, W. P. Norred, W. J. Chamberlain, E. Wang, A. H. Merrill, Jr., G. Motelin, V. R. Beasley, et al. 1993. Alteration of tissue and serum sphinganine to sphingosine ratio: an early biomarker of exposure to fumonisin-containing feeds in pigs. *Toxicol. Appl. Pharmacol.* **118:** 105–112.

222. Hu, R., G. Li, Y. Kamijo, T. Aoyama, T. Nakajima, T. Inoue, K. Node, R. Kannagi, M. Kyogashima, and A. Hara. 2007. Serum sulfatides as a novel biomarker for cardiovascular disease in patients with end-stage renal failure. *Glycoconj. J.* **24:** 565–571.

223. Villanueva, L., A. Navarrete, J. Urmeneta, R. Geyer, D. C. White, and R. Guerrero. 2007. Monitoring diel variations of physiological status and bacterial diversity in an estuarine microbial mat: an integrated biomarker analysis. *Microb. Ecol.* **54:** 523–531.

224. Kato, M., Y. Muto, K. Tanaka-Bandoh, K. Watanabe, and K. Ueno. 1995. Sphingolipid composition in *Bacteroides* species. *Anaerobe.* **1:** 135–139.