Ether Lipids as Anticancer Agents: Focus on Non-Phosphorus Cationic Glycerolipids

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Abstract: This review focuses on the synthesis and mechanisms of antitumor activity of cationic non-phosphorus analogs of edelfosine. The role of variable length and mode of conjugation of the spacer group, the types of cationic 'head', and the length of the substituent at C(2) atom of the glycerol backbone are discussed, providing the basis for rational design of lipophilic anticancer drugs, in particular, for elimination of multidrug resistant cells.

Keywords: Ether lipids, Non-phosphorus cationic glycerolipids, Anticancer drugs, Edelfosine (ET-18-OMe).

INTRODUCTION

Ether glycerolipids are the focus of intense investigations. Initially a major interest to this class of compounds was bound to their use as components of liposomes, i.e. vehicles for delivery of genetic constructs and small molecule drugs to eukaryotic cells [1], and to studies of artificial membranes [2]. Further studies broadened the therapeutic potential of ether glycerolipids. From the structural viewpoint, these glycerolipids are predominantly the derivatives of long chain 1,2-dialkylglycerols. Introduction of short chain substituent at the position C(2) of the glycerol backbone yields the compounds with clinically important characteristics such as platelet activating factor (PAF) antagonism, antiviral (in particular, anti-human immunodeficiency virus type 1), antibacterial and anticancer activities [3-6].

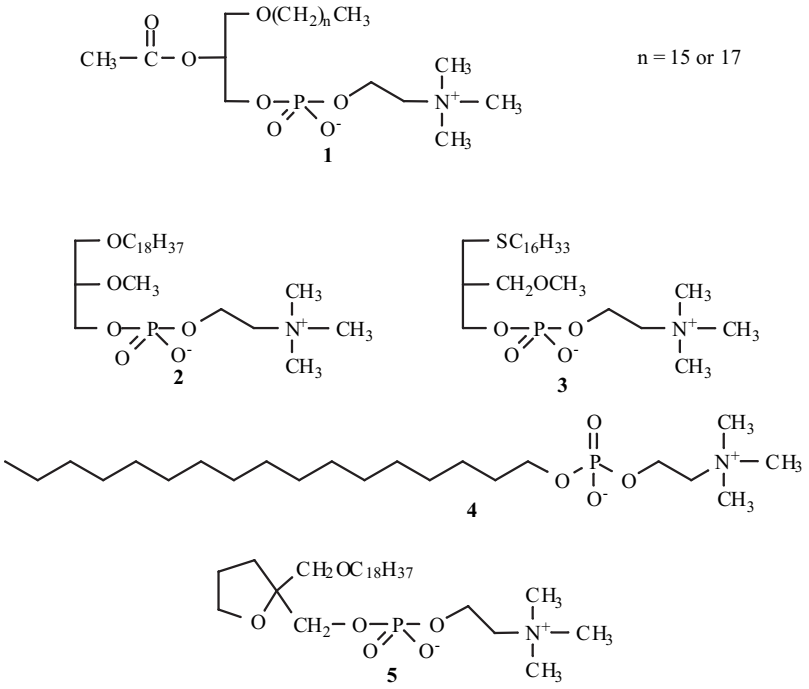

Fig. (1). PAF and some of its clinically used analogs.

Ether glycerolipids became the focus of intensive studies with the discovery of PAF (**1**; (Fig. **1**)).

Based on PAF structure, a number of its structural analogs have been synthesized. The prototypic drug edelfosine (ET-18-OMe) (**2**; (Fig. **1**)) with methyl substituent at the C(2) atom of glycerol, demonstrated the ability to preferentially kill cancer cells while sparing non-malignant counterparts [reviewed in ref. 7]. Structurally

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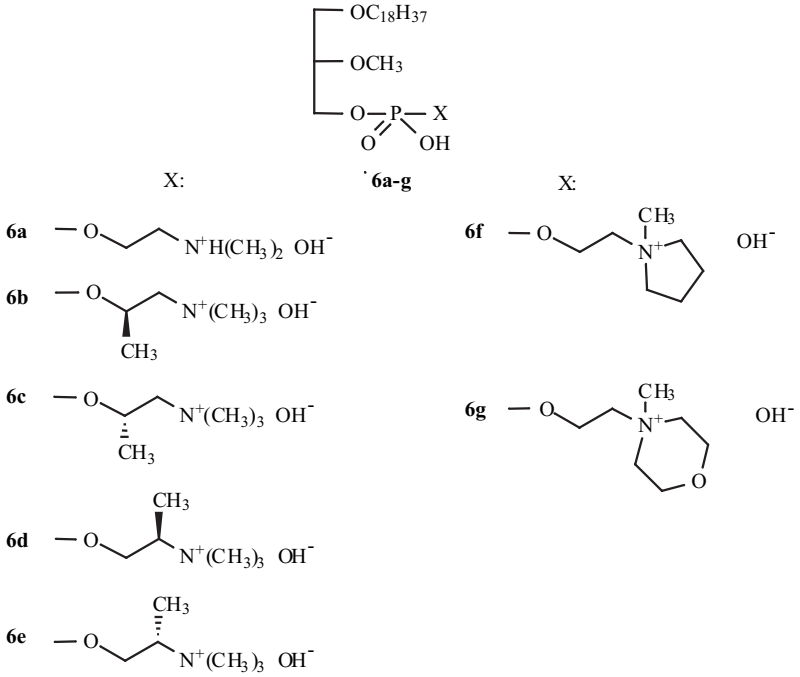


