Pergamon

Bioorganic & Medicinal Chemistry, Vol. 5, No. 8, pp 1469–1479, 1997

PII: S0968-0896(97)00097-7
0968-0896/97 $17.00 + 0.00

Antitumor Agents—CLXVII.† Synthesis and Structure–Activity Correlations of the Cytotoxic Anthraquinone 1,4-Bis-(2,3-Epoxypropylamino)-9,10-Anthracenedione, and of Related Compounds

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Abstract—1,4-Bis-(2,3-epoxypropylamino)-9,10-anthracenedione (**3**) was synthesized in this laboratory and was found to be a potent antitumor agent. Derivatives of this compound containing anthraquinone, naphthoquinone, and quinone skeletons were also prepared and evaluated for in vitro cytotoxic activity in several cell lines. These molecules were designed as bifunctional antitumor agents with the potential to act as (1) intercalating agents due to their planar backbones, and (2) alkylating agents due to the presence of alkylating moieties in their side chains. Compounds with an anthraquinone skeleton and propylamino side chains containing epoxides or halohydrins as the alkylating species showed greater activity than similar compounds with naphthoquinone or quinone skeletons. Compounds without these alkylating functionalities (e.g., with alkene or amino groups) were generally inactive. Hydroxy substitution on the planar skeleton in conjunction with alkylating side chains gave compounds with the most potent cytotoxic activity. The position of the hydroxy groups and side chains could be varied without substantially affecting activity. Activity was retained when an epoxypropyloxy side chain was substituted for the epoxypropylamino side chain in the parent compound. © 1997 Elsevier Science Ltd.

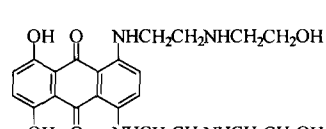
Introduction

Cancer chemotherapy continues to be an important research avenue. Combination chemotherapy using antineoplastic agents with different mechanisms of action is one method employed to combat this disease. Thus, a single molecule containing two functional groups, each with a different mechanism of action, also could prove beneficial in cancer treatment.

Intercalating agents represent one class of antineoplastic agents. Such compounds contain a planar chromophore that inserts between two base pairs in the DNA helix causing a local untwisting of the helix resulting in miscoding and possible cell death.[2] Mitoxantrone,[3–10] (Fig. 1) which contains a planar anthraquinone skeleton, is a clinically useful antineoplastic agent. Other anthraquinone-type analogues, such as bisantrene,[11–13] chrysophanol, and emodine,[14,15] also have shown significant in vivo antineoplastic activity.

Alkylating agents (e.g., cyclophosphamide and busulfan) are a second class of antineoplastic drugs. This large, diverse group of compounds contains reactive groups that are capable of covalently modifying a variety of biological molecules.[16] Teroxirone (Fig. 1) is a 1,3,5 triazine, which contains alkylating epoxide moieties in its amino side chains; this compound has been reported to exhibit antineoplastic activity.[17]

In order to combine features of both intercalating and alkylating agents, the known anthraquinone 1,4-bis-(2,3-epoxypropylamino)-9,10-anthracenedione (com-



Mitoxantrone

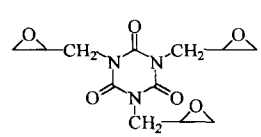

Teroxirone

Figure 1.

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†For Part 166, see ref 1.

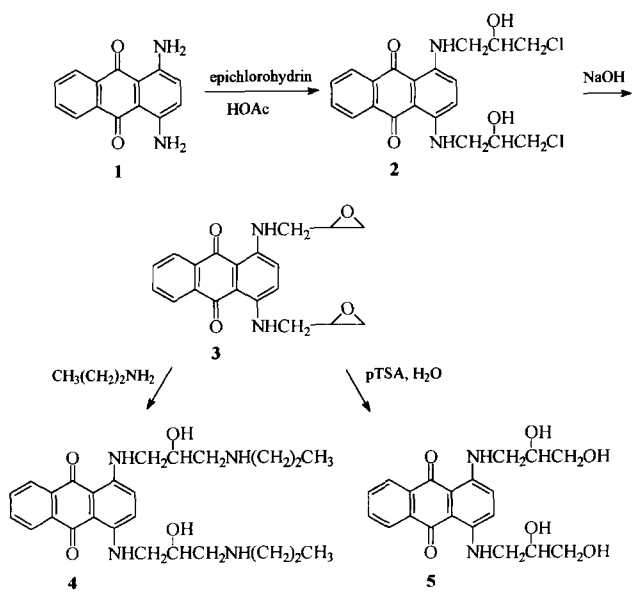
pound **3**, Scheme 1) was synthesized in this laboratory. This substituted anthraquinone contains the planar skeleton and diamino side chain substitution pattern of mitoxantrone and the alkylating epoxide moiety of teroxirone. Compound **3** showed significant and selective activity in preliminary in vitro studies with an ED_{50} of less than 40 ng/mL against human epidermoid carcinoma (KB cells). Based on this promising result, a structure–activity relationship (SAR) study was implemented to explore the importance of: (1) the epoxide moiety, (2) the number and position of the side chains, (3) the addition of hydroxy groups to the planar anthraquinone skeleton, (4) the length of the alkylamino group, (5) the type of planar backbone, and (6) the substitution of alkoxy for alkylamino side chains.

Chemistry

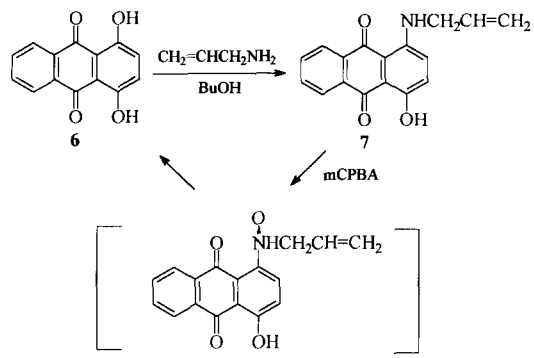
The anthraquionone series

Scheme 1 shows the synthesis of the parent compound (**3**) and related anthraquinones. Reaction of 1,4-diaminoanthraquinone (**1**) and epichlorohydrin under acidic conditions gave the bis-propylaminohalohydrin (**2**). Compound **2** under basic conditions was converted to the bis-propylaminoepoxide (**3**). In order to explore the importance of the epoxide, two additional compounds (**4** and **5**) were synthesized as shown in Scheme 1. The basicity of the molecule was increased in derivative (**4**), which was prepared via nucleophilic attack of propylamine on **3**. The more hydrophilic diol (**5**) was prepared by treatment of **3** with *p*-toluenesulfonic acid.[18,19]

Analogue **7** with only one amino side chain was synthesized by the method of Zee-Cheng et al.[20] (Scheme 2). However, attempted epoxidation of **7** with



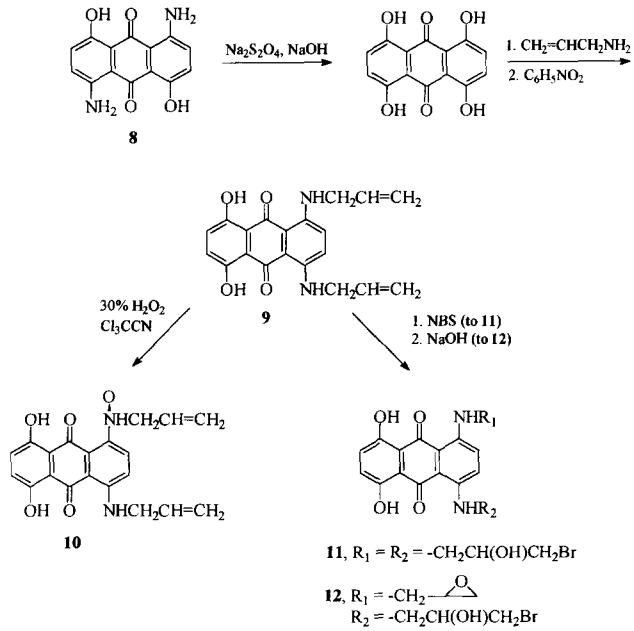
Scheme 1.



