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Heterocyclic Quinones. XIII.[1] Dimerization in the Series of 5,8-Quinazolinediones: Synthesis and Antitumor Effects of Bis(4-amino-5,8-quinazolinediones)

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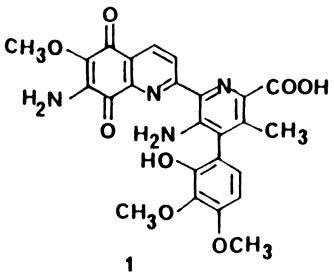
With the aim of obtaining new antitumor drugs more active than previously described 5,8-quinazolinediones, a series of dimers of 5,8-quinazolinediones linked in the 4-position by a simple or a substituted α,ω-diaminopolymethylenic chain was studied. The structure–activity relationships of these compounds are discussed as functions of the length of the chain, presence or absence of other functional groups, nature of these functional groups, position of the chain and nature of the substituents in the 6 and(or) 7-positions. When bis(5,8-quinazolinediones) were substituted in the 6-position with a methoxyl group, the dimerization showed a variable effect on cytotoxicity toward L1210 leukemia cells. Bis[4-amino-bis-6,7(1-aziridinyl)-5,8-quinazolinediones] which exhibited high cytotoxic activity (IC_{50} 0.0037 to 0.018 μM) were further screened *in vivo* for activity against murine P388 leukemia. Antitumor activity was increased by the dimerization of the molecule. The most potent compound bears an additional tertiary amino function on the chain.

Keywords——bis(4-amino-5,8-quinazolinedione); antitumor activity; aziridinyl quinone; cytotoxicity; dimerization; Fremy's salt; heterocyclic quinone; 5,8-quinazolinedione

Streptonigrin (**1**) exhibited broad spectrum inhibition of various tumors but was very toxic.[2] The principal functionality responsible for its activity seemed to be the 5,8-quinolinedione moiety. According to Lown and Sim,[3] single-strand cleavage of covalently closed circular deoxyribonucleic acid (DNA) was due to OH free radicals generated from the reduced quinone function and oxygen.

In connection with our research on nitrogen heterocyclic quinones, we have previously described 6,7-bis(1-aziridinyl)-5,8-quinazolinedione (**2a**)[4] which was very cytotoxic towards L1210 leukemia cells and active *in vivo* against P388 lymphocytic leukemia.



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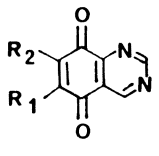

2a: $R_1 = R_2 =$ N

2b: $R_1 = OCH_3$ $R_2 = H$

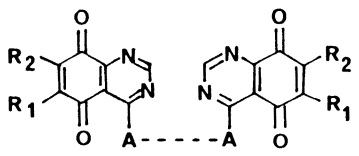

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Chart 1

It is well-known,[5,6] in pharmacomodulation, that dimeric compounds are often more active than the initial compounds. The symmetrical dimerization of a ligand can indeed lead to a very large increase of its binding affinity for a receptor as shown, for example, by the dimers of daunomycins[7] in which the dicarboxylic acid hydrazide functions are bridged by simple methylene chains of variable length, dimers of 9-aminoacridine joined *via* their amino group with a variable linking chain[7] or dimers of pyridocarbazoles.[5] In most cases, the relationship between the structure of the linking chain and the biological properties of these molecules is still unclear since DNA binding properties are not greatly influenced by the linker chain's length, flexibility or rigidity. However, in some cases, such as piperidyl-linked bis-pyridocarbazoles, the ligand rigidity is a determinant of sequence specificity, mechanism of bis-intercalation, and antitumor activity.[7]

We applied this concept to 5,8-quinazolinediones. We expected to increase their activity by preparing bis(5,8-quinazolinediones) in which the two nuclei are linked by an α,ω-diaminopolymethylenic chain. Among many possibilities, we chose to link the two heterocyclic nuclei at the 4,4′-positions for the following reasons; in nitrogen 6-membered heterocycles, the increase in basicity is greater when the amino function is in the 4-position than when it is in the 2-position.[8] It is well known that electrostatic binding to DNA phosphate groups is facilitated when molecules are sufficiently basic. Ambrose *et al.* showed that tumor cells apparently have a higher negative surface charge suggesting that basic compounds should be concentrated in such cells.[9]

In this paper, we wish to report the synthesis, and cytostatic and antitumor activities, of bis(4-amino-5,8-quinazolinediones) (**3**) in which A—A is an α,ω-diaminopolymethylenic chain onto which various functional groups can be introduced in order to modify either the partition coefficient or DNA affinity. Some other derivatives were also studied.

Synthesis

The methoxyl group in the *p*-position relative to the nitrogen of the quinoline nucleus was important for the biological activity of streptonigrin.[10] Consequently, such a group has been preserved on the 5,8-quinazolinedione nucleus. Because of the instability of the methoxy *p*-quinone group, the quinonic function was introduced in the last step of the synthesis.