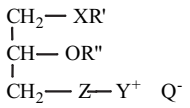
Fig. (2). Compounds **6a-g**, structural analogs of ET-18-OMe.

close analogs of ET-18-OMe (compounds **6a-g**, (Fig. **2**) also showed high antitumor activity [8], confirming the perspectiveness of these lipids as anticancer agents. Most importantly, the mechanism of antitumor effect of edelfosine involves selective targeting of lipid microdomains in tumor cells and activation of Fas/CD95/APO-1 mediated apoptosis [9]. Edelfosine is internalized into lipid rafts of tumor cells via endocytosis [10, 11]. Once in the cell, edelfosine triggers recruitment of Fas-associated death domain protein, procaspases-8 and -10, c-Jun N-terminal kinase, and Bid, the molecules critical for initiation of apoptosis [9]. Thus, death receptor (Fas) and mitochondrial apoptotic routes are spaciously linked, resulting in disruption of the mitochondrial transmembrane potential, production of reactive oxygen species, caspase-3 activation, cleavage of poly(ADP-ribose) polymerase, and DNA fragmentation [12]. However, in certain cell types Fas signaling is not affected by edelfosine; rather, the cytotoxicity of the drug is associated with inhibition of phosphatidylcholine synthesis [10]. Other important targets of ether lipids in rafts are protein kinase C and Raf-1, the components of the anti-apoptotic mitogen activated protein kinase cascade [13, 14]. Edelfosine evokes no DNA damage and is not mutagenic [6], which makes it advantageous over many conventional chemotherapeutics. Furthermore, octadecyl-(N,N-dimethyl-piperidinio-4-yl)-phosphate (D-21266; Perifosine; patented by Zentaris GmbH, Frankfurt on Main, Germany) induced cell cycle arrest by activating $p21^{Waf-1}$ in a p53-independent manner, suggesting clinical efficacy of this agent for tumors with altered p53 pathway [15]. In preclinical settings edelfosine synergized with paclitaxel and teniposide in killing leukemia cells [16]. In clinical trials edelfosine and structurally close thioether phosphocholine ilmofosine (BM 41.440) (**3**; (Fig. **1**)), hexadecylphosphocholine (miltefosine; **4**) and cyclic lipid SRI 62.834 (**5**) showed promising potency in patients with advanced solid tumors [17, 18], head and neck cancer [19], non-small cell lung cancer [20], mammary gland carcinoma [21, 22] and in efficient purging of leukemia cells from the autologous bone marrow transplant [23]. These properties indicate the uniqueness of ether glycerolipids for cancer chemotherapy.

This review analyzes the synthesis and structure-activity relationship (SAR) of antitumor cationic glycerolipids. In particular, evidence is provided in support of edelfosine-like non-phosphorus ether lipids as perspective anticancer agents. Beyond the scope of this article are the applications of ether lipids in the areas other than oncology (see, e.g., [24]).

NON-PHOSPHORUS CATIONIC GLYCEROLIPIDS: SYNTHESIS AND SAR

The synthesis of non-phosphorus alkyl lipids requires fewer stages and is less laborious since no phosphorus containing groups have to be linked. These compounds are likely to have long-lasting biological activity due to the resistance to phospholipases. The majority of biologically active non-phosphorus cationic lipids can be presented as **7** (Fig. **3**) [2, 25-27].



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where X – O, S, OCONH;

R' – long chain (C_{10}–C_{20}) alkyl, alkenyl or acyl;

R" – long chain (C_{10}-C_{20}) or short chain (C_1-C_4) alkyl substituent;

Z - no spacer or a C_{1-8} spacer group of alkyl, acyl or amide types;

Y^+ - ammonium or sulfonium aliphatic 'head' with short (C_1-C_3) substituents of alkyl type, or heterocyclic 'head' with positively charged N or S atoms (pyridinic or thiazolinic 'heads');

Q^- - counterion (Hal^-, AcO^-, TsO^-).

Fig. (3). Compound **7**, general structure of biologically active cationic non-phosphorus lipids.

The R' and R" radicals can be linked to form the dioxalane cycle with a long chain alkyl substituent (compound **8**; R' = $C_{17}H_{35}$, R" = H) (Fig. **4**) [28, 29]:

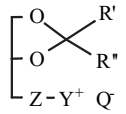

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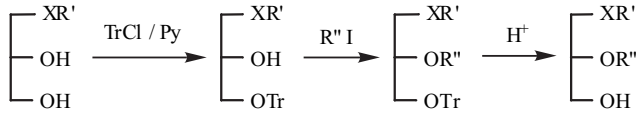
Fig. (4). Compound **8**, glycerolipid with dioxalane cycle.