Scheme 2.

m-chloroperbenzoic acid was unsuccessful as the starting 1,4-dihydroxyanthraquinone (**6**) was reformed, presumably through formation of the *N*-oxide and subsequent rearrangement (vide infra).

The 5,8-dihydroxylated analogues of the parent compound were synthesized following the methods of Marshall[21] and Murdock et al.[22] as shown in Scheme 3. It was anticipated that the addition of hydroxy groups on the anthraquinone ring would enhance binding and activity.[22] Oxidation of 1,5-diamino-4,8-dihydroxyanthraquinone (**8**) with sodium thiosulfate gave leuco-1,4,5,8-tetrahydroxyanthraquinone.[21] Treatment of the latter compound with allylamine followed by air oxidation[22] afforded the desired 5,8-dihydroxy-1,4-dipropyleneaminoanthraquinone (**9**). Treatment of **9** with trichloroacetonitrile and 30% hydrogen peroxide afforded the *N*-oxide derivative (**10**) instead of the expected diepoxide. However, both alkene groups were converted to halohydrins in compound **11** by treatment of **9** with *N*-bromosuccinimide.[23–25] Analogue **11** was then converted to the monoepoxide (**12**) by treatment



Scheme 3.

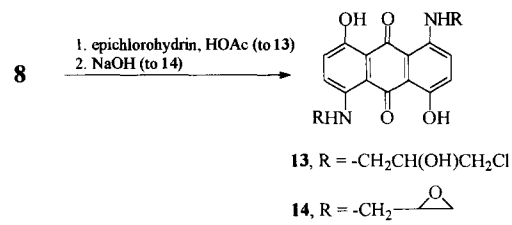
with base. The *bis*-epoxide was not formed in the reaction.

The 1,5-disubstituted series was synthesized (Scheme 4) in order to explore the spatial requirement for activity. The dihalohydrin (**13**) was formed from 1,5-diamino-4,8-dihydroxyanthraquinone (**8**), then was treated with base to give the expected diepoxide (**14**).

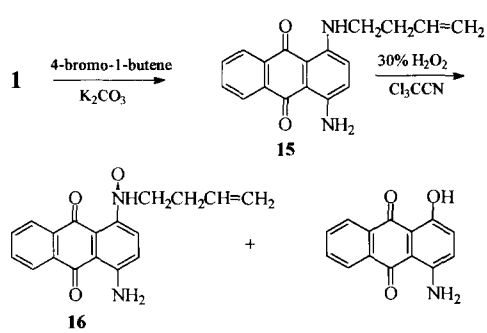
The length of the alkyl side chain was extended from three to four carbons as shown in Scheme 5. The monoalkylated species (**15**) was formed in the reaction between 1,4-diaminoanthraquinone (**1**) and 4-bromo-1-butene in the presence of base. In a manner analogous to the oxidation of **7**, when treated with trichloroacetonitrile and 30% hydrogen peroxide, **15** afforded a 1:1 mixture of the *N*-oxide (**16**) and 1-hydroxy-4-aminoanthraquinone. Heating the reaction mixture also failed to give epoxide and resulted in a total conversion of **16** to the latter compound.

The naphthoquinone series

The naphthoquinone backbone modification reduces the size of the planar region, alters the spatial distance, and changes the reactivity of the nitrogen from aniline-like to a vinylogous amide. The syntheses of the epoxide and halohydrin naphthoquinones **18** and **19** are shown in Scheme 6. Reaction of 2-amino-3-chloronaphthoquinone and 3-bromo-1-propene under basic conditions afforded 2-(2,3-propyleneamino)-3-chloronaphthoquinone (**17**).[26] The desired epoxide (**18**) was then obtained by oxidation of **17**.[26] The halohydrin naphthoquinone (**19**) was synthesized by reaction of the 2-amino-3-chloronaphthoquinone and an excess of epichlorohydrin in acetic acid. The resulting halohydrin



Scheme 4.



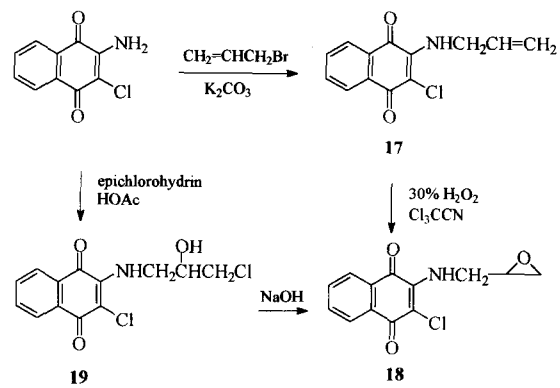
Scheme 5.

(**19**) was converted to the epoxide (**18**) by treatment with sodium hydroxide in methanol.

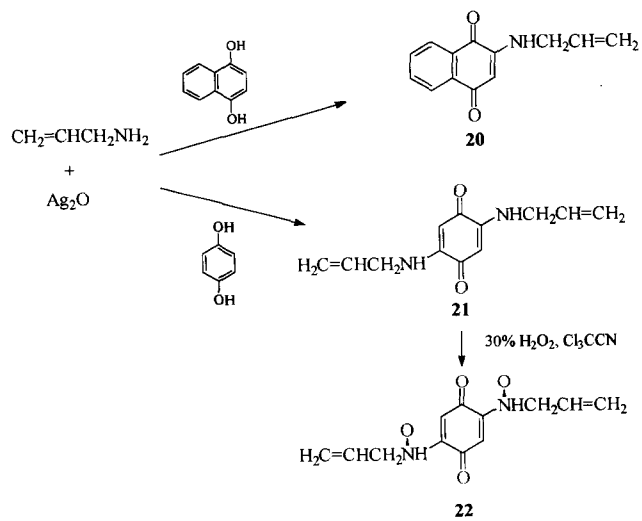
Synthesis of a 2,3-disubstituted naphthoquinone was attempted via displacement of the chloride in **17** with an amine. However, displacement using ammonium hydroxide either neat or in solution was unsuccessful. Liquid ammonia with and without sodium amide also failed to give the desired product.[27–29] The nonchlorinated derivative (**20**) of **17** was synthesized via reaction of 1,4-dihydroxynaphthalene with silver oxide and allylamine (Scheme 7). Based on preliminary biological results (Table 1) for compounds **17–19**, the epoxide derivative of **20** was not synthesized.

The quinone series

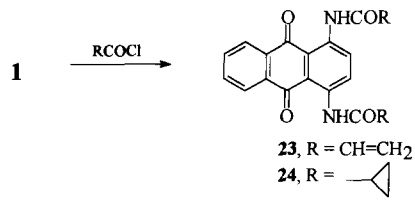
A few derivatives containing only the quinone backbone were prepared. As shown in Scheme 7, reaction of 1,4-hydroquinone with allylamine yielded **21**. Attempts to form the epoxide from the alkene using either trichloroacetonitrile and 30% hydrogen peroxide or *m*-chloroperbenzoic acid resulted in the formation of the *N*-oxide **22**.[22] Attempted reduction of the quinone (**21**) to the dihydroxy species with sodium borohydride



Scheme 6.



Scheme 7.

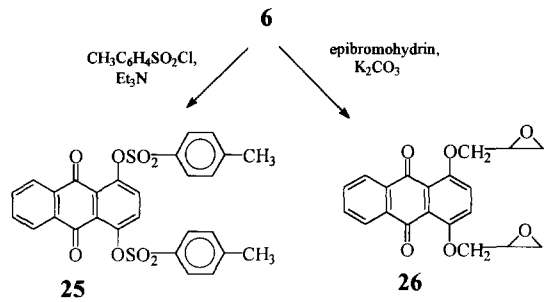


Scheme 8.

gave only starting material. Based on preliminary biological data (Table 1), emphasis was placed on the anthraquinone backbone and no additional quinone analogues were synthesized.