The key compound was the 5-benzyloxy-4-chloro-6-methoxyquinazoline (**8**) which was prepared from 2-benzyloxy-3-methoxy-6-nitrobenzaldehyde (**4**). Compound **4** was synthesized from commercial 2-hydroxy-3-methoxybenzaldehyde according to Julia *et al.*[11] The aldehyde (**4**) was oxidized to the acid (**5**) by potassium permanganate in 71% yield. The nitro group of **5** was reduced to an amino group by using ferrous sulfate in an ammoniacal medium in 79% yield. The unstable amino acid (**6**) could not be purified. Heterocyclization of **6** to give **7** could not be achieved by heating **6** with formamide according to Niementowski's classical method.[12] Compound **7** was obtained in a fair yield (80%) by heating the crude amino acid (**6**) with *s*-triazine in the presence of piperidine according to Kreutzberger and Uzbek.[13] Reaction of **7** in benzene with an equimolecular amount of phosphoryl chloride in the presence of triethylamine yielded 81% of the chlorinated compound **8**.

Reaction of compound **8** with an α,ω-diamine (**9**, A = NH or N) in the presence of triethylamine at room temperature produced only the bis(5-benzyloxy-6-methoxy-4-quinazolinamine) (**10**, A = NH or N) even if the α,ω-diamine was used in excess. Compound **10** was debenzylated to **11** either by heating with trifluoroacetic acid (TFA) or by hydrogenolysis in the presence of palladium on activated carbon. The methoxyquinone (**12**) was specifically obtained by oxidation of **11** with potassium nitrosodisulfonate (Fremy's salt).[14]

We studied the effect of various factors on cytotoxicity: length of the polymethylenic chain between the nitrogens in the 4-position (compounds **12a**, **12b**), the presence of aliphatic