In general, synthesis of major ether glycerolipids includes the modifications of 1,2-dialkylglycerol **9** at the position 3. The most widely utilized approach is illustrated by the synthesis of compound **9** using trityl protection group (Scheme **1**) [28].



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Scheme 1. Synthesis of 1,2-dialkylglycerol **9**.

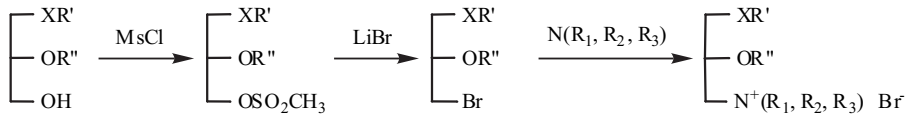
Based on this principle, the ether lipids **10** have been synthesized in which the ammonium type cationic group is linked directly to the glycerol fragment. The reactions included mesylation of diglyceride, replacement of mesyl residue for bromine followed by quaternization of the respective tertiary amine (Scheme **2**) [4, 28].

A variety of methods has been developed to obtain the compounds with a spacer group between hydrophilic and hydrophobic domains of the lipid molecule. For example, lipid **13** with quaternary ammonium 'head' separated from the glycerol fragment by several methylene groups can be synthesized based on compound **11** (Scheme **3**) which is transferred into positively charged glycerolipids **13** according to Scheme **2** [28].

Alternatively, *rac*-1-alkyl-2-benzylglycerol **12** can be used as an initial compound for synthesis of lipids **13**. The resulting molecule is constructed by step-by-step linkage of structural domains (pathway **A**, Scheme **4**) or by linking the hydrophilic domain-spacer complex to the hydrophobic domain followed by quaternization of tertiary amine (pathway **B**, Scheme **4**) [30, 31].

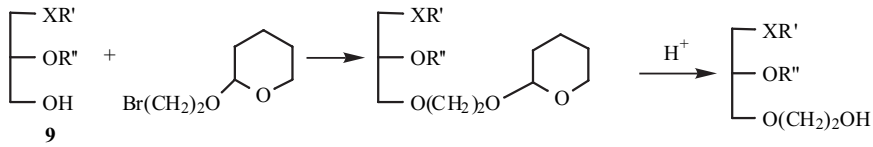
Morris-Natschke *et al.* have reported the synthesis of positively charged glycerolipids **14** with thioethyl spacer group introduced into the lipid molecule by thioalkylation (Scheme **5**) [32]. The subsequent processes include the reactions as in Schemes **2** and **4A.**

Scheme **6** depicts the synthesis of compounds **15** with aliphatic or heterocyclic substituents at the position C(2) of glycerol and polyethylene glycol residues as spacers (Scheme **6**, **A**). Also, the reaction can be modified using trytile protective group (Scheme **6**, **B**) [33].



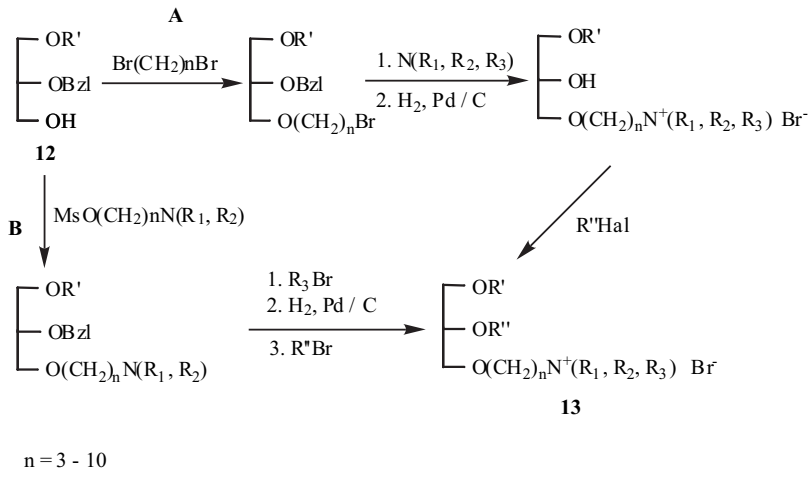
9 **10**

Scheme 2. Synthesis of lipids **10** with cationic 'head' linked directly to the glycerol backbone.



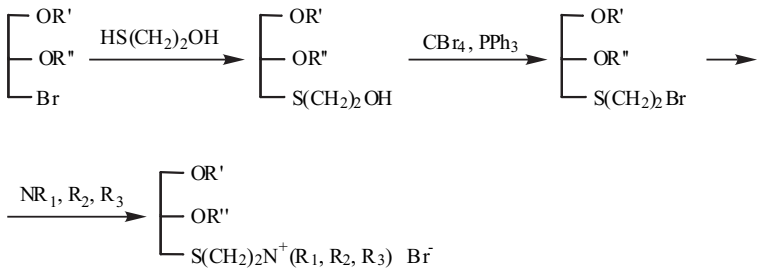
9 **11**

Scheme 3. Synthesis of compound **11**.



n = 3 - 10

Scheme 4. Synthesis of cationic glycerolipids **13** with alkyl aliphatic spacer.