Additional analogues

Seven related compounds were also synthesized after the initial SAR study. Reaction of 1,4-diaminoanthraquinone (**1**) with acryloyl chloride or cyclopropanecarbonyl chloride under basic conditions gave the *bis*-substituted amido anthraquinones **23** and **24**, respectively (Scheme 8). As shown in Scheme 9, reaction of 1,4-dihydroxyanthraquinone (**6**) with *p*-toluenesulfonyl chloride gave a *bis*-sulfonic ester, **25**, while reaction of **6** with epibromohydrin gave **26**, the *bis*-epoxypropyloxy analogue of **3**. Reaction of epibromohydrin with 1-aminoanthraquinone (Scheme 10) and with 1,4-dihydroxynaphthalene (Scheme 10) gave two further derivatives of **3**: the mono-epoxypropylamino anthraquinone **27** and the *bis*-epoxypropyloxy naphthalene **28**. A final compound, naphthoquinone **29**, contains cyclopropylmethylamino functionalities and was prepared in two steps from 5,8-dihydroxy-1,4-naphthoquinone: reduction with $SnCl_2$ followed by reaction with cyclopropylmethylamine (Scheme 11).



Scheme 9.

Results and Discussion

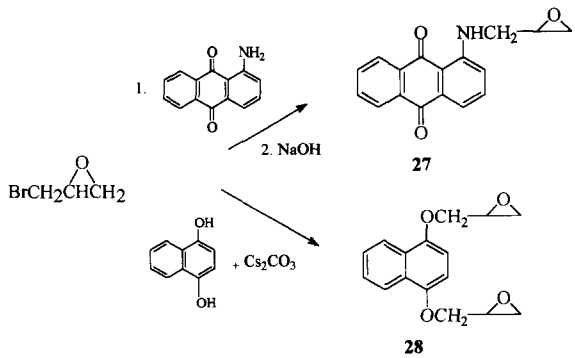
The initial 22 compounds were tested in five in vitro cytotoxicity screening assays; L1210, P388, A549, HCT-8, and KB. The results are shown in Table 1. ED_{50} values greater than 4 μg/mL are considered inactive. Several structure–activity relationships can be found from this data.

Compounds without any alkylamino side chains [1,4-diaminoanthraquinone (**1**), 1,4-dihyroxyanthraquinone (**6**), and 1,5-diamino-4, 8-dihydroxyanthraquinone (**8**)] were inactive (with the exception of the ED_{50} of 1.20 μg/mL shown by compound **1** in the KB assay). The two anthraquinone compounds containing secondary amine

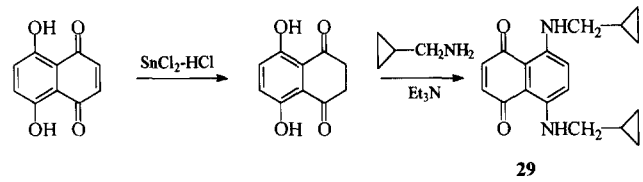
Table 1. Cytotoxicity results in L1210, P388, A549, HCT-8, and KB cell lines

	Cytotoxicity ED_{50} (μg/mL)[a]				
Compd (Scheme #)	**L 1210**	**P 388**	**A 549**	**HCT-8**	**KB**
1 (1)	>10	>10	6.93	8.75	1.20
2 (1)	>10	4.91	1.00	9.79	8.25
3 (1)	5.10	4.40	>10	3.30	<0.10
4 (1)	>10	8.00	7.80	9.40	4.40
5 (1)	>10	6.60	>10	>10	>10
6 (2)	>10	5.13	6.91	6.92	>10
7 (2)	3.10	0.51	>10	>10	>10
8 (3)	>10	>10	>10	>10	6.40
9 (3)	>10	5.50	>10	>10	>10
10 (3)	5.80	5.55	>10	5.97	6.19
11 (3)	0.44	0.42	>10	0.75	5.50
12 (3)	0.52	<0.10	0.10	0.62	0.34
13 (4)	0.52	0.42	0.70	0.61	0.46
14 (4)	0.44	0.49	0.55	0.56	0.32
15 (5)	>10	>10	>10	>10	>10
16 (5)	5.10	5.41	3.84	4.64	4.38
17 (6)	>10	6.90	5.50	4.60	>10
18 (6)	5.50	4.40	>10	4.89	—
19 (6)	3.90	0.72	4.80	3.50	4.20
20 (7)	6.50	6.00	5.50	2.70	>10
21 (7)	>10	>10	>10	>10	>10
22 (7)	0.92	4.08	>10	6.40	—

[a]Mitoxantrone has an ED_{50} of 0.25 g/mL in the 1210 assay.



Scheme 10.



Scheme 11.

(4) and diol (5) moieties in the side chains also were inactive in all assays.

Although an olefin is a potential reactive site, its presence alone does not assure cytotoxicity as most analogues with a propyleneamino functional group (**9**, **10**, **17**, **21**) were relatively inactive as were both compounds with a butyleneamino group (**15** and **16**). However, while the 1,4-*bis*-propyleneamino anthraquinone **9** was inactive in all assays, the monosubstituted compound **7** [1-(2,3-propyleneamino)-4-hydroxy-9,10-anthracenedione] showed high activity (ED_{50} = 0.51 μg/mL) in the P388 assay and some activity in the L1210 assay (ED_{50} = 3.10 μg/mL). A related monosubstituted naphthoquinone, 2-(2,3-propyleneamino)-1,4-naphthoquinone, **20**, was moderately cytotoxic (ED_{50} = 2.70 μg/mL) in the HCT-8 assay. Compound **22**, the *N*-oxide analogue of the inactive *bis*-(2,3-propylene-amino)quinone (**21**), was very selective and active (ED_{50} = 0.92 μg/mL) in the L1210 assay.

The majority of active compounds contained either an epoxide (**3**, **12**, **14**), which is a very reactive alkylating group, or a halohydrin (**2**, **11**, **13**, **19**), which can either serve as an alkylating species itself or may be transformed in vivo to an epoxide. Compound **3**, [1,4-*bis*-(2,3-epoxypropylamino)-9,10-anthraquinone], the parent compound for this SAR study, exhibited the highest activity in the KB assay (ED_{50} < 0.10 μg/mL) and was also selective for this cell line in the preliminary testing. Its synthetic precursor, *bis*-halohydrin **2**, showed selective cytotoxicity in the A549 assay (ED_{50} = 1.00 μg/mL).

Four active anthraquinones (**11**, **12**, **13**, and **14**) contain two hydroxy groups on the planar skeleton in addition to the alkylating groups in the side chains. Compounds **11** and **12** have the amino side chains on the same ring and are 1,4-dialkylamino-5,8-dihydroxy substituted, while compounds **13** and **14** have the amino groups on different rings and are 1,5-dialkylamino-4,8-dihydroxy substituted. Both substituent patterns retained high activity. Compound **11**, a *bis*-halohydrin, showed good activity in the murine (P388, L1210) and human colon (HCT-8) cell lines. Conversion of one halohydrin group to an epoxide gives **12**, which was active in all five cell lines and very active in the P388 assay (ED_{50} < 0.10 μg/mL). The 1,5-dialkylamino-4,8-dihydroxy compounds (**13** and **14**) were equipotent in all five assays (ED_{50} values from 0.32 to 0.70 μg/mL); neither the halohydrin (**13**) nor epoxide (**14**) showed any selectivity.

