Sphingolipid Metabolism Inhibitors and Cell Function

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Abstract: It has been widely shown that ceramide, ceramide phosphate, sphingosine and sphingosine-1-phosphate are molecules involved in cell proliferation, differentiation and apoptosis. The regulation of the enzymes that control their metabolism has an important role for cellular fate. Many studies in the field have shed light on sphingolipid metabolism inhibition and on its role in cell processes such as gene expression, DNA duplication and RNA transcription. The aim of this review is to collect, in a systematic way, the recent advances in the field of sphingolipid metabolism inhibitors, with special emphasis on their effects on cell function.

INTRODUCTION

The name sphingolipid was born at the end of the XIX century on the basis of the molecule structure similar to the riddle of the sphinx. These molecules are complex lipids, localized in animal and plant membranes and in some forms of lower life, which contain sphingoid bases and have a very complex metabolism.

The sphingolipid biosynthetic pathway initiates with the condensation of the serine with palmitoyl-CoA to form 3-ketodihydrosphingosine or 3-ketosphinganine, a serine palmitoyl transferase (SPT) catalyzed reaction. 3-ketodihydrosphingosine is reduced to dehydrosphingosine or sphinganine by a NADPH dependent 3-ketosphinganine reductase. An acylation in the presence of acyl-CoA produces dihydroceramide by ceramide synthase (CerS). Dihydroceramide is then converted to ceramide through the introduction of the double bond trans-4,-5 by dihydroceramide desaturase.

The ceramide can be transformed to sphingomyelin (SM) by two different ways: the first one involves the reaction with CDP-choline catalyzed by ceramide choline phosphotransferase and the second, described subsequently, consists in phosphocholine transfer from lecithin to ceramide by phosphatidylcholine: ceramide phosphocholine transferase or sphingomyelin synthase (SM-synthase). The synthesized SM can be used as source of phosphocholine to form phosphatidylcholine in the presence of diacylglycerol by reverse-sphingomyelin-synthase (RSM-synthase) freeing ceramide, or can be degraded to ceramide and phosphocholine by acid or neutral sphingomyelinase (aSMase or N-SMase). The new-produced ceramide can be deacylated by acid or neutral ceramidase (AC or NC) to form sphingosine, which can be phosphorylated by a sphingosine kinase 1 (SphK1) or by sphingosine kinase 2 (SphK2) to sphingosine phosphate, can be phosphorylated by ceramide kinase (CerK) or can be transformed to glucosylceramide and dihydroglucosylceramide then converted to lactosylceramide and to more complex sphingolipids. Today many inhibitors of the sphingolipid metabolism have been studied (Table **1**). They act at different steps reducing the level of molecules involved in signal transduction such as ceramide, ceramide phosphate, sphingosine and sphingosine-1-phosphate

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INHIBITORS OF SPHINGOLIPID BIOSYNTHETIC PATHWAY

Serine Palmitoyl Transferase (SPT)

The inhibition of SPT, first enzyme of sphingolipid synthesis, is responsible for the reduction either of sphingosine and ceramide or SM. The selective inhibitors of this enzyme are beta-chloroalanine which maintains low sphingosine content for a long time [1] and L-cycloserine which blocks the sphinganine synthesis [2]. Analogs of sphingosine such as cytotoxic cyclic analogs 99 and *Z*-4-methylsphingosine 2-azidosphingosine [3] interfere with SPT. In addition, SPT is inhibited with natural products [4] as stereoisomers of the antifungal sphingofungin B [5] which act by C-14 hydroxyl group and antifungal antibiotic myriocin [6] which does not need of C4-OH, C6 double bond and C14-keto groups, as well as C3-OH configuration, for its biological activity [6-8]. Myriocin blocks in this way the ceramide de novo synthesis in Jurkat acute leukemia cells [9] and in particular the formation of long-chain ceramide species which stimulate proteasomal activation with subsequent activation of caspases [10]. Sphinganine and not ceramide analogues, such as C2 and C6, are not able to reverse the inhibitory effect of myriocin [11]. Reducing the ceramide content, myriocin is responsible for the decrease of SM concentration in the liver [12].