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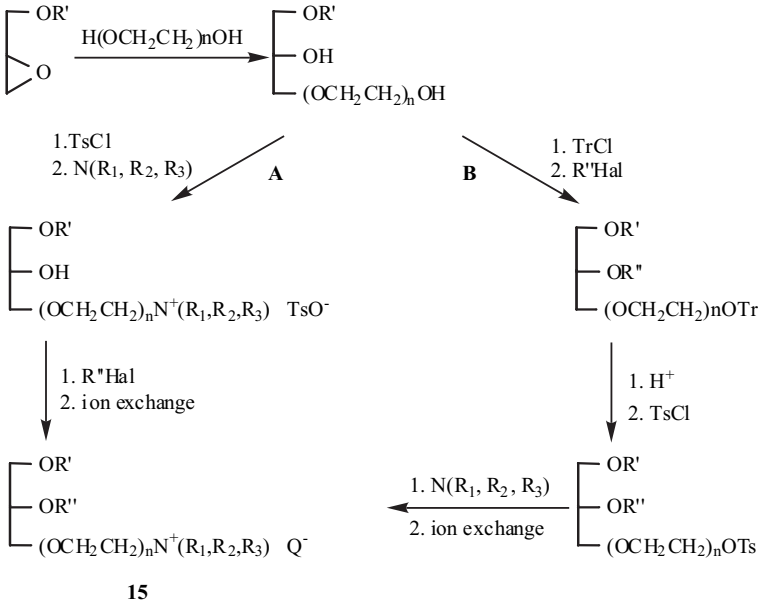
Scheme 5. Synthesis of glycerolipids **14**.



n = 0-2, 4

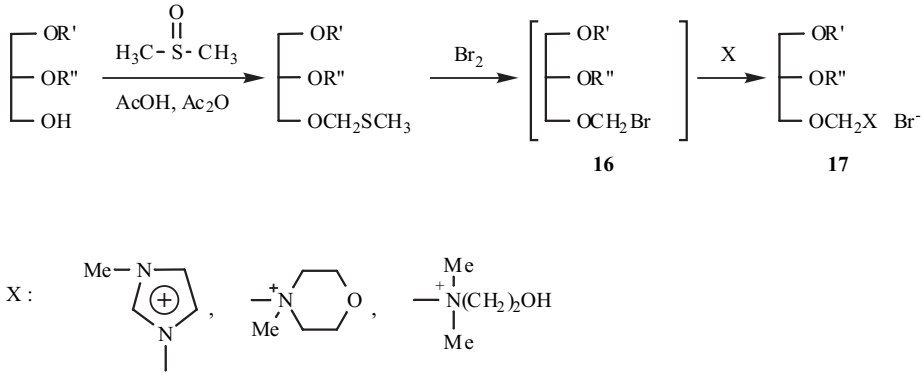
R'' = n-C_3H_7, n-C_4H_9, $CH_2CO_2^-$, CH_2CF_3, CH_2COMe,

15

Scheme 6. Reactions for obtaining glycerolipids **15**.

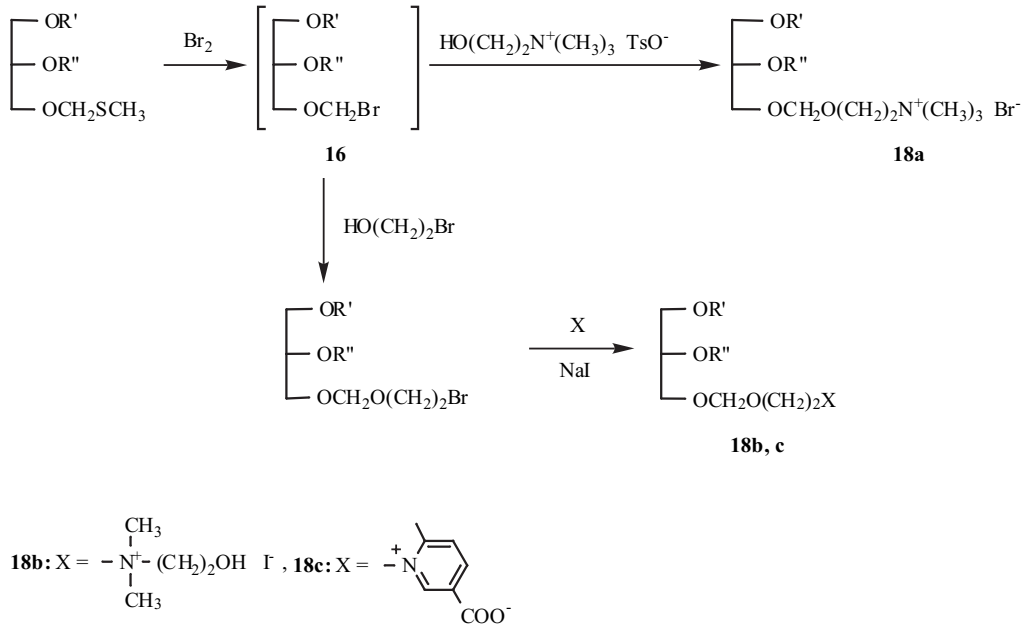
Methylthiomethyl ethers of *rac*-dialkylglycerols have been used to prepare glycerolipids **17** with the polar 'head' separated from the glycerol fragment by methylene group (Scheme **7**) [34, 35]. The key component of the reaction is α-bromoether **16** formed in situ upon interaction of methylthiomethyl ether of dialkylglycerol with bromine. This brominated ether readily reacts with various nucleophilic agents, in particular, with tertiary aliphatic or heterocyclic amines.