Interestingly, the most active alkene compound, **7**, also contains a hydroxy group on the anthraquinone skeleton. Furthermore, comparison of compound **2** (a *bis*-chlorohydrin without hydroxy groups on the anthraquinone skeleton) and **11** (a 5,8-dihydroxy-anthraquinone-*bis*-bromohydrin) shows greatly increased cytotoxicity in three cell lines with the latter compound (**11**) but the selectivity found in the A549 cell line with the non-hydroxylated compound (**2**) is lost. Thus, the presence of hydroxy groups may cause increased activity but also may lead to decreased selectivity. Preparation of the non-hydroxylated analogues of the 1,5-dialkylamino-4,8-dihydroxy compounds, **13** and **14**, would be useful to further identify the importance of the hydroxy groups or substitution pattern as would preparation of the diepoxide analogue of **12**. The hydroxy groups might act as additional binding sites or might align the side chains in a specific orientation for optimal activity. The hydroxy groups might also create the optimum balance between hydrophilicity and lipophilicity necessary for the transport of compounds across the cellular membrane. The SAR of substituents other than hydroxy groups has not yet been investigated.

Comparison of the planar skeletons (quinone, naphthoquinone, anthraquinone) shows, in general, more activity with compounds containing the anthraquinone backbone. For example, all anthraquinone derivatives containing epoxides or halohydrins showed activity; however, with a naphthoquinone backbone, the one epoxide-containing compound, **18**, was inactive in all assays and its halohydrin derivative, **19**, showed good activity only in the P388 assay (ED_{50} = 0.72 μg/mL). A comparison of alkene compounds with different backbones (**21**-quinone, **20**-naphthoquinone, and **7**-anthraquinone) showed no activity for the quinone, moderate activity in one assay (HCT-8) for the naphthoquinone, and good activity in one assay (P388) and moderate activity in a second assay (L1210) for the anthraquinone. Activity increased as the planar skeleton enlarged.

After the initial screening, the parent compound (**3**) and its secondary amino and diol analogues (**4** and **5**, respectively), together with seven related compounds (**23–29**) were evaluated by the National Cancer Institute (NCI) in its human tumor cell line cytotoxicity panel. As in the initial screening, the secondary amine and diol

substituted anthraquinones (**4** and **5**) showed no activity in this extended screening. The two amido (**23** and **24**) and the sulfonic ester (**25**) substituted anthraquinones and the cyclopropylmethylamino napthoquinone (**29**) also were inactive. Compounds **3**, **26**, **27**, and **28** did show activity in this screening, and their data are shown in Table 2 as log GI_{50} values (GI_{50} = molar concentration required to inhibit 50% of cell growth). Data are reported only when the value in that cell line is less than the mean value (median log GI_{50}) over all cell lines tested.

The parent naphthoquinone, **3**, the bis-epoxypropylamino substituted compound, showed good activity in several cell types and was extremely active (log $GI_{50} < -8$) in many leukemia cell lines and in one melanoma (LOX1MVI) cell line. Its monosubstituted analogue (**27**) was less active than the disubstituted compound; however, compound **27** was not isolated in pure form. Compound **26**, a bio-isostere of compound **3** with an oxygen replacing the amine in the side chains, showed similar levels of activity to those of **3**. Deletion of the middle quinone ring from **26** gives the bis-epoxypropyloxy naphthalene **28**, which was somewhat less active than the anthraquinone. Hydroxylation of these derivatives would be an interesting future SAR study.

Conclusions

Derivatives of 1,4-bis-(2,3-epoxypropylamino)-9,10-anthracenedione (**3**) containing alkene, epoxide, halohydrin, diol, and secondary amine functional groups in the alkylamino side chains and with quinone or naphthoquinone skeletons have been prepared and tested for in vitro antineoplastic activity. Results showed that, in general, analogues with no alkyl side chains, alkene, secondary amines, diols, or side chains containing four instead of three carbons were less cytotoxic, while compounds containing alkylating epoxide or halohydrin moieties exhibited greater activity. While hydroxylation of the planar skeleton may increase cytotoxicity, it may also decrease the

Table 2. Inhibition of in vitro cancer cell growth

	Cytotoxicity log GI_{50} (M)[a]					Cytotoxicity log GI_{50} (M)[a]			
Cell line	**3**	**26**	**27**	**28**	**Cell line**	**3**	**26**	**27**	**28**
Leukemia					***Melanoma***				
CCRF-CEM	−8.00	−8.00	−6.40	−6.57	LOX1MVI	−8.00	−7.56	−6.26	−6.29
HL-60 (TB)	−8.00	−7.34	−5.72	−6.49	MALME-3M				
K-562	−6.90	−6.33	−5.64	−5.70	M 14	−6.20	−6.19	−5.22	
MOLT-4	−8.00	−7.81	−6.17	−6.45	SK-MEL-28				
RPMI-8226				−5.52	SK-MEL-5				
SR	−7.60	−7.19	−5.63	−6.46	UACC-257				
Non-Small Cell Lung Cancer					UACC-62	−6.17	−6.00	−5.15	
A549/ATCC					***Ovarian Cancer***				
EKVX			−5.19		IGROV1			−5.25	
HOP-62		−6.03		−5.52	OVCAR-3		−6.11	−5.40	−5.86
HOP-92					OVCAR-4				
NCI-H226					OVCAR-5				
NCI-H23	−6.10	−6.10	−5.47	−5.60	OVCAR-8	−6.16	−6.14	−5.65	−5.66
NCI-H322M					SK-OV-3				
NCI-H460		−6.23			***Renal Cancer***				
NCI-H522					786-O	−6.14	−6.28	−5.37	−5.68
Colon Cancer					A 498				
COLO-205					ACHN	−6.66	−6.46	−5.53	−5.72
HCT-2998					CAKI-1	−5.99		−5.32	−5.76
HCT-116	−6.77	−6.29	−5.77	−5.87	RXF-393			−5.64	−5.58
HCT-15			−5.14		SN 12 C	−6.18	−6.34	−5.54	−5.65
HT-29					TK-10				
KM12			−5.24		UO-31			−5.60	
SW-620		−6.31	−5.74	−5.90	***Prostate Cancer***				
CNS Cancer					PC-3			−5.29	
SF 268	−7.06	−6.52	−5.55	−6.24	DU-145	−5.63		−6.34	
SF 295				−5.54	***Breast Cancer***				
SF 539				−5.66	MCF 7	−7.30	−6.87	−6.88	−6.05
SNB-19					MCF7/ADR-RES			−5.30	
SNB-75					MDA-MB-231/ATCC				
U 251			−5.26	−5.63	HS 578 T				
					MDA-MB-435			−5.51	−5.61
					MDA-N			−5.47	−5.55
					HT-549				
					T-47 D				−5.72

[a]Compounds **4**, **5**, **23–25**, and **29** were also tested in these cell lines. These compounds were inactive in all cell lines with GI_{50} values greater than 10^{-4} M.

selectivity found with the non-hydroxylated epoxypropylamino (**3**) and epoxypropyloxy (**26**) compounds. Emphasis remains on the parent structure with optimization of the nature and pattern of substitution as a future development goal. Coupling synthetic efforts with molecular modeling techniques could aid in future analogue design.

The compounds in this study may be bifunctional cytotoxic agents. They contain both a planar intercalating-type functional group and alkylating-type epoxide and halohydrin side chains. Therefore, the compounds may intercalate into the DNA helix while the alkylating moieties react with nearby macromolecules. Together, the two active species in the compound may disrupt the normal functional processes of the tumor cell. The preliminary data presented in this study shows that bifunctional anthraquinones can show potent and selective cytotoxicity. Their potential as antineoplastic agents would require further mechanistic and in vivo studies.