Ceramide-Synthase (CerS)

Besides SPT inhibitors, ceramide synthesis is also inhibited by molecules which act specifically on CerS as Fumonisin B1 and mycotoxin, appertaining to fumonisins family, produced by the fungus Fusarium verticillioides [13]. The specificity of Fumonisin B1 for this enzyme has been demonstrated in LLC-PK1 cells in which the decrease of pERK2 phosphorylation in response to Fumonisin B1 is

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Table 1. Inhibitors of Sphingolipid Metabolism. The Table Lists Inhibitors, Enzyme Inhibited, Cell Type, and References

Inhibitor	Enzyme	Cell Type	Reference
Alpha-ketoamides (5-7)	AC	liver	Org Biomol Chem 2005;3(20):3707-12.
Beta- chloroalanine	SPT	skin	Crit Rev Ther Drug Carrier Syst 1991;8(3):193-210.
		skin	Int J Pharm 2002;238(1-2):43-50.
C11AG	N-SMase	macrophages	Drugs Exp Clin Res 2003;29(1):5-13
		Jurkat T-cell lymphoma cells	Biochem Pharmacol 2005;69(8):1141-8.
Ceramide analogues	AC	human HaCaT keratinocytes and human melanoma cells	FEBS Lett 2002;516(1-3):47-52.
Chlorogentisylquinone	N-SMase	brain	J Antibiot (Tokyo) 2001;54(11):882-9
d,l-PDMP	GCerS	brain	J Neurochem 1999;72(3):1040-9.
D609	SMS	U937 human monocytic leukemia cells	Exp Cell Res 2004;292(2):385-92
D-DPMA	GCerS	human cancer cell	J Lipid Res 1995;36(3):611-21.
		brain	J Lipid Res 1987;28(5):565-71.
		PC12 cells	J Biol Chem 1998;273(40):26001-7.
Desipramine	AC	DU145, 5637, and Hela cancer cell line	FEBS Lett 2006;580(19):4751-6.
		fibroblasts	Biol Chem Hoppe Seyler 1994;375(7):447-50.
D-MAPP	N-C	mesangial cells	Gene 2003;315:113-22.
D-threo- PPPP	GCerS	fibroblasts	J Biochem 2000;127(3):485-91.
Extracts of Sporormiella aus-tralis	sphinganine N-acyltransferase	fungi	J Antibiot (Tokyo) 1995;48(5):349-56.
F-12509	CerK	mast cells	Biochim Biophys Acta 2005;1738(1-3):82-90.
Fumonisin B1	CerS	LLC-PK(1) cells	Food Chem Toxicol 2005;43(1):123-31.
		porcine renal epithelial cells	Cell Biol Toxicol 2004;20(4):197-212.
		liver	Toxicol Sci 2006;94(2):388-97.
		LLC-PK1 cells	J Toxicol Environ Health A 2004;67(23-24):2085-94.
		tumour cells	Biochim Biophys Acta 2002;1585(2-3):188-92.
Glutathione	N-SMase	Molt-4 cells	J Biol Chem 1997;272(26):16281-7.
GT11	dihydroceramide desaturase	Jurkat A3 cells	Org Biomol Chem 2005;3(20):3707-12.
		liver	Angew Chem Int Ed Engl 2001;40(10):1960-1962.
		primary cultured cerebellar	Mol Pharmacol 2004;66(6):1671-8.
GW4869	N-SMase	MCF7 cells	J Biol Chem 2002;277(43):41128-39.
LCL204	SphK1	prostate cancer	Cancer Chemother Pharmacol 2008;61(2):231-42.
l-Cycloserine	SPT	neutrophils	J Biol Chem 2003;278(2):974-82.
		MDA-MB 468 breast cancer cells	Mol Cancer Ther 2002;1(9):719-26
L-threo-sphinganine	SphK1	swiss 3T3 fibroblasts	J Biol Chem 1998;273(36):23585-9
Manumycin A	N-SMase	brain	Chembiochem 2001;2(2):141-3.
Minopentol backbone (AP1)	CerS	HT29 cells	J Biol Chem 1998;273(30):19060-4.
MS-209	SMS	breast cancer and non-small-cell lung cancer	Curr Opin Investig Drugs 2004;5(12):1340-7.