Using methylthiomethyl ethers of *rac*-dialkylglycerols, we have synthesized lipids **18a-c** with the spacer group



16 **17**

Scheme 7. Glycerolipids **17** with cationic 'head' separated from the glycerol backbone by methylene group.



Scheme 8. Synthesis of glycerolipids **18** with acetal type spacer.

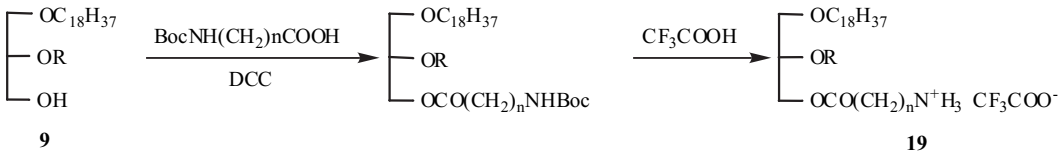
linked to the glycerol backbone via the acid-labile acetal bond (Scheme **8**) [35, 36].

We reported the synthesis of ether glycerolipids with acyl type spacers. Acylation of 1,2-dialkylglycerols **9** using long chain Boc-protected amino acids (followed by removal of the protective group) yielded a series of compounds **19** with the spacers of different length (Scheme **9**) [37].

In lipids **21** the spacer group is represented by single valerate residue. The initial 1,2-dialkylglycerols **9** were acylated by chloroanhydride of 5-bromovaleric acid. The cationic 'head' was introduced by quaternization of *N,N*-dimethylethanolamine with bromides **20** (Scheme **10**) [37].

Importantly, our cationic ether glycerolipids **17-21** vary in one particular structural domain, namely, in the substituents at C(2) atom of the glycerol backbone, cationic 'heads' and the length of the spacer group. Furthermore, these compounds have functional hydroxy, carboxy or amino groups in the polar domain. This allows for conjugating other molecules/atoms aiming at the increased cytotoxicity and/or delivery of spin probes [38], radioactive isotopes [39] and boronated compounds (in boron neutron capture therapy) [40] to the tumor.

To summarize the data on previously reported non-phosphorus anticancer ether glycerolipids, we group these compounds into four categories:

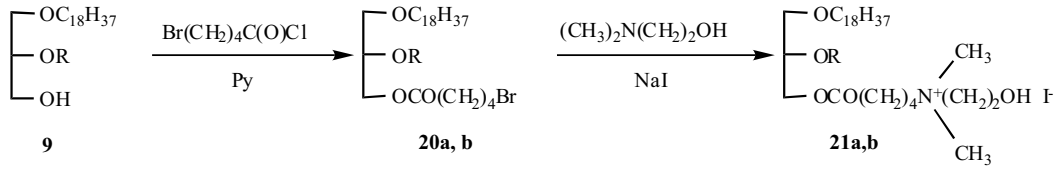


R = ethyl, allyl

n = 5, 10

DCC: *N,N*-dicyclohexylcarbodiimide

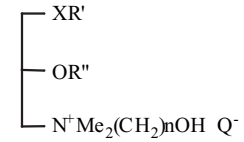
Scheme 9. Synthesis of positively charged lipids **19**.



R: **a** ethyl,
b allyl

Scheme 10. Reactions for obtaining lipids **21**.