Experimental

All melting points were taken on a Fisher–Johns melting point appartus and are uncorrected. Infrared spectra were recorded on a Perkin–Elmer 1320 Infrared spectrophotometer and are reported as cm^{-1}. Proton NMR spectra were obtained on either a Bruker-300 or a Varian 400 MHz NMR spectrometer, and all chemical shifts are reported in ppm from the internal standard TMS. Carbon-13 spectra were obtained on a Varian 400 MHz NMR spectrometer, and all chemical shifts are reported in ppm. Elemental analyses were performed by Atlantic Microlabs Norcross, Georgia or by Robertson Laboratories, Madison, New Jersey. Mass spectral analyses were determined on a V.G. Analytical Incorporation High Resolution 70F mass spectrometer or on a A.E.I. MS-90 mass spectrometer. Analytical thin-layer chromatography (TLC) was carried out on Analtech precoated silica gel GF plates. Universal Adsorbents silica gel (32–63 mesh) was used for both gravity and flash column chromatography. All new target compounds were characterized by melting point, ^1H NMR and IR spectral data, elemental and/or mass spectrometric analysis and, for some compounds, ^{13}C NMR spectrometry.

1,4-Bis-(3-chloro-2-hydroxypropylamino)-9,10-anthracenedione (2). To 10.0 g (0.042 mol) of 1,4-diaminoanthraquinone (**1**) in 200 mL of HOAc was added 84.5 mL (1.1 mol) of epichlorohydrin. The solution was stirred for 30 min at 90 °C and the solvent was removed under reduced pressure. The product was purified by silica gel flash chromatography (eluant: CH_2Cl_2:MeOH, 25:1) and recrystallized from CH_2Cl_2–Et_2O-hexanes to give 12.5 g (70%) of pure blue solid with mp 167–169 °C; ^1H NMR ($CDCl_3$) δ 3.21 (b, 2H, OH), 3.43 (m, 4H, CH_2), 3.51 (m, 2H, CH), 3.75 (d, J = 7.0 Hz, 4H, CH_2), 4.19 (2H, NH), 7.03 (s, 2H, ArH-2 and -3), 7.69 (dd, J = 3.2 and 5.5 Hz, 2H, ArH-6 and -7), 8.27 (dd, J = 3.2 and 5.5 Hz, 2H, ArH-5 and -8); IR (cm^{-1}) 3350, 1640, 1570, 1250, 1170, 1010, and 720. Analysis ($C_{20}H_{20}N_2O_4Cl_2 \cdot 0.25\ H_2O$) C, H, N.

1,4-Bis-(2,3-epoxypropylamino)-9,10-anthracenedione (3). To 8.3 g (0.02 mol) of **2** in 500 mL of MeOH at 60 °C was added 3.5 g of NaOH. The blue solution was stirred at room temperature for 4 h. The MeOH was removed under reduced pressure and the product purified by silica gel flash chromatography (eluant: CH_2Cl_2:MeOH, 25:1). Recrystallization from CH_2Cl_2-hexanes yielded 6.2 g (90%) of product melting at 189–190 °C; ^1H NMR ($CDCl_3$) δ 2.74 (m, 2H, CH_2), 2.87 (m, 2H, CH_2), 3.26 (m, 2H, CH), 3.57 (m, 2H, CH_2), 3.78 (m, 2H, CH_2), 7.32 (s, 2H, ArH-2 and -3), 7.71 (dd, J = 3.2 and 5.5 Hz, 2H, ArH-6 and -7), 8.34 (dd, J = 3.2 and 5.5 Hz, 2H, ArH-5 and -8), 10.76 (s, 2H, NH); IR (cm^{-1}) 3400, 3070, 2910, 1640, 1540, 1510, 1220, 1010, 900, and 730. Analysis ($C_{20}H_{18}N_2O_4 \cdot 0.25\ H_2O$) C, H, N.

1,4-Bis-(3-propylamino-2-hydroxypropylamino)-9,10-anthracenedione (4). To a stirring solution of 1.0 mL (12.2 mmol) of propylamine and 10 mL CH_2Cl_2 was added 200 mg (0.6 mmol) of **3**.[30,31] The purple solution was refluxed for 4.5 h, washed with H_2O, and dried over $MgSO_4$. The $CHCl_3$ was removed under reduced pressure and the product purified by silica gel flash chromatography (eluant: CH_2Cl_2:MeOH:NH_4OH, 9:1:2) to yield 173 mg (65%) of product with a mp of 163–164 °C; ^1H NMR ($CDCl_3$) δ 1.14 (t, J = 7.4 Hz, 6H, CH_3), 1.4 (b, 2H, NH), 1.7 (m, 4H, CH_2), 2.75 (m, 4H, CH_2), 2.92 (m, 2H, CH), 3.45 (s, 2H, OH), 3.69 (dd, J = 4.59 and 12 Hz, 2H, CH_2), 3.57 (dd, J = 6.7 and 13 Hz, 2H, CH_2), 4.15 (b, 2H, NH), 7.60 (s, 2H, ArH-2 and -3), 7.77 (m, 2H, ArH-6 and -7), 8.38 (m, 2H, ArH-5 and -8), and 10.56 (s, 2H, NH); IR (cm^{-1}) 3280, 3060, 2910, 2840, 1640, 1560, 2910, 1170, and 720; MS *m/z* Calcd for $C_{26}H_{36}O_4N_4$: 468.56; Found $[M]^+$ 469. Anal. ($C_{26}H_{36}N_4O_4 \cdot 0.5\ H_2O$) C, H, N.

1,4-Bis-(2,3-dihydroxypropylamino)-9,10-anthracenedione (5). To a suspension of 250 mg (0.7 mmol) of **3** in 20 mL of H_2O:MeOH (10:1) was added 25 mg (0.1 mmol) of *p*-toluenesulfonic acid,[18,19] and the reaction mixture was refluxed for 6 h. The product then was extracted with CH_2Cl_2 and dried over $MgSO_4$. The CH_2Cl_2 was removed under reduced pressure and the product purified by silica gel gravity chromatography (eluant: $CHCl_3$:MeOH:NH_4; 7:1:2). Recrystallization from EtOH–$CHCl_3$–hexanes yielded 158 mg (58%) of a blue product decomposing at 96–98 °C; ^1H NMR ($CDCl_3$) δ 1.59 (b, 1 H, OH), 2.25 (b, 1 H, OH), 3.45 (dd, J = 7.2 and 14 Hz, 4 H, CH_2), 3.62 (m, 4 H, CH_2), 3.91 (m, 2 H, CH), 4.6 (b, 2 H, NH), 7.42 (s, 2H, ArH-2 and -3), 7.67 (m, 2 H, ArH-6 and -7), and 8.25 (m, 2 H, ArH-5 and -8); IR (cm^{-1}) 3400, 3300, 2910, 1640, 1560, 1250, 1000, and 720; MS *m/z* Calcd for $C_{20}H_{22}N_2O_6$: 386.4; Found $[M+1]^+$ 387, 355, 338, and 315. Anal. ($C_{20}H_{22}N_2O_6 \cdot 2.5\ H_2O$) C, H, N.

1-(2,3-Propyleneamino)-4-hydroxy-9,10-anthracenedione (7). To 2.0 g (8.3 mmol) of 1,4-dihydroxyanthra-

quinone (**6**, quinizarin) in 50 mL of *n*-BuOH was added 0.5 mL (12.49 mmol) of allylamine.[20] The suspension was stirred for 48 h at 50 °C. After the magenta solution was cooled to room temperature, 100 mL of Et_2O was added. The resulting precipitate was filtered, washed three times Et_2O and purified by silica gel gravity chromatography (eluant: hexanes: EtOAc, 4:1) to give 1.65 g (70%) of a purple solid with mp 109–110 °C; ^{1}H NMR ($CDCl_3$) δ 4.08 (m, 1 H, CH_2), 5.31 (m, 2 H, CH_2), 5.99 (m,1 H, CH), 7.19 (d, J = 9.4 Hz, 2 H, ArH-2 and -3), 7.78 (m, 2 H, ArH-6 and -7),8.35 (m, 2 H, ArH-5 and -8), 10.43 (s, 1 H, NH), and 13.71 (s, 1 H, OH). Anal. ($C_{17}H_{13}NO_3$) C, H, N.