Table 1 Cont…

Myriocin	SPT	Jurkat cells	J Biol Chem 2001;276(48):44848-55.
		T-cell hybridomas and T-cell blasts	Cell Death Differ 2003;10(2):193-202.
Myriocin derivates	SPT	lymphocyte	J Antibiot (Tokyo) 1996;49(9):846-53
ISP-1/Myriocin	SPT	Ramos cells	J Biol Chem 2003;278(17):14723-31.
N-(n-butyl) deoxynojirimycin	GCerS	GM95 mouse melanoma cells	Faseb J 2003;17(9):1144-6
N,N-dimethylsphingosine	SPHK1	human A549 epithelial lung carcinoma cells	Cell Signal 2005;17(10):1203-17.
N-butyldeoxynojirimycin	GCerS	brain	J Neurochem 1999;72(3):1040-9.
	glucosylceramide transferase	brain	Acta Paediatr Suppl 2005;94(447):69-75
	glucosidases and glucosyltransferases	HL-60 cells	J Biol Chem 1994;269(11):8362-5.
NO/cGMP	A-SMase	dendritic cells	J Immunol 2004;173(7):4452-63.
N-octyl beta-valienamine	beta-glucocerebrosidase	liver	Bioorg Med Chem 1998;6(10):1955-62
OGT2378	GcerS	MEB4 melanoma cells	Cancer Res 2003;63(13):3654-8.
PDMP and PPPP	GCerS	hepatoma cells	Biochem Biophys Res Commun 2001;288(1):269-74.
		KB cell lines	Br J Cancer 1999;81(3):423-30
Scyphostatin	N-SMase	endothelial cell	Am J Physiol Heart Circ Physiol 2004;287(3):H1344-52.
		brain	J Antibiot (Tokyo) 1999;52(6):525-30
Scyphostatin analogue	N-SMase	monocytes, macrophages, hepatocytes	Chembiochem 2005;6(4):726-37
		brain	Bioorg Med Chem 2001;9(11):2901-4.
SG12 and SG14	SphK2	platelets	Bioorg Med Chem. 2005 May 16;13(10):3475-85
Sphingofungin B and C	SPT	Saccharomyces cerevisiae	J Biol Chem 1992;267(35):25032-8
SM analogues	SMase	PC-12	Nippon Yakurigaku Zasshi 2002;120(1):67P-69P
		PC-12	Bioorg Med Chem Lett 2001;11(10):1277-80
		Bacillus cereus	Org Lett 2000;2(17):2627-9
		Bacillus cereus	Org Lett 2003;5(16):2801-4.
SR33557	A-SMase	P388/ADR cells	J Biol Chem 1991;266(29):19858-60
		Rinm5F cells	Autoimmunity 2000;32(4):241-54.
XM462	dihydroceramide desaturase	Jurkat A3 cells	ChemMedChem 2008; 3(6):946-53.

independent of sphingosine and ceramide derived from SM and it is due only to the inhibition of de novo ceramide biosynthesis [14]. Curiously, after 5 daily treatment with Fumonisin B1 in mice, the SPT and acid sphingomyelinase (aSMase) activity and sphingoid bases content increase significantly in the liver to maintain a balance of cellular sphingolipids [15]. This effect is specific since another hepatotoxicant, as acetaminophen, induces liver regeneration similar to Fumonisin B1 but does not produce similar effects on liver sphingolipid metabolizing enzymes [16]. Nevertheless it has been shown that Fumonisin B1 enters with difficulty into the cells determining a low inhibitor effect on CerS. The higher effect is obtained by australifungin, a micotoxin isolated from *Sporormiella australis*, but its functional groups present a very high chemical reactivity which is responsible for its limited use [17]. A toxic metabolite of hydrolyzed fumonisin, AP1 and the fumonisin-related AAL-toxin are also potent CerS inhibitors [18].