Table 1. Cytotoxicity of Lipids with 'Reverted' Choline as a Cationic 'Head'



10, 22

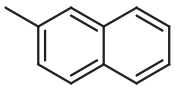
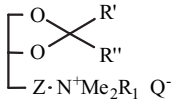
Compound	X	R'	R"	n	Q^-	IC_{50}, μM*	Cell line	Refs
10a	S	$C_{16}H_{33}$	Me	2	Br	4.66±0.27	HL-60	41, 42
10b	S	$C_{16}H_{33}$	Et	2	Br	3.72±0.05	HL-60	42
10c	S	$C_{16}H_{33}$	Et	3	Br	2.95±0.36	HL-60	42
10g	S	$C_{18}H_{37}$	Me	3	Br	3.55±0.22	HL-60	41, 42
10e	O		Me	2	Br	>10	HL-60	41, 42
10j	S	$C_{18}H_{37}$	H	2	I	0.35±0.05	KB	43
10t	S	$C_{16}H_{33}$	C_6H_{11}	2	Br	7.15	HL-60	32
22	S	$C_{16}H_{33}$	CH_2OMe (instead of OR")	3	Br	3.46	HL-60	33

Table 2. Cytotoxicity for HL-60 Leukemia Cells of Cationic Lipids Containing the Dioxalane Cycle [41, 42]



8a-c

Compound	R'	R"	R_1	Z	IC_{50}, μM
8a	$C_{17}H_{35}$	H	$(CH_2)_3OH$	-	6.47±0.24
8b	Me	Me	$(CH_2)_2OH$	-	>10
8c	$C_{17}H_{35}$	Me	Me	$O(CH_2)_4$	2.82±0.42

1 Lipids with cationic 'head' in the form of 'reverted' choline (Table **1**);

2 Dioxalane cycle containing cationic lipids (Table **2**);

3 Lipids with ammonium group linked directly to the glycerol backbone (Table **3**);

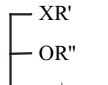
4 Lipids with cationic 'head' linked to the glycerol fragment via the spacer (Table **4**).

The majority of 'reverted' cholines (group 1) were less active than the compounds in groups 3 and 4 whose activities appeared to be comparable with that of reference compound ET-18-OMe (Table 4). The presence of hydroxy group in the cationic 'head' leads to somewhat lower cytotoxicity. The role of the heteroatom (S or O) at the position C(1) of glycerol has not yet been elucidated (compounds **10b** and **10h**; Table 3) [41]. The optimal lenght of the alkyl substituent at C(1) atom of glycerol is C_{14}–C_{19}. Shortening of this interval (compounds **8b;** Table 2 and **10o;** Table 3), as well as the replacement of alkyl for an aromatic system (compound **10e;** Table 1) resulted in decreased cytotoxicity. Limited rotational mobility of dioxalane cycle at C(1) and C(2) atoms of the glycerol fragment resulted in decreased potency (Table 2).

The presence of methoxy or ethoxy groups at C(2) does not influence the antitumor activity (compare the activities of compounds **10a** and **10b**; Table 1). Also, replacement of methoxy group for heterocyclic pyrimidine type base causes no significant loss of activity (**15a** vs **15d**; Table 4). In contrast, the alkyl substituent with 5 or more carbon atoms in its chain decreases the antitumor potency: compound **10s** was ~3-fold less potent than **10d**; Table 3).

The SAR analysis of compounds of groups 3 (**10l** vs **10m; 10p** vs **10h**; Table 3) and 4 (**13d** vs **13f; 15d** vs **15f**; Table 4) reveals that the cationic 'head' (e.g., with

Table 3. Cytotoxicity of Glycerolipids with Ammonium Group Directly Linked to Glycerol



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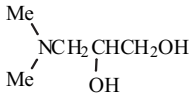
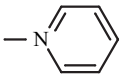
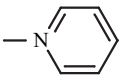
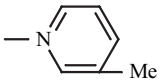
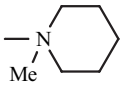
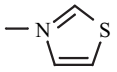
Compound	X	R'	R"	Y^+	Q^-	IC_{50}, μM	Refs
10d	S	$C_{16}H_{33}$	Me	NMe_3	Br	2.20±0.30	41
10h	O	$C_{16}H_{33}$	Me	NMe_3	Br	1.59±0.17	41
10i	O	$C_{16}H_{33}$	H (instead of OR')	NMe_3	Br	1.61±0.10	41
10k	S	$C_{16}H_{33}$	Me		Br	3.62±0.04	32
10l	O	$C_{16}H_{33}$	Et	NEt_3	Br	0.68±0.11	41
10m	O	$C_{16}H_{33}$	Et		Br	1.01±0.06	41
10n	O	$C_{16}H_{33}$	H (instead of OR')		Br	0.82±0.24	41
10o	O	C_8H_{17}	Me	NMe_3	Br	21.15±5.40	32
10p	O	$C_{16}H_{33}$	Me		Br	1.07±0.30	32
10q	O	$C_{18}H_{37}$	Et		Br	2.92	32
10r	S	$C_{16}H_{33}$	Me		Br	8.75	32
10s	S	$C_{16}H_{33}$	C_5H_{11}	NMe_3	Br	6.14	32

quaternary nitrogen) is a necessary prerequisite for cytotoxicity. No discernible difference in antitumor activity is observed for amines of aliphatic or heterocyclic type. However, compound **10r** (Table 3) with S atom in the polar 'head' appeared to be ~4-fold less potent. The reasons for this decrease remain to be elucidated.