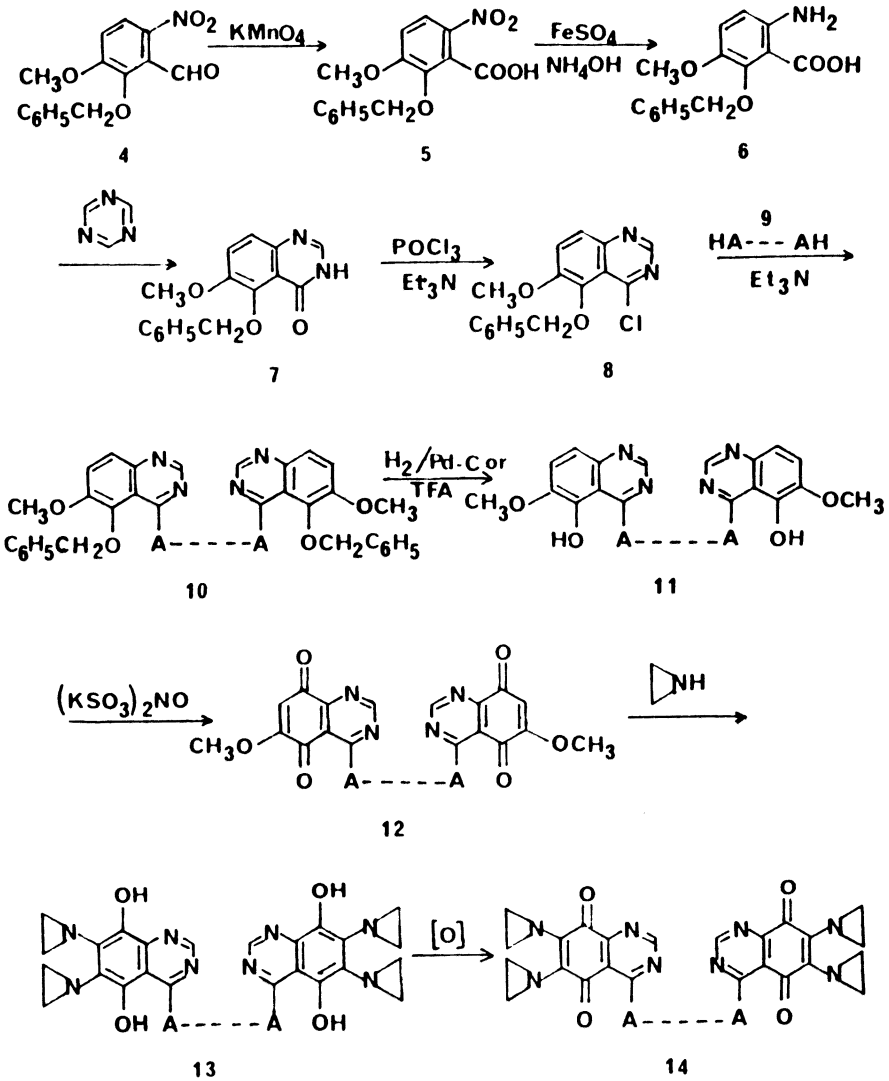


Chart 2

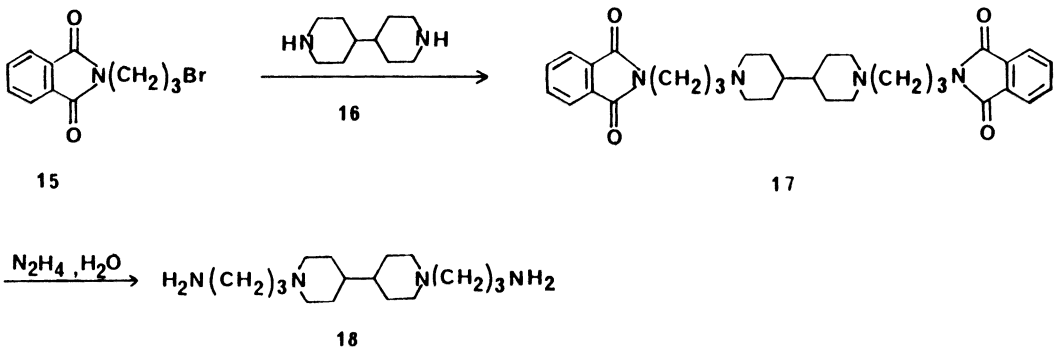


Chart 3

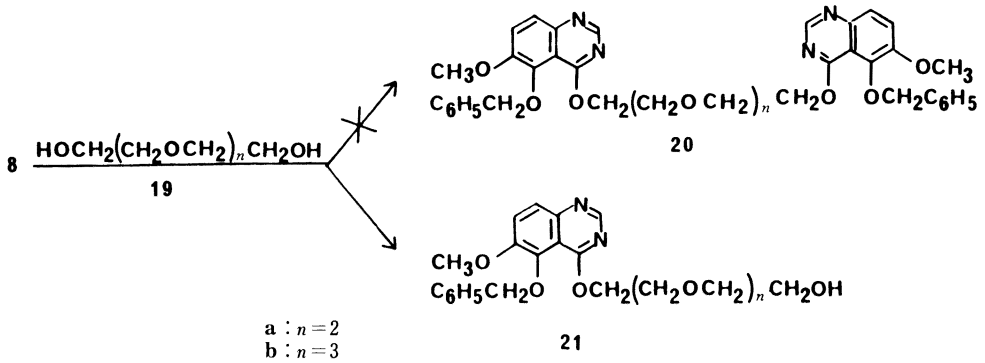


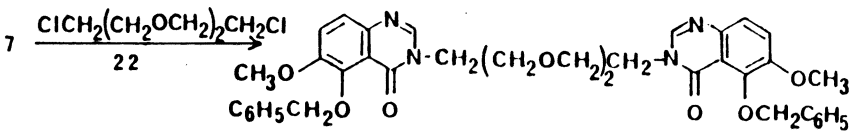
Chart 4

or heterocyclic tertiary amino groups (compounds **12c** and **12d** respectively.[15]) The triamine suitable for the synthesis of **12c** could be compared with polyamines such as spermidine, which have been reported to inhibit the growth of some animal tumors.[16,17] The chain of **12d** has been used in the synthesis of 7*H*-pyridocarbazole dimers.[5] Compound **18** was prepared from *N*-(3-bromopropyl)phthalimide (**15**) and 4,4′-bipiperidine (**16**).

In compound **12c**, the tertiary amino group was easily quaternarized by iodomethane to give **12g**. Introduction of a tertiary amino group, which increases the basicity, and the presence of a quaternary ammonium function are two factors which can facilitate DNA-binding. We also used aliphatic or cyclic chain with ether functions (compounds **12e** and **12f**) in order to obtain more hydrophilic compounds. Use of diazacrown, compound **12f**, was based on the ability of synthetic ionophores to complex various metal ions (alkali, alkaline earth, iron, *etc.*) and to facilitate their transport through biological membrane.[18]

To investigate if antitumor activity was caused by the presence of a diamino function in the 4 and 4′-positions of 5,8-quinazolinediones, we wanted to replace it by an α,ω-diether chain in compound **20** (Chart 4). Unfortunately, the reaction of the chlorinated compound **8** with triethylene glycol (**19a**) or tetraethylene glycol (**19b**) in the presence of various bases (sodium hydride, silver(I) oxide, potassium carbonate) or in neutral medium yielded only **21** without any **20**. The best result was obtained when potassium carbonate in acetone was used. Compound **21** did not react with **8** to give **20**.[19]

In the same way, we compared the cytotoxicity of bis(4-amino-quinazolinediones) with that of quinones (**25**) derived from bis(3-substituted-4(3*H*)-quinazolinone) (**24**). The quinone (**25**) was synthesized as indicated in Chart 5.