N-Acyl-Sphingosine Dehydrogenase or Dihydroceramide Desaturase

The cyclopropene ceramide analog GT11 has been described as the first selective inhibitor of the dihydroceramide desaturase [19]. GT11 is a competitive inhibitor and its analogs with N-hexanoyl and N-decanoyl moieties inhibit the enzyme with similar effects [20]; differently urea and

thiourea analogs have significantly lower effect [21]. A new inhibitor of this enzyme is XM462 which reduces the viability in Jukat A3 cells cultured in serum-free medium [22].

Cytidyldiphosphocholine Transferase

The cAMP analogs as chlorophenylthio-cAMP reduce strongly CTP: phosphocholine cytidylyltransferase activity in cultured rat hepatocytes [23]. In HaCaT cells this enzyme is inhibited by sphingomyelin analogs as acetyl-erythro-sphingosine-1-phosphocholine and N-octanoyl-erythro-sphingosine-1-phosphocholine [24] whereas reversible inhibitors are sphingosine and lysosphingolipids [25].

Sphingomyelin Synthase (SMS)

In U937 human monocytic leukemia cells, SMS is inhibited by tricyclodecan-9-yl-xanthogenate [26] but its pivaloyloxymethyl analog results more potent [27]. Also MS-209, a quinolone-derivative, has been described as a SMS inhibitor [28]. In addition, the products of SM metabolism, ceramide and sphingosine, inhibit SMS restoring the homeostasis between SM and ceramide pools [29].

INHIBITORS OF SPHINGOMYELIN UTILIZATION

The existence of different kinds of sphingomyelinase (SMase) has been widely described [30]. The Mg^{++}-dependent, membrane-associated, neutral SMase (N-SMase) is inhibited, in a dose-dependent manner, by glutathione at physiological concentrations [31]. An active compound which inhibits N-SMase, scyphostatin, has been extracted from a discomycete, Trichopeziza mollissima SANK 13892 [32]. This inhibitor determines a 50% inhibition of N-SMase at the concentration value of 1.0 microM whereas to inhibit lysosomal aSMase approximately 50-fold greater concentration is required [33]. Scyphostatin, inhibiting N-SMase, prevents downstream of mitogen-activated protein kinases [34]. Also scyphostatin analogs 3a and 3b have inhibitor effects on N-SMase; in particular the primary hydroxy group in compound 2 is important for this activity [35]. Scyphostatin analog 14, a chemically and metabolically stabilized compound lacking the epoxy function of the natural congener and carrying a palmitic acid group instead of the native trienoyl residue induces N-SMase inhibition in several systems (monocytes, macrophages, hepatocytes) [36]. Chlorogentisylquinone, purified from the culture broth of a fungal strain FOM-8108 isolated from a marine environment by solvent extraction, silica gel chromatography and Sephadex LH-20 chromatography, inhibits N-SMase rat brain membranes [37]. One inhibitor discovered in the screening on N-SMase, GW4869, functions as a noncompetitive inhibitor of the N-SMase *in vitro* but it does not inhibit aSMase [38]. An irreversible specific inhibitor of N-SMase is manumycin A [39] whereas competitive inhibitors have been extracted from Abies nephrolepis, Acer tegmentosum, and Ginkgo biloba [40]. A SM methylene analog, synthesized by Hofmann rearrangement of the alpha-hydroxyethyl-beta-hydroxy amide 4 followed by the intramolecular oxazolidinone ring formation as the key steps, has been designed as a N-SMase inhibitor [41]. Also SM nitrogen analog 1 synthesized by continuous Hofmann and Crutius rearrangement as key steps in constructing the 3-hydroxy-1,2-diamine structure in the backbone of 1 has SMase inhibitor effect [42]. In PC-12 neurons another analog of SM, difluoromethylene, inhibits N-SMase activity [43]. Short-chain analogs of N-palmitoylsphingosine-1-phosphate are non-competitive inhibitors with the same level of inhibitory activity of schyphostatin [44]. N-SMase inhibitor, undecylidene aminoguanidine C11AG, blocks lipopolysaccharide-stimulated SM degradation and NF kappa B activation [45]. SR33557, an aSMase inhibitor, counteracts TNF-alpha-mediated effect of IL-1beta-induced NF-kappaB activation [46]. Nitric oxide/cyclic guanosine 3',5'-monophosphate pathway inhibits aSMase in a mouse model of lipopolysaccharide-induced sepsis [47] whereas desipramine induces proteolytic degradation of aSMase [48].