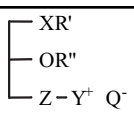
For the compounds of fourth series (Table 4) the growth inhibitory activity slightly decreased with the length of the spacer group (e.g., **13b** vs **13c**); in general, the potency of these lipids is similar to that of ET-18-OMe. The degree of oligomerization of ethyleneoxy group in compounds **15** plays no significant role in antitumor activity (compounds **15a** vs **15b**; Table 4).

It is worth noting that we herein provide general SAR data relevant to drug discovery within the above chemical classes. To unambigously evaluate the perspectiveness of the ether lipid, and to specify the efficacy of certain structural modifications in cytotoxicity, it is necessary to compare the congeners with structures as close as possible. From this viewpoint our approach to synthesize compounds **17-21** that differ in one single domain seems straightforward for addressing the role of individual structural units in cytotoxicity of antitumor ether glycerolipids. Even spatial isomers of certain ether lipids can have different activities, and cytotoxic potential frequently appears to depend on the cell type (see below).

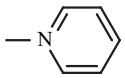
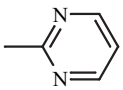
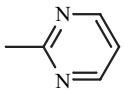
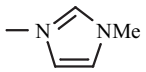
Unresolved Issues for Non-Phosphorus Cationic Glycerolipids: Lessons from Edelfosine

Antineoplastic activities of ether lipids with different configuration of the optical center vary for individual cell types [46]. The lifespan of mice with engrafted S-180 sarcoma cells was longer if animals were treated with the *sn*-3 isomer of ET-18-OMe compared with the *sn*-1 isomer, whereas the latter compound was more efficient against MM46 mammary gland carcinoma transplants [47]. For

Table 4. Cytotoxicity of Lipids with Cationic 'Head' Linked to Glycerol Via the Spacer



13 - 15, 23

Compound	R'	R"	Z	Y^+	Q^-	IC_{50}, μg/ml	Cell type	Refs
ET-18-OMe	$C_{18}H_{37}$	Me	$OPO_3^-(CH_2)_2$	NMe_3	-	2.5±0.3[a] 0.63	HL-60 KB 3-1	32, 33, 41
13a	$C_{16}H_{33}$	Me	$O(CH_2)_2$	NMe_3	Br	1.85±0.15[a]	HL-60	41
13b	$C_{16}H_{33}$	Me	$O(CH_2)_2$	NMe_3	Br	2.30±0.37	HL-60	41
13c	$C_{16}H_{33}$	Me	$O(CH_2)_4$	NMe_3	Br	3.79±0.13[a]	HL-60	41
13d	$C_{16}H_{33}$	Et	$O(CH_2)_4$	NMe_3	Br	1.86±0.04	HL-60	41
13f	$C_{16}H_{33}$	Et	$O(CH_2)_4$		Br	0.78±0.02	HL-60	41
13g	$C_{16}H_{33}$	H (instead of OR')	$O(CH_2)_4$	NMe_3	Br	1.56±0.10[a]	HL-60	41
13h	$C_{18}H_{37}$	Me	$O(CH_2)_2$	NMe_3	Cl	1,25 0.16	HL-60 KB 3-1	33
14	$C_{16}H_{33}$	Me	$S(CH_2)_2$	NMe_3	Br	2.4	HL-60	32
15a	$C_{18}H_{37}$	Me	$(OCH_2CH_2)_2$	NMe_3	Cl	1,25 0.32	HL-60 KB 3-1	33
15b	$C_{18}H_{37}$	Me	$(OCH_2CH_2)_5$	NMe_3	Cl	1,25 0.16	HL-60 KB 3-1	33
15c	$C_{18}H_{37}$	C_4H_9	$(OCH_2CH_2)_2$	NMe_3	Cl	2.5	HL-60	33, 44
15d	$C_{18}H_{37}$		$(OCH_2CH_2)_2$	NMe_3	Cl	0.31 0.32	HL-60 KB 3-1	33, 44, 45
15e	$C_{18}H_{37}$	CH_2CF_3	$(OCH_2CH_2)_2$	NMe_3	Cl	2.5	HL-60	33, 44
15f	$C_{18}H_{37}$		$(OCH_2CH_2)_2$		Cl	0.63	HL-60	33, 44
23	$C_{18}H_{37}$	$COCH_2Ac$	$(OCH_2CH_2)_2$	NMe_3	Cl	20.0	HL-60	33, 44

[a]μM

cultured HL-60 leukemia cells the *sn*-3 isomer of ET-18-OMe was the most potent, the *sn*-1 isomer was the least cytotoxic, and the racemic mixture of both compounds displayed an intermediate activity [48]. Also, the optical isomers BM 41.440 also showed differential potency depending on cell type [49]. It is plausible to suggest that the cytotoxicity of optical isomers of non-phosphorus ether lipids would vary; this, in turn, presumes that such isomers should be tested in broad number of biological models.

This requirement is further substantiated by differential sensitivity of individual tumor to ether lipids [42, 50]. Moreover, the activity of ether lipids for cultured cells may or may not be paralleled by the potency in animal studies. This reiterates the necessity of properly matched cell culture/animal models. On the other hand, search for the compound(s) with increased toxicity should be carried out cautiously keeping in mind that these lipids can evoke non-selective killing of normal cells.