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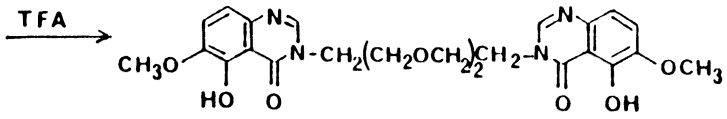

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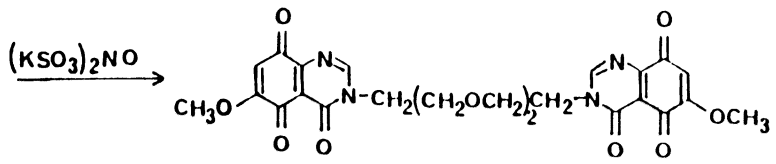

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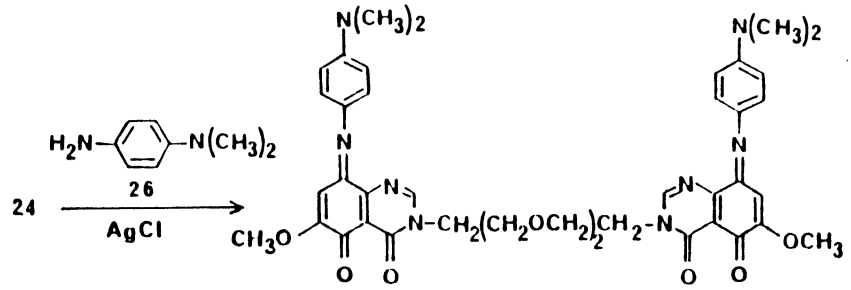

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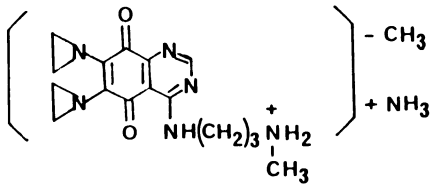
Chart 5

It is well known[20] that 4(3*H*)-quinazolinones can be alkylated by a halogenated derivative in an alkaline medium on the nitrogen in the 3-position. The 4(3*H*)-quinazolinone (**7**) was condensed with 1,8-dichloro-3,6-dioxaoctane (**22**) in the presence of sodium hydride to produce the bis-quinazoline (**23**) in 57% yield. The quinone (**25**) was obtained in 20% yield *via* the phenol (**24**) according to the above method. In this case, the phenol (**24**) was oxidized into **25** in weakly alkaline medium, and consequently dibasic sodium phosphate was to be used instead of monobasic potassium phosphate.

As Hodnett *et al.* showed[21] that some benzoquinonimines are very active *in vivo* on murine sarcoma 180, we prepared the bis-quinonimine (**27**) in 73% yield by the reaction of *N*,*N*-dimethyl-1,4-benzenediamine (**26**) with the phenol (**24**) in the presence of silver chloride according to their method[21] (Chart 5). On the other hand, preparation of the corresponding oxime by the same method[21] could not be achieved.

Because of the ester-like properties of **12**, the methoxyl group in the quinone nucleus was

replaced by nucleophilic reagents. Aziridine was interesting because of its bioalkylating properties as seen in AZQ or NSC-182986.[22,23] In this case, the presence of two aziridino groups could provide bis-alkylating compounds. Treatment of **12** with two equivalents of aziridine in methanol was generally unsuccessful. Use of aziridine in excess, without any solvent, gave the 4,4′-bis[6,7-bis(1-aziridinyl)-5,8-quinazolinediones] (**14**) *via* the hydroquinones (**13**) as intermediates. Compound **14d** has not yet been isolated because of its instability. Reaction of aziridine with methoxyquinone (**14f**) produced several inseparable compounds. The aziridinylquinones (**14**) had to be immediately purified by flash column chromatography. Often accurate elemental analysis could not be obtained because the quinones (**14**) solvated variable quantities of the purification solvents. However, infrared (IR) proton nuclear magnetic resonance (^{1}H-NMR) and mass spectrum (MS) data were consistent with the assigned structures. For compound **14c**, a $(M+H)^+$ peak was not found, but a peak at *m/z* 331 could be assigned to the ion (**28**). Such a fragmentation was also found for the analogous 6-methoxyquinone (**12c**) at *m/z* 279 beside the $(M+H)^+$ peak at *m/z* 522.



28

Pharmacology

***In Vitro* Cytotoxicity on L1210 Leukemia Cells**——Cytotoxicity toward L1210 leukemia cells was determined. Dose–effect relationships of the various compounds tested were determined from the regression line in a plot of percent cell growth inhibition as a function of the logarithm of the dose. From these curves, the dose of drug reducing the cell growth by

TABLE I. Effect of Bis(4-amino-5,8-quinazolinediones) and Derivatives on the Growth of L1210 Cells

Compd. No.	IC_{50}[a]		Correlation coefficient
	ng/ml	μM	
2b[4]	593	3.12	>0.95
12a	>1000	>2.29	–
12b	1215	2.40	0.97
12c	694	1.25	0.95
12d	854	1.30	0.96
12e	1846	3.52	0.99
12f	>10000	>15.7	—
12g	>10000	>14.5	—
2a[4]	20	0.083	0.95
14a	6.58	0.011	0.99
14b	2.23	0.0037	0.99
14c	3.01	0.0048	0.99
14e	12.7	0.018	0.99
25	>10000	>19.0	—
27	6674	8.18	0.99
Doxorubicin[b]	28	0.048	0.99

a) IC_{50}: drug concentration that decreases the growth rate of the cells by 50% after 48 h of culture. They are determined by a least-squares plotting of the experimental data. *b*) Doxorubicin was used for reference.