INHIBITORS OF CERAMIDE UTILIZATION

The ceramide new-synthesized or derived from SM metabolism can be phosphorylated, glycosylated to form glucosylceramide and galactocerebroside or degraded to sphingosine which can be phosphorylated by sphingosine-1-kinase (SphK1). Many enzymes are involved in this metabolism and different inhibitors have been described. The ceramide kinase (CerK) is inhibited specifically by a novel F-12509A olefin isomer, K1, which does not act on sphingosine kinase and diacylglycerol kinase [49]. N-butyldeoxynojirimycin and N-butyldeoxygalactonojirimycin, synthetic analogs of the iminosugars, polyhydroxypiperidine alkaloid deoxynojirimycin, have inhibitory activity against the glucosyl ceramide synthase (GCerS) [50, 51]. Also *N*-nonyldeoxynojirimycin [52] and the imino sugar OGT2378 [53] show effects on GCerS in different cell studies. D-threo-1-phenyl-2-benzyloxycarbonylamino-3-pyrrolidino-1-pr opanol is the most potent inhibitor [54]. 1-phenyl-2-decanoylamino-3-morpholino-1-propanol inhibits the human GCerS, but not the same enzyme present in other organisms [55]. SR33557 inhibitor consists of four isomers and only D-threo is the active form on murine GCerS [56]. Moreover some isofagomine analogues of the alkaloid fagomine bearing an alkyl chain on the C6 position [57]. *N*-acyl derivatives of valienamine lack inhibitory activity, while the corresponding *N*-alkyl analogs are very strong GCerS inhibitors; the *N*-octyl derivative of valienamine is the most potent of the series [58]. The inhibition of glucosylceramidase (GCase) has been obtained by 1-phenyl-2-decanoylamino-3-morpholino-1-propanol (PDMP) and 1-phenyl-2-hexadecanoylamino-3-pyrrolidino-1-propanol [59]. The acid ceramidase (AC) activity present in human HaCaT keratinocytes and human melanoma cells is suppressed by ceramide analogs (1S,2R)-2-N-myristoylamino-1-phenyl-1-propanol and (1R,2R)-2-N-myristoylamino-1-(4-nitrophenyl)-1,3-propan-diol [60]. Also desipramine induces a time- and dose-dependent down regulation of AC [61] whereas D-erythro-(1R,2R)-2-N-myristoylamino-1-(4-nitrophenyl)-1,3 propandiol is used as inhibitor of neutral ceramidase (NC) [62]. The new inhibitor of AC is LCL204 which in DU145 PCa cells induces a reduction of sphingosine and an increase of ceramide level [63]. Compounds extracted from fungi as sphingosine derivatives, DL-threo-dihydrosphingosine and dimethylsphingosine, inhibit the SphK1 activity [64]. The inhibition of basal SphK1 activity is obtained by N,N-dimethylsphingosine [65]. DL-threo-dihydrosphingosine, besides SphK1 inhibition, increases the sensitivity of HEK-293 cells to fumonisin B1, inhibitor of CerS [66]. While N,N-dimethyl-

sphingosine displays inhibitory effects for both SphK1 and SphK2, synthetic sphingoid analogs, as SG12 and SG14, have specific inhibitory effects on SphK2 [67].

ROLE ON CELL FUNCTION

Sphingolipid metabolism produces second messengers as ceramide, sphingosine and sphingosine-1-phosphate which have different functions in cell life. For instance, while ceramide can mediate and induce cell death, sphingosine-1-phosphate results to be a second messenger for cell survival and proliferation and protection against ceramide mediated apoptosis [68-70]. Sphingolipid metabolites are interconvertible, it is not the individual quantitative level to determine cell fate, but their relative levels [71]. This observation has brought some researchers to propose the "rheostat ceramide/ sphingosine-1-phosphate" model indicating that an equilibrium between these two molecules is crucial to determine a cell survival or death [72] In addition, the biological effects of all sphingolipid metabolites may vary according to cell type, depending on the extracellular stimulus, on the relation between the different metabolite concentrations and the involved subcellular compartment, on cell cycle phase and on cell development [73-75]. The regulation of sphingolipid metabolites depends on their specific enzyme activity and therefore, the enzyme inhibitors are able to modify cell function (Fig. **1**).