Clinical use of ether lipids is frequently hampered by their side effects, predominantly by hemolytic activity. At the concentrations only slightly above the therapeutic range, ET-18-OMe evoked a significant damage of erythrocytes (a 50% hemolysis at 16 μM ET-18-OMe) [51]. This drawback is supposed to be overcome by incorporating ether lipids into the liposomes, with dioleylphosphatidylethanolamine, cholesterol and dioleylphosphatidylcholine as helper lipids of complementary molecular shape [52, 53]. The liposomes containing cholesterol and ET-18-OMe proved to be particularly stable and demonstrated relatively low hemolytic activity; a 50% hemolysis was achieved only with 661 μM ET-18-OMe [53]. These considerations imply that liposomal forms of non-phosphorus ether lipids might be devoid of hemolytic side effects.

Non-Phosphorus Cationic Glycerolipids are Potent for Tumor Cells with Overexpressed Lipid Microdomains

Considering the unique mechanisms of anticancer effects of ET-18-OMe and structurally related compounds, namely, their ability to target lipid microdomains and disrupt its constituents (see Introduction), the non-phosphorus cationic glycerolipids could be potent for tumor cells with well developed rafts. Multidrug resistance (MDR), a major obstacle for therapeutic success in cancer patients, is frequently associated with overexpression of lipid microdomains [54, 55]. It has been demonstrated that a key molecule that mediates the resistant phenotype(s), the efflux pump P-glycoprotein, is localized in these compartments [56]. We hypothesized that cationic glycerolipids might efficiently kill MDR tumor cells. The MCF-7Dox subline selected from MCF-7 human breast carcinoma cell line for survival in the presence of antitumor drug adriamycin was resistant to this agent (fold resistance 9.9 compared to the parental cells) and to P-glycoprotein transported drugs taxol (fold resistance 72.5), vincristine (14.2) and mitoxantrone (54.1) [57]. The MCF-7Dox cells expressed P-glycoprotein that was co-localized with lipid microdomains (A.Shtil, unpublished). Of note, in the parental cells (and therefore in MCF-7Dox subline) caspase 3, a critical effector of many apoptotic pathways, is not expressed [58]. Furthermore, in MCF7Dox cells the expression of pro-apoptotic $\alpha v\beta 3$ integrin is down-regulated, whereas collagenases are markedly activated, and the invasiveness of extracellular matrix is increased. These cells are resistant to anchorage dependent apoptosis (anoikis) [57]. However, our non-phosphorus lipid **21a** (with 'reverted' choline as a cationic 'head') was equally cytotoxic for MCF-7 and MCF-7Dox cells ($IC_{50} \sim 4$ μM within the initial 24 h of exposure) (our unpublished observations). These results show that non-phosphorus ether lipids are active against highly malignant cells otherwise resistant to a variety of stress stimuli including anticancer drugs. Definitely, more studies are necessary to answer the question of whether lipid rafts are targeted by non-phosphorus cationic glycerolipids. If these compounds retain an important therapeutic advantage of ether lipids, i.e., the capability to selectively eliminate tumor cells, and if they prove the potency for MDR tumors, the non-phosphorus cationic glycerolipids would emerge as attractive candidates for cancer therapy.

CONCLUDING REMARKS

Anticancer ether lipids possess several advantages for clinical use: 1) they target mostly tumor cells while non-malignant counterparts remain spared; 2) ether lipids may serve as carriers of other therapeutically valuable molecules/atoms to the tumor site; 3) the mechanism of cytotoxicity involves the interference of ether lipids with rafts where many intermediates crucial for cell viability are localized; 4) ether lipids do not interact with DNA and thereby cause no mutagenic side effect, a drawback of many conventional chemotherapeutics that limits their use in repetitive courses of treatment. In particular, the non-phosphorus cationic glycerolipids as a class of anticancer agents deserve further investigation. Indeed, the synthesis of these agents has been developed, and this procedure allows to omit the reactions of incorporation of phosphorus containing groups. Importantly, cellular effects of these compounds are long-lasting due to stability in the presence of phospholipases. The results of testing in cell culture and animal models, and the initial data on the ability to kill pleiotropically resistant tumor cells, provide strong basis for the perspective of non-phosphorus cationic glycerolipids in anticancer drug development.

ACKNOWLEDGEMENTS

We are grateful to Dr. M. Maslov for helpful discussions. This study was supported by grants of the Russian Foundation for Basic Research (96-03-33383-a, 01-03-332334-a, 03-03-32482-a and 04-03-32452-a), Russian Federation President's grant for support of leading national scientific schools (MK-2134.2004.3; NSh-2013.2003.3), International Science and Technical Committee (1813p), and Scientific University Priority Research Program (203.05.04.005). N.V.P. and A.A.S. equally contributed to this article.

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Received: June 9, 2005 **Revised: July 30, 2005** **Accepted: August 1, 2005**