1,4,5,8-Leucotetrahydroxy-9,10-anthracenedione. To 5.0 g (0.02 mol) of 1,5-diamino-4,8-dihydroxyanthraquinone (**8**) and 10.0 g (0.25 mol) of NaOH in 200 mL of distilled H_2O under nitrogen atmosphere at 100 °C was added dropwise 10.0 g (0.06 mol) of $Na_2S_2O_4$ dissolved in 50 mL of H_2O.[21] The solution was refluxed for 1 h at 10 °C, cooled to room temperature then cooled further in an ice bath and placed at 5 °C for 12 h. The brown suspension was filtered under nitrogen atmosphere, and the dark solid with a green metallic reflex was dried in a vacuum oven, crushed, and suspended in 200 mL of hot H_2O with gentle stirring. The suspension was slowly acidified with concentrated HCl and the resulting brown precipitate was filtered, washed with H_2O, and dried to yield 4.1 g (80%) of a dark solid with a green metallic reflex. Decomposition at 280 °C; ^{1}H NMR ($CDCl_3$) δ 1.55 (s, 4 H, CH_2), 3.05 (s, 2 H, OH), and 7.32 (s, 2 H, ArH).

1,4-Bis-(2,3-propyleneamino)-5,8-dihydroxy-9,10-anthracenedione (9). To 2.0 g (7.2 mmol) of leuco-1,4,5,8-tetrahydroxyanthraquinone in 25 mL of EtOH under nitrogen atmosphere was added 8.0 mL (140 mmol) of allylamine and the solution was stirred at 60 °C for 4 h.[22] The reaction was then cooled and stirred at 25 °C for 15 h. The resulting precipitate was filtered and washed twice with EtOH and twice with hexanes to yield 2.06 g (81%) of a dark solid with a green reflex. The dark crystals were dissolved in 42 mL of nitrobenzene and the now magenta colored solution was refluxed for 2 h and filtered. The filtrate was cooled to 5 °C for 15 h. The suspension (now brilliant blue) was filtered to yield 2.1 g (90%) of dark needles with a melting point of 198–199 °C; ^{1}H NMR ($CDCl_3$) δ 3.95 (m, 4 H, CH_2), 5.43 (m, 4 H CH_2), 5.92 (m, 2 H, CH), 7.01 (s, 2 H, ArH-2 and -3), and 7.08 (s, 2 H, ArH-6 and -7); IR (cm^{-1}) 3220, 3080, 2900, 1600, 1560, 1450, 1190, 960, 900, and 820. Analysis ($C_{20}H_{18}N_2O_4$) C, H, N.

1-(2,3-Propyleneamino)-4-(2,3-propyleneamino)-5,8-dihydroxy-9,10-anthracenedione-*N*-oxide (10). To a stirring solution of 84 mg (0.2 mmol) of **9** in 38 mL of CH_2Cl_2:MeOH (25:1) and 0.1 mL (1.2 mmol) of Cl_3CCN at −5 °C was added dropwise 1.2 mL (198 mmol) of 30% H_2O_2, which was previously adjusted to a pH of 6.8 with K_2HPO_4.[31] The blue biphasic mixture stirred for 3 h in an ice bath and a color change from blue to fuschia was noted. An amount of 50 mL of hexane was added, the resulting precipitate filtered, and the filtrate washed 2 × H_2O, 2 × Na_2SO_3, 2 × brine and dried over $MgSO_4$. Silica gel flash chromatography (eluant: hexanes:EtOAc, 4:1) produced 60 mg (69%) of a red crystal with a melting point of 210–212 °C; ^{1}H NMR ($CDCl_3$) δ 4.06 (m, 2 H, CH_2), 4.10 (m, 2 H, CH_2), 5.34 (m, 4 H CH_2), 5.95 (m, 2 H, CH), 7.03 (d, J = 9.2 Hz, 1 H, ArH-3), 7.27 (m, 2 H, ArH-6 and -7), and 7.49 (d, J = 9.4 Hz, 1 H, ArH-2); IR (cm^{-1}) 3380, 3210, 1685, 1600, 1380, 1200, 1100, 910, 820, and 750; MS *m/z* Calcd for $C_{20}H_{18}N_2O_5$: 366; Found $[M]^+$ 366, 364, 309, 268, 255, 241, 80, and 55.

1,4-Bis-(3-hydroxy-2-bromopropylamino)-5,8-dihydroxy-9,10-anthracenedione (11). To a vigorously stirred solution of 5.0 g (1.4 mmol) of **9** in 50 mL of DME and 25 mL of H_2O at −5 °C was added 0.5 g (2.8 mmol) of *N*-bromosuccinimide.[32] The solution was stirred for 2 h, then the product was extracted into CH_2Cl_2 and dried over $MgSO_4$. The solvent was removed in vacuo and the product purified by silica gel chromatography (eluant: hexanes:EtOAc, 4:1) followed by another silica gel column (eluant: CH_2Cl_2) to give 0.25 g (32%) of a blue solid with mp 105–109 °C; ^{1}H NMR ($CDCl_3$) δ 3.61 (m, 2 H, CH), 3.78 (dd, J = 6.6 and 14.12 Hz, 4 H, CH2), 3.92 (m, 4 H, CH_2), 4.14 (b, 2 H, OH), 4.32 (b, 2 H, NH), 7.02 (s, 2 H, ArH-2 and -3), 8.98–9.16 (m, 2 H, ArH-6 and -7), (m, 1 H, NH), 10.29 (m, 1 H, NH), 12.51 (b, 1 H, OH), and 13.09 (b, 1 H, OH); IR (cm^{-1}) 3420, 3160, 1680, 1600, 1560, 1500, 1180, and 790; MS *m/z* Calcd for $C_{20}H_{20}N_2O_6Br_2$: 544.2; Found $[M]^+$ 544, 540, 380, 270, 241, 82, 80, and 56.

1-(2,3-Epoxypropylamino)-4-(3-hydroxy-2-bromopropylamino)-5,8-dihydroxy-9,10-anthracenedione (12). Bromohydrin **11** was converted to epoxide **12** in a similar manner as **3** to **4**. Yield 67% ^{1}H NMR ($CDCl_3$) δ 2.76 (m, 1 H, CH_2), 2.91 (m, 1 H, CH_2), 3.56 (m, 1 H, CH), 3.82 (m, 1 H, CH), 3.91 (m, 2 H, CH_2), 3.91 (m, 2 H, CH_2), 4.04 (m, 2 H, CH2), 4.24 (b, 1 H, OH), 4.54 (b, 2 H, NH), 7.25 (m, 2 H, ArH-2 and -3), 9.10 (d, J = 9.8 Hz, 1 H, ArH-5 or -8), 9.13 (d, J = 9.8 Hz, 1 H, ArH-5 or -8), 10.23 (b, 2 H, NH), 12.61 (b, 1 H, OH), and 13.09 (b, 1 H, OH); IR (cm^{-1}) 3400, 3110, 2910, 1650, 1250, 1200, and 810; MS *m/z* Calcd for $C_{20}H_{20}N_2O_6Br$: 463.36; Found $[M]^+$ 464, 369, 353, 337, 283, 270, 11, 80, 67, and 56.

[1,5-(Chloro-2-hydroxypropylamino)-4,8-dihydroxy]-9,10-anthracenedione (13). Chlorohydrin **13** was prepared from **8** in an analogous manner as **3** from **2**. Recrystallized from EtOAc-hexanes; yield 56%; mp 300 °C; ^{1}H NMR ($CDCl_3$) δ 3.31 (b, 2 H, OH), 3.42 (m, 4 H, CH_2), 3.61 (m, 2 H, CH), 3.65 (d, J = 7.2 Hz, 4 H, CH_2), 3.90 (b, 1 H, NH), 4.1 (b, 1 H, OH), 7.09 (m, 2 H, ArH-2 and -6), 7.47 (m, 2 H, ArH-3 and -7), and 9.60 (s, 2 H, OH); IR (cm^{-1}) 3458, 3310, 1620, 1570, 1220, and 730;MS *m/z* Calcd for $C_{20}H_{20}N_2O_6Cl_2$: 454.29. Found: $[M]^+$ 454, 418, 382, 283, 270, and 57.