TABLE II. Effect of Bis[4-amino-6,7-bis(1-aziridinyl)-5,8-quinazolinediones] (**14**) on P388 Lymphocytic Leukemia[a]

Compd. No.	Dose per inject. mg/kg	Toxicity survivors[b]	ΔP (J_5-J_1)[c]	MST[d] (control)	T/C[e] (%)
12d	10	10/10	−1.1	10.6	103
	5	10/10	−0.1	10.6	100
	2.5	10/10	+0.5	10.6	98
14b	12.5	04/06	−3.2	14.0	133[f]
	10.0	06/06	−3.0	13.3	127[f]
	7.5	06/06	−1.0	12.8	122
	5.0	10/10	−1.4	12.6	120
	2.5	06/06	0	11.7	111
Controls (**13**)	—	—	+2.6	10.5	—
14c	7.5	06/06	−5.0	6.4	61
	5.0	10/10	−4.2	17.6	168[f]
	2.5	10/10	−1.8	15.7	150[f]
	1.25	06/06	−1.0	14.4	137[f]
Controls (**13**)	—	—	+2.6	10.5	—
14e	15	07/10	−4.1	16.0	145[f]
	10	10/10	−1.8	14.8	135[f]
	5	10/10	−1.1	14.6	133[f]
	2.5	10/10	−0.5	12.6	115
Controls (**25**)	—	—	+1.3	11.0	—

a) For the method, see the experimental section. *b*) Number of survivors on day 5/number of treated mice. *c*) Weight between day 1 and day 5. *d*) MST = median survival time in days. *e*) T/C% = MST of treated animals over controls × 100; *f*) significant if T/C% ≥ 125.

50% after 48 h as compared to the controls (IC_{50}) was estimated. Cytotoxicity of synthesized quinones is summarized in Table I.

***In Vivo* Antitumor Activity**——Four compounds, **12d**, **14b**, **14c** and **14e**, which had a IC_{50} equal to/or lower than 1 μM were retained for *in vivo* study on P388 leukemia. Tumor cells were intraperitoneally (i.p.) grafted and the treatment (one injection) was given on day 1 by the same route. The results are listed in Table II. They were confirmed in a second experiment.

Results and Discussion

Cytotoxicity

On the basis of the data obtained by *in vitro* screening, the structure–activity relationships of the bis(4-amino-5,8-quinazolinediones) are considered to be as follows. In the case of the 6-methoxyquinones, dimerization of 6-methoxy-5,8-quinazolinedione (**2b**) (IC_{50} 3.12 μM) through a simple α,ω-diaminopolymethylenic chain that bears two methylenes, compound **12a** (IC_{50} 2.29 μM), or seven methylenes, compound **12b** (IC_{50} 2.40 μM), did not improve the activity. Additional tertiary amino substituents either aliphatic, compound **12c** (IC_{50} 1.25 μM), or heterocyclic, compound **12d** (IC_{50} 1.30 μM), significantly increased the cytotoxicity whereas quaternization of the tertiary amino function afforded the inactive compound **12g** ($IC_{50} > 14.5$ μM). The presence of ether functions on the chain, compound **12e** (IC_{50} 3.52 μM), was somewhat unfavorable. The presence of a diazacrown, compound **12f** ($IC_{50} > 15.7$ μM), was extremely unfavorable. Replacing the bis(4-aminoquinazoline) structure by a bis(3-substituted-4(3*H*)quinazolinone) structure afforded the inactive compound **25** ($IC_{50} > 19$ μM), whereas the presence of a quinonimine function is more favorable, compound **27** (IC_{50} 8.18 μM).

Comparison of the bis[4-amino-6,7-bis(1-aziridinyl)-5,8-quinazolinediones] (**14a**, **b**, **c**, **e**) with the 6,7-bis(1-aziridinyl)-5,8-quinazolinedione (**2a**) (IC_{50} 0.083 μM)[4] showed that in all cases the cytotoxicity was increased in the dimeric compounds (from five- to twenty-one-fold), and was up to ten times higher than that of doxorubicin used as a reference.

In Vivo Antitumor Activity

6,7-Bis(1-aziridinyl)-5,8-quinazolinedione (**2a**) was active *in vivo* against P388 leukemia in mouse[4] (T/C = 134%, treatment schedule: i.p., one injection at day 1, 20 mg/kg) (see footnote in Table II). Considering the lowest dose which gave T/C $\simeq$ 130%, it appears that antitumor activity is increased by the dimerization of the molecule *via* an α,ω-diaminopolymethylenic chain (compound **14b**, T/C = 133% for 12.5 mg/kg; compound **14c**, T/C = 137% for 1.25 mg/kg; compound **14e**, T/C = 133% for 5 mg/kg). Compound **14c** with a tertiary amino group on the chain exhibited the highest potency among the compounds reported here. Compound **12d** substituted by a methoxyl group at the 6-position did not show antitumor activity.