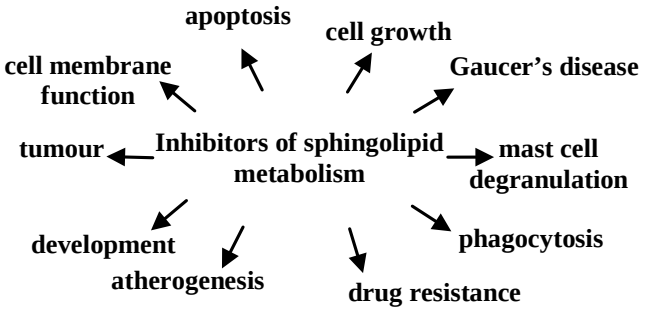

Fig. (1). Sphingolipid metabolism inhibitors in physiological and pathological processes.

Apoptosis and Cell Growth

Apoptosis is modified by myriocin which, by acting on SPT, interferes with the early steps of CD95-mediated caspase activation inducing the block of activation-induced cell death in T-cell hybridomas and T-cell blasts [76]. Also tricyclodecan-9-yl-xanthogenate, by inhibiting SMS, induces apoptosis in U937 human monocytic leukemia cells [77] such as Fumonisin B1, by inhibiting CerS, regulates serum deprivation-induced cell death in LLC-PK1 cells [78]. Moreover Fumonisin B1 abrogates the proapoptotic effects of palmitate on L6 myotubes [78]. Inhibition of basal SphK1 activity, obtained by N,N-dimethylsphingosine, induces a spontaneous apoptosis in A549 carcinoma cells [66]. In contrast to pro-survival SphK1, the protein SphK2, a nuclear protein [79], inhibits cell growth and enhances apoptosis [80]. In fact the suppression by small interfering RNA treatment prevents serum deprivation- or drug-induced apoptosis in HEK293 cells [81]. Three multidrug-resistant KB cell lines, KB-C1, KB-A1 and KB-V1 are induced to apoptosis preferentially respect to the drug-sensitive cell lines by two inhibitors of GCase, PDMP and 1-phenyl-2-hexadecanoylamino-3-pyrrolidino-1-propanol [60]. Difluoromethylene analog of SM inhibits N-SMase activity in bovine brain microsomes suppressing TNF-induced apoptosis of PC-12 neurons at a low concentration [44]. In the same cells, nine difluoromethylene analogues of SM suppress cell death induced by serum deprivation [82]. In Jurkat T-cell lymphoma cells, treatment with C11AG, by inhibiting N-SMase, enhances the sensitivity to apoptosis [46]. SR33557, an aSMase inhibitor, avoids apoptosis induced by TNF in ML-1a cells [83]. Treatment with D-threo- PDMP, but not with another GCerS inhibitor, N-butyldeoxynojirimycin, results in a dose-dependent reduction of the growth rate [84]. D-threo-1-phenyl-2-decanoylamino-3-morpholino-1-propanol inhibits NGF-induced neurite outgrowth of PC12 cells [85]. Differently GT11, by inhibiting dihydroceramide desaturase, has cell protective properties in primary cultured cerebellar neurons [86].

Development

PDMP, by inhibiting glucosphingolipid synthesis, triggers oocyte meiotic maturation representing an important regulator of oocyte meiosis [87]. Moreover PDMP-treated fish embryos are fully viable with no evidence of developmental abnormality [88].

Membrane Structure and Function

Chloroalanine, inhibitor of SPT, has been used to investigate the role of skin sphingosine synthesis inhibition in the permeation of levodopa, a hydrophilic drug, across rat skin [89]. Moreover beta- chloroalanine causes paradoxical effects on barrier permeability homeostasis [90] and delays mammalian epidermal barrier recovery after acute perturbation [91]. In essential fatty acid deficient mice, with a chronic disturbance in barrier function, beta-chloroalanine impairs recovery of barrier structure and function [92].