[1,5-(2,3-Epoxypropylamino)-4,8-dihydroxy]-9,10-anthracenedione (14). Chlorohydrin **13** was converted to

epoxide **14** as for **4** from **3**. Recrystallized from $CHCl_3$–MeOH; yield 41%; mp 185–186 °C; ^1H NMR ($CDCl_3$) δ 2.73 (m, 2 H, CH_2), 2.86 (m, 2 H, CH_2), 3.51 (m, 2 H, CH), 3.65 (m, 4 H, CH_2), 3.83 (b, 1 H, NH), 3.98 (b, 1 H, NH), 7.29 (s, 2 H, ArH-2 and -6), 7.39 (m, 2 H, ArH-3 and -7), and 9.81 (s, 2 H, OH); IR (cm^{-1}) 3440, 3260, 2910, 1560, 1250, and 760; MS *m/z* Calcd for $C_{20}H_{18}N_2O_6$: 382.3; Found $[M+1]^+$ 383, 382, 237, 71, and 57.

1-(3,4-Butyleneamino)-4-amino-9,10-anthracenedione (15). To 3.0 g (13 mmol) of **1** in 50 mL of CH_3CN was added 3.5 g (25.2 mmol) of K_2CO_3 and 9.0 mL (0.05 mol) of 4-bromo-1-butene diluted in 15 mL of CH_3CN. The solution was stirred at 60 °C for 36 h. The solvent was removed under reduced pressure and the product was purified on a silica gel gravity column (eluant: CH_2Cl_2) to yield 2.3 g (64%) of a blue solid with melting point of 116–119 °C; ^1H NMR ($CDCl_3$) δ 2.56 (q, J = 6.6 Hz, 1 H, CH_2), 3.46 (t, J = 7.05, 2 H, CH_2), 4.15 (m, 1 H, NH), 5.24 (m, 2 H, CH_2), 5.91 (m, 1 H, CH), 7.04 (d, J = 9.4 Hz, 1 H, ArH-3), 7.15 (d, J = 9.4 Hz, 1 H, ArH-3), 7.75 (m, 2 H, ArH-6 and -7), and 8.35 (m, 2 H, ArH-5 and -8); IR (cm^{-1}) 3380, 3240, 2820, 1565, 1530, 1260, 995, 910, and 720; MS *m/z* Calcd for $C_{18}H_{16}N_2O_2$: 292.322; Found $[M]^+$ 292.120, 251, 239, 223, and 55.

1-(3,4-Butyleneamino)-4-amino-9,10-anthracenedione-*N*-oxide (16). Compound **16** was prepared by the method described for compound **10**.[26] Silica gel flash chromatography (eluant: CH_2Cl_2:MeOH, 25:1) produced 1-hydroxy-4-aminoanthraquinone and the *N*-oxide (43%); mp 93–97 °C; ^1H NMR ($CDCl_3$) δ 2.54 (dd, J = 13. 3 and 6.6 Hz, 2 H, CH_2), 3.43 (dd, J = 13.3 and 6.6 Hz, 2 H, CH_2), 5.25 (m, 2 H, CH_2), 5.91 (m, 1 H, CH), 7.00 (d, J = 9.4 Hz, 1 H, ArH-3), 7.57 (d, J = 9.4 Hz, 1 H, ArH-2), 7.81 (m, 2 H, ArH-6 and -7), 8.14 (s, J = 7.4 Hz, 1 H, ArH-5), and 8.22 (d, J = 7.4 Hz, 1 H, ArH-8); IR (cm^{-1}) 2910, 1665, 1635, 1590, 1510, 1350, 1250, and 720; MS *m/z* Calcd for $C_{18}H_{16}N_2O_3$:308; Found $[M-1]^+$ 307, 293, 281, 240, 206, and 55.

2-(2,3-Propyleneamino)-3-chloro-1,4-naphthoquinone (17). 2-Amino-3-chloro-1,4-naphthoquinone was reacted with allyl bromide as in the reaction of **1** with 4-bromo-1-butene (see compound **15**). Essentially pure orange product was obtained by silica gel flash chromatography (eluant: $CHCl_3$:MeOH, 25:1); remaining impurities were removed by stirring in a solution of hexane–Et_2O (4:1). Yield 74%, mp 111–112 °C; ^1H NMR ($CDCl_3$) δ 4.45 (m, 2 H, CH_2), 5.24 (m, 2 H, CH_2), 6.00 (m, 1 H, CH), 6.18 (b, 1 H, NH), 7.64 (m, 1 H, ArH-6 or -7), 7.74 (m, 1 H, ArH-6 or -7), 8.04 (m, 1 H, ArH-5 or -8), and 8.14 (m, 1 H, ArH-5 or -8); ^{13}C NMR (DEPT) 45.9 (CH_2), 115.5 (CH_2), 125.78 (CH), 126.47(CH), 132.65(CH), 134.83(CH), 135.80 (CH); IR (cm^{-1}) 3300, 1670, 1590, 1550, 1295, 930, and 720; MS *m/z* Calcd 247.67, Found $[M]^+$ 247.0396, [M+2] 249, 212, 56. Anal. ($C_{13}H_{10}NO_2Cl$) C, H.

2-(2,3-Epoxypropylamino)-3-chloro-1,4-naphthoquinone (18). Compound **17** was reacted with 30% H_2O_2 using an analogous procedure to the reaction of **9** to give **10**.[26] Recrystallized from Et_2O–CH_2Cl_2-hexanes; yield 72%; mp 116–117 °C; ^1H NMR ($CDCl_3$) δ 2.61 (m, 1 H, CH_2), 2.84 (m, 1 H, CH_2), 3.30 (m, 1 H, CH), 3.91 (m, 1 H, CH_2), 4.32 (m, 1 H, CH_2), 6.17 (b, 1 H, NH), 7.64 (m, 1 H, Ar H-6 or -7), 7.74 (m, 1 H, Ar H-6 or -7), 8.04 (m, 1 H, Ar H-5 or -8), 8.14 (m, 1 H, Ar H-5 or -8); IR (cm^{-1}) 3310, 1670, 4610, 1510, 1290, 1250, 900, 850, 720. Anal. ($C_{13}H_{10}NO_3Cl$·0.5 H_2O) C, H.

2-(3-Chloro-2-hydroxypropylamino)-3-chloro-1,4-naphthoquinone (19). 2-Amino-3-chloro-1,4-naphthoquinone was reacted with epichlorohydrin as in the preparation of compound **2**. Recrystallized from Et_2O–CH_2Cl_2-hexanes; yield 55%; mp 208–210 °C; ^1H NMR ($CDCl_3$) δ 3.67 (m, 2 H, CH2), 3.72 (s, 1 H, OH), 4.29 (dd, J = 5.6 and 12 Hz, 2 H, CH_2), 5.22 (m, 1 H, CH), 5.60 (b, 1 H, NH), 7.70 (m, 2 H, ArH-6 and -7), and 8.11 (dd, J = 12 and 28 Hz, 2 H, ArH-5 and -8); IR (cm^{-1}) 3450, 3300, 1670, 1610, 1380, 1270, 845, and 710. Anal. ($C_{13}H_{11}NO_3Cl_2$) C, H, N.