Conclusion

We prepared a series of bis(4-amino-5,8-quinazolinediones) in which the two nuclei were linked by an α,ω-diaminopolymethylenic chain, with the aim of increasing antitumor activity of 5,8-quinazolinediones. In the series of 6-methoxyquinones (**12**), dimerization had a variable effect on the cytotoxicity. On the other hand, in the series of 6,7-bis(1-aziridinyl)quinones (**14**), the simple process of dimerization led to a large increase of cytotoxicity and antitumor effect on P388 leukemia. The most potent compound **14c** bears an additional tertiary amino function on the chain. There appears to be no obvious relationship between the activity of bis-quinazolinediones and the linker chain structure.

Experimental

All melting points were determined on a Maquenne apparatus and are uncorrected. IR spectra were obtained on a Perkin–Elmer 157G spectrometer. ^{1}H-NMR spectra were measured with a Bruker 270 MHz spectrometer with trimethylsilane $(Me_3Si)_2$ as an internal reference. DCI/NH_3 mass spectra were recorded on a Nermag R10-10C instrument. Thin layer chromatography (TLC) was carried out on Merck GF 254 silica gel plates. Flash column chromatography was performed on silica gel (SiO_2, Lichroprep Si60, Merck).

a) Chemistry——2-Benzyloxy-3-methoxy-6-nitrobenzoic Acid (**5**): A 10% aqueous solution of potassium permanganate (700 ml) was added dropwise at 40 °C to a solution of 2-benzyloxy-3-methoxy-6-nitrobenzaldehyde (**4**)[11] (100 g, 345 mmol) in acetone (1.4 l). Stirring was continued at 40 °C for 7 h. After filtration to remove the precipitate, acetone was evaporated off under reduced pressure. Acidification with HCl solution afforded a precipitate, which was filtered off, washed with H_2O and purified by dissolution in 3 M KOH solution, treatment with charcoal, filtration and precipitation by HCl solution to give **5** (74 g, 71%), mp 176 °C. IR (KBr): 2900 (ν_{OH}), 1710 (ν_{CO}), 1515 and 1350 (ν_{NO_2}), 1050 (OCH_3) cm^{-1}. ^{1}H-NMR (DMSO-d_6) δ: 3.94 (3H, s, CH_3), 4.98 (2H, s, CH_2), 7.31 (6H, m, C_6H_5 and H_4), 7.91 (1H, d, J = 8 Hz, H_5), 10 (1H, OH). *Anal.* Calcd for $C_{15}H_{13}NO_6$: C, 59.40; H, 4.32; N, 4.62. Found: C, 59.06; H, 4.35; N, 4.75.

6-Amino-2-benzyloxy-3-methoxybenzoic Acid (**6**): A solution of $FeSO_4 \cdot 7H_2O$ (269 g) in H_2O (1.1 l) was added to a boiling solution of **5** (42 g, 149 mmol) in 8 M NH_4OH (1 l). The mixture was refluxed for 0.5 h, then the precipitate was filtered off and washed with H_2O. The filtrate was adjusted to pH 4 with HCl. The resulting solid was filtered off, and washed with H_2O to give **6** (32 g, 79%), which was not further purified because of its instability, mp 76—78 °C. IR (KBr): 3200—2900 (ν_{OH}, $\nu^+_{NH_3}$), 1710 (ν_{CO}), 1590 (ν_{COO^-}), 1050 (OCH_3) cm^{-1}. ^{1}H-NMR (DMSO-d_6) δ: 3.73 (3H, s, OCH_3), 4.96 (2H, s, CH_2), 6.63 (1H, d, J = 8 Hz, H_5), 7.03 (1H, d, J = 8 Hz, H_4), 7.35 (8H, m, C_6H_5 and $\nu^+_{NH_3}$).

5-Benzyloxy-6-methoxy-4(3*H*)-quinazolinone (**7**): A mixture of **6** (30 g, 110 mmol), *s*-triazine (8.9 g, 110 mmol) and piperidine (7.5 ml) in dry EtOH (675 ml) was refluxed under N_2 for 30 h. After the reaction mixture had cooled, the precipitate was separated by filtration, and washed with EtOH and then with ethyl ether to give **7** (24.82 g, 80%), which was used without further purification. Flash chromatography using ethyl acetate and then ethyl acetate–MeOH (9.5 : 0.5) as eluents gave an analytical sample melting at 200 °C. IR (KBr): 330 (ν_{NH}), 1680 (ν_{CO}), 1060 (OCH_3) cm^{-1}. ^{1}H-NMR (DMSO-d_6) δ: 3.79 (3H, s, OCH_3), 4.90 (2H, s, CH_2), 7.1—7.6 (7H, m, C_6H_5, H_7 and H_8), 7.80 (1H,