Tumor Process and Drug Resistance

The iminosugar OGT2378, inhibitors of GCerS, is used to reduce tumorigenic capability of MEB4 melanoma cells [54]. D-threo- PDMP exhibits anti-tumor activity against two different Shope carcinoma cell lines [93]. Both threo and erythro racemic compounds of SR33557, an aSMase inhibitor, induces a strong inhibition of human cancer cell growth [94]. Moreover GCerS inhibition by 1-phenyl-2-hexadecanoylamino-3-pyrrolidino-1-propanol enhances tumor cell sensitivity to vincristine [95]. 1-phenyl-2-hexadecanoyl amino-3-pyrrolidino-1-propanol and PDMP enhance doxoru bicin-elicited ceramide elevation in HepG2 hepatoma cells but only the second inhibitor potentiates the modest apoptotic response to doxorubicin treatment [96]. The GCase inhibition has been implicated in drug resistance; in fact in the absence of functional Gcase, melanoma cells do not respond to anticancer drugs [97].

Other Functions

The inhibition of CerK is responsible for mast cell degranulation [50]. L-cycloserine, by inhibiting SPT, enhances phagocytosis in COS-1 cells [98]. Reducing the ceramide content, myriocin is responsible for the decrease of SM concentration in the liver [12] accompanied by a decrease of total cholesterol and triglyceride plasma levels and of atherosclerotic lesions demonstrating that inhibition of SM synthesis reduces atherogenesis [12].

N-butyldeoxynojirimycin and the imino sugar OGT2378, inhibitors of GCerS, are used in therapy for treating type I Gaucher disease [99]. Treatment with D-threo PDMP, but not with another GCerS inhibitor, causes cell death in NG108-15 cells, suggesting that this inhibitor is toxic for treatment of Gaucher's disease [85]. In chinese hamster ovary cells it induces lysosomes vacuolization [100].

CONCLUSIONS

In the last ten years the definition of "fundamental structural components of biological membranes" has become reductive for the sphingolipids. In fact, this class of lipids constitutes a source of bioactive molecules which, together with the enzymes of the sphingolipid metabolism, are involved in signal transduction and in some fundamental cellular mechanisms as proliferation and its arrest, differentiation and apoptosis, embryogenesis and ageing regulation. Different inhibitors of the sphingolipid metabolism are able to block the synthesis and/or utilization of the sphingolipids at various levels regulating the metabolite molecule concentrations. The direct consequence is the modification of cell proliferation, differentiation and/or apoptosis lipid-induced. Therefore inhibitors of sphingolipid metabolism enzymes could be used to regulate cell function in physiological and pathological conditions.

ABBREVIATIONS

AC	=	Acid ceramidase
aSMase	=	Acid sphingomyelinase
CerK	=	Ceramide kinase
CerS	=	Ceramide synthase
GCase	=	Glucosyl ceramidase
GCerS	=	Glucosyl ceramide synthase
NC	=	Neutral ceramidase
N-SMase	=	Neutral sphingomyelinase
PDMP	=	1-Phenyl-2-decanoylamino-3-morpholino-1-propanol
PPPP	=	1-Phenyl-2-palmitoylamino-3-pyrrolidino-1-propanol (P4)
SM	=	Sphingomyelin
SMS	=	Sphingomyelin-synthase
SphK1	=	Sphingosine kinase 1
SphK2	=	Sphingosine Kinase 2
SPT	=	Serine palmitoyl transferase

ACKNOWLEDGEMENTS

We wish to acknowledge support from Ministero dell' Università e Ricerca (PRIN project), ASI (Agenzia Spaziale Italiana), and the Fondazione Cassa di Risparmio di Perugia. We also thank Dr. Ilaria Bernardini, and Remo Lazzarini for the discussion and Silvano Pagnotta for the figure preparation.

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Received: June 04, 2008 Revised: July 07, 2008 Accepted: July 17, 2008