2-(2,3-Propyleneamino)-1,4-naphthoquinone (20). To 0.5 g (3.12 mmol) of 1,4-dihydroxynaphthalene in 50 mL of Et_2O under nitrogen atmosphere was added dropwise 1.0 mL (9.36 mmol) of allylamine and 0.5 g of $MgSO_4$. After refluxing for 30 min, 2.8 g (12.5 mmol) of Ag_2O was added slowly and the reaction again refluxed for 2 h. After cooling to room temperature, the reaction stirred for 36 h. The reaction was filtered with the aid of Celite, washed 2 × Et_2O and 3 × $CHCl_3$ (10 mL portions), and the entire process was repeated with fresh Celite. The solvent was removed under reduced pressure and product was obtained by purification on silica gel column (eluant: CH_2Cl_2:MeOH, 15:1) and recrystallization from Et_2O-hexanes to yield 0.5 g (45%): mp 113–114 °C; ^1H NMR ($CDCl_3$) δ 3.83 (apparent triplet, J = 5.65 Hz, 2 H, CH_2), 5.32 (m, 2 H, CH_2), 5.74 (s, 1 H, CH), 5.88 (m, 1 H, CH), 6.03 (b, 1 H, NH), 7.59 (m, 1 H, ArH-6 or -7), 7.71 (m, 1 H, ArH-6 or -7), and 8.11 (m, 2 H, ArH-5 and -8); 13C (DEPT) 45.04 (CH_2), 101.58 (CH), 118.17 (CH_2), 126.72 (CH), 131.54 (CH), 131.96(CH), 133.56(CH), 134.76(CH); IR (cm^{-1}) 3320, 1670, 1600, 1560, 1500, 1250, 1000, 920, and 720. Anal. ($C_{13}H_{11}NO_2$) C, H, N.

2,5-Bis-(2,3-propyleneamino)quinone (21). Dihydroquinone was reacted with allylamine and $MgSO_4$ in an identical manner to the preparation of **20**. Recrystallized from MeOH–acetone; yield 76%; mp 192–193 °C; ^1H NMR ($CDCl_3$) δ 3.79 (m, 4 H, CH_2), 5.28 (m, 6 H, CH_2), 5.85 (m, 2 H, CH), and 6.65 (b, 2 H, NH); IR (cm^{-1}) 3280, 1640, 1545, 1480, 1100, 990, and 930; MS *m/z* Calcd 218.25, Found $[M]^+$ 218.1051. Anal. ($C_{12}H_4N_2O_2$·0.25 H_2O) C, H.

2,5-Bis-(2,3-propyleneamino)quinone-*N*-oxide (22). Compound **22** was prepared from **21** in a similar manner to the synthesis of **18**.[26] Recrystallized from Et_2O–CH_2Cl_2-hexanes; yield 70%; mp 153–154 °C; ^1H NMR ($CDCl_3$) δ 3.87 (m, 2 H, CH_2) 4.54 (m, 2 H, CH_2),

5.22–5.36 (m, 4 H, CH_2), 5.81 (s, 2 H, CH), 5.85(m, 2 H, CH), and 6.45 (b, 2 H, CH). Anal. ($C_{12}H_{14}N_2O_4$) C, H.

1,4-Bis-acrylamidoanthraquinone (23). The preparation of this material was based on modifications of a similar literature procedure.[33] Triethylamine (1.5 mL, 10.76 mmol) followed by acryloyl chloride (9.58 g, 105.85 mmol) was added to a cooled (0–5 °C) solution of **1** (2.5 g, 10.49 mmol) in 150 mL of toluene. The reaction was allowed to warm slowly and stir overnight at room temperature After heating at 86 °C for 1 h, the mixture was diluted with 500 mL of toluene, filtered to clarify, then concentrated under reduced pressure. The resultant red residue was redissolved in toluene and washed with dilute aqueous NaCl solution followed by saturated aqueous NaCl solution. The volume of the solution was reduced and crystallization was induced by addition of hexane–Et_2O to give 2.72 g (75% yield) of blood red crystals. The material was further purified by recrystallization from DMF. mp 208–210 °C; ^{1}H NMR (200 MHz, $CDCl_3$) δ 5.9, (dd, 1H, $COC\underline{H}CH_2$), 6.6, (m, 2H, $COCHC\underline{H}_2$), 7.8, (m, 1H, Ar-H), 8.2 (m, 1H, Ar-H), 9.2 (s, 1H, Ar-H), 12.8 (s, NH).

1-(Epoxypropyl)aminoanthraquinone (27). Epibromohydrin and 1-aminoanthraquinone were reacted in hot glacial HOAc as in the preparation of **3** from **2** to give 87% yield of 1-(3-bromo-2-hydroxypropyl)aminoanthraquinone. The ^{1}H NMR spectra in $CDCl_3$ and in DMSO-d_6 were consistent with the proposed structure of the desired compound. Sodium hydroxide (2 N, 3.6 mL, 7.2 mmol) was added, with vigorous stirring, to a solution of 1-(3-bromo-2-hydroxypropyl)aminoanthraquinone (2.00 g, 5.55 mmol) dissolved in 30 mL of DMF containing 10 mL of EtOH. After stirring for 5 min, the reaction mixture was diluted with 100 mL of H_2O, cooled and then filtered. The isolated solid precipitate was air dried, then chromatographed on silica gel and eluted with EtOAc–hexane (1:3). Pure material could not be isolated. The ^{1}H NMR spectrum in $CDCl_3$ was consistent with the proposed structure of the desired compound. This material contains approximately 25–30% 1-aminoanthraquinone, as determined by GLC.

1,4-Bis-(2′,3′-epoxypropyloxy)naphthalene (28). Epibromohydrin (6.16 g, 44.95 mmol) was added to a mixture of 1,4-dihydroxynaphthalene (2.40 g, 14.98 mmol) and cesium carbonate (10.74 g, 32.96 mmol) in 100 mL of MEK and the resultant mixture was held at reflux temperature for 24 h. The reaction mixture was filtered and concentrated, under reduced pressure to a brown solid residue. This material was purified by column chromatography [100 g of silica gel and eluted with hexane–EtOAc (2:1)] (41% yield) and further purified by crystallization with hexane–EtOAc to afford white needle-like crystals. mp 110–111 °C; ^{1}H NMR (200 MHz, $CDCl_3$) δ 2.84, (dd, 1H, $C\underline{H}_aH_b$), 2.98, (dd, 1H, $CH_a\underline{H}_b$), 3.48, (m, 1H, CH), 4.10, (dd, 1H, $C\underline{H}_cH_d$), 4.35, (dd, 1H, $CH_c\underline{H}_d$), 6.70, (s, 1H, Ar-H), 7.54, (m, 1H, Ar-H), 8.26, (m, 1H, Ar-H).

The preparations of compounds **24–26** and **29** have been reported in the literature.[34,35] the physical and spectral data of the synthetic samples were consistent with those given therein.

Biological screening. In the initial in-house screening, compounds **1–22** were tested according to the NCI protocols.[36] Five in vitro cytotoxicity screening systems were employed: L-1210 (murine lymphocytic leukemia cells), P-388 (murine lymphoid neoplastic cells), A-549 (human lung carcinoma), HCT-8 (human colon carcinoma), and KB (human epidermoid carcinoma of the nasopharynx). In the in vitro test systems, evaluation was based on the effective dose that inhibited cell growth by 50% of the control group (ED_{50}). For pure compounds, values with an average less than or equal to 4 μg/mL were considered significantly cytotoxic. Values greater than 4 μg/mL were considered inactive. The target compound, 1,4-bis-(2,3-epoxypropylamino)-9,10-anthracenedione (**3**), was used as the standard in the biological screening.

Acknowledgements

This investigation was supported by a grant (CA-17625) from the National Cancer Institute awarded to K. H. Lee. We would like to thank Dr David Millington, Duke University Pediatric Department, Duke Medical Center, Durham, North Carolina and Dr Dean Mabry, School of Public Health, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina for mass spectral data, Dr Y. C. Cheng of the University of North Carolina at Chapel Hill Cancer Research Center for the biological data, and Dr David L. Harris of the Department of Chemistry and Mr James Gilbert of the Department of Medicinal Chemistry, University of North Carolina for NMR data.

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(*Received in U.S.A.* 2 *December* 1996; *accepted* 3 *March* 1997)