TABLE III. Physicochemical Data for Bis(4-amino-5-benzyloxy-6-methoxyquinazolines) (**10**)

Compd. No.	mp (°C) (Recryst. solvent)[a]	Yield (%)	IR (cm^{-1})[b]		^{1}H-NMR δ (J=Hz)	Formula	Analysis (%) Calcd (Found)		
			ν_{NH}	OCH_3			C	H	N
10a[c]	196 (B)	74	3390	1090	3.46 (4H, d, J=6, CH_2), 3.91 (6H, s, OCH_3), 5.03 (4H, s, OCH_2), 7—7.25 (10H, m, C_6H_5), 7.38 (2H, d, J=8, H_7), 7.55 (2H, d, J=8, H_8), 8.05 (2H, t, J=6, NH), 8.35 (2H, s, H_2)	$C_{34}H_{32}N_6O_4$	69.38 (69.44	5.48 5.52	14.28 13.92)
10b[c]	123.5 (B–P)	97	3400	1100	1.0 (6H, quint, J=6, CH_2), 1.31 (4H, quint, J=6, NH–CH_2–$\underline{CH_2}$), 3.27 (4H, q, J=6, NH–$\underline{CH_2}$), 3.93 (6H, s, OCH_3), 5.13 (4H, s, OCH_2), 7.3—7.5 (12H, m, C_6H_5, H_7), 7.51 (2H, d, J=9, H_8), 7.89 (2H, t, NH), 8.38 (2H, s, H_2)	$C_{39}H_{42}N_6O_4$	71.10 (71.08	6.43 6.48	12.76 12.68)
10c[d]	Oil[e]	56	3490	1180	2.20 (4H, quint, J=6, NH–CH_2–$\underline{CH_2}$), 2.93 (3H, s, N–CH_3), 3.08 (4H, t, J=6, N(CH_3)$\underline{CH_2}$), 4.22 (4H, q, J=6, NH–$\underline{CH_2}$), 4.89 (6H, s, OCH_3), 6.11 (4H, s, OCH_2), 8.22—8.38 (12H, m, C_6H_5, H_7), 8.55 (2H, d, J=10, H_8), 8.91 (2H, t, J=6, NH), 9.13 (2H, s, H_2)	$C_{39}H_{43}N_7O_4$[f]			
10d[c]	162 (A)	68	3400	1110	0.98—2.8 (18H, m, N⟨⟩), 1.53 (4H, quint, J=6, NH–CH_2–$\underline{CH_2}$), 2.20 (4H, t, J=6, CH_2N<), 3.38 (4H, q, J=6, NH–$\underline{CH_2}$), 3.98 (6H, s, OCH_3), 5.15 (4H, s, OCH_2), 7.3—7.5 (12H, m, C_6H_5, H_7), 7.55 (2H, d, J=8, H_8), 7.95 (2H, t, J=6, NH), 8.38 (2H, s, H_2)	$C_{48}H_{58}N_8O_4 \cdot H_2O$	69.54 (69.47	7.30 7.12	13.52 13.78)
10e[c]	139 (M)	60	3400	1120	3.26 (4H, s, O–CH_2–CH_2–O), 3.40 (4H, q, J=6, NH–$\underline{CH_2}$), 3.53 (4H, t, J=6, NH–CH_2–$\underline{CH_2}$–O), 3.85 (6H, s, OCH_3), 5.07 (4H, s, $O\underline{CH_2}$–C_6H_5), 7.2—7.4 (10H, m, C_6H_5), 7.31 (2H, d, J=8, H_7), 7.47 (2H, d, J=8, H_8), 8.21 (2H, t, J=6, NH), 8.35 (2H, s, H_2)	$C_{38}H_{40}N_6O_6 \cdot 1/4\,H_2O$	66.99 (66.98	5.99 5.96	12.34 12.79)
10f[c]	138 (M)	74		1120	3.38 (8H, s, O–CH_2–CH_2–O), 3.62 (8H, t, J=6, N–CH_2), 3.87 (8H, t, J=6, N–CH_2–$\underline{CH_2}$), 4.80 (4H, s, $O\underline{CH_2}$–C_6H_5), 7.27 (10H, m, C_6H_5), 7.41 (2H, d, J=8, H_7), 7.56 (2H, d, J=8, H_8), 8.38 (2H, s, H_2)	$C_{44}H_{50}N_6O_8 \cdot 3/4\,H_2O$	65.69 (65.49	6.45 6.59	10.45 10.52)

a) Abbreviations used for solvents are: A (acetonitrile), B (benzene), M (MeOH), P (petroleum ether). *b*) IR spectra were measured by the KBr disc method. *c*) ^{1}H-NMR spectra were measured in $CDCl_3$. *d*) ^{1}H-NMR spectra were measured in DMSO-d_6. *e*) Purified by flash chromatography using ethyl acetate then MeOH as eluents. *f*) Because of the instability of **12c**, elemental analysis was not attempted, MS m/z: 674 [$(M+H)^+$].

TABLE IV. Physicochemical Data for Bis(4-amino-5-hydroxy-6-methoxyquinazolines) (**11**)

Compd. No.	Synthetic[a] method	Yield	IR (cm^{-1})[b] ν_{NH}, ν_{OH}	IR (cm^{-1})[b] OCH_3	^{1}H-NMR δ (J=Hz)	Formula
11a[c]	A	85	3000—3400	1130	3.91 (6H, s, OCH_3), 4.13 (4H, m, CH_2), 7.02 (2H, d, J=8, H_7), 7.73 (2H, d, J=8, H_8), 8.66 (2H, s, H_2), 10.04 (2H, m, NH)	$C_{20}H_{20}N_6O_4$
11b[d]	A	Quant	3450	1100	1.33 (6H, quint, J=6, CH_2), 1.60 (4H, quint, J=6, NH–CH_2–CH_2), 3.62 (4H, q, J=6, NH–CH_2), 3.87 (6H, s, OCH_3), 7.0 (2H, d, J=8, H_7), 7.60 (2H, d, J=8, H_8), 8.53 (2H, s, H_2), 10.22 (2H, NH)	$C_{25}H_{30}N_6O_4$
11c[d]	B	Quant	3400	1095	1.82 (4H, quint, J=6, NH–CH_2–CH_2), 2.33 (3H, s, CH_3), 2.64 (4H, t, J=6, $N(CH_3)CH_2$), 3.53 (4H, q, J=6, NH–CH_2), 3.69 (6H, s, OCH_3), 6.38 (2H, d, J=8, H_7), 7.13 (2H, d, J=8, H_8), 8.16 (2H, s, H_2)	$C_{25}H_{31}N_7O_4$
11d[d]	A	Quant	3400	1125	0.71—2.75 (18H, m, N⬡), 2.00 (4H, quint, J=6, NH–CH_2–CH_2), 2.95 (4H, t, J=6, CH_2–N⬡), 3.55 (4H, q, J=6, NH–CH_2), 3.67 (6H, s, OCH_3), 6.42 (2H, d, J=8, H_7), 7.10 (2H, d, J=8, H_8), 8.15 (2H, s, H_2)	$C_{34}H_{46}N_8O_4$
11e[c]	A	80	2900, 3300	1100	3.70 (18H, m, OCH_3 and CH_2), 6.95 (2H, d, J=8, H_7), 7.10 (2H, s, OH), 7.48 (2H, d, J=8, H_8), 8.52 (2H, s, H_2), 9.68 (2H, t, J=6, NH)	$C_{24}H_{28}N_6O_6$
11f[d]	A	93			3.35 (6H, s, OCH_3), 3.50 and 4.50 (24H, m, CH_2), 7.05 (2H, s, OH), 7.18 (2H, d, J=8, H_7), 7.60 (2H, d, J=8, H_8), 8.20 (2H, s, H_2)	$C_{30}H_{38}N_6O_8$

a) Methods A and B are described in the text. *b*) IR spectra were measured by the KBr disc method. *c*) ^{1}H-NMR spectra were measured in DMSO-d_6-TFA. *d*) ^{1}H-NMR spectra were measured in DMSO-d_6.

s, H_2), 11.82 (1H, NH). *Anal.* Calcd for $C_{16}H_{14}N_2O_3$: C, 68.07; H, 5.00; N, 9.93. Found: C, 67.83; H, 5.01; N, 9.81.

5-Benzyloxy-4-chloro-6-methoxyquinazoline (**8**): A mixture of **7** (16 g, 57 mmol), phosphoryl chloride (5.2 ml, 57 mmol) and triethylamine (21 ml, 147 mmol) in dry C_6H_6 (480 ml) was heated at 80°C under N_2 for 3 h. After being poured onto ice, the reaction mixture was extracted with C_6H_6. The solvent layer was washed with H_2O, 10% aqueous $NaHCO_3$ then H_2O until chlorides were eliminated, dried over Na_2SO_4 and evaporated under reduced pressure to afford **8** (13.7 g, 80%), which was used without further purification. Flash chromatography using CH_2Cl_2–ethyl acetate (9.7 : 0.3) as an eluent gave an analytical sample melting at 137.5 °C. ^{1}H-NMR (DMSO-d_6) δ: 3.87 (3H, s, OCH_3), 4.49 (2H, s, CH_2), 7.3—7.6 (6H, m, C_6H_5 and H_7), 7.69 (1H, d, J=8 Hz, H_8), 8.64 (1H, s, H_2). *Anal.* Calcd for $C_{16}H_{13}Cl\ N_2O_2$: C, 63.90; H, 4.36; N, 9.31. Found: C, 63.76; H, 4.73; N, 8.95.

Bis(4-Amino-5-benzyloxy-6-methoxyquinazolines) (**10**). General Procedure: The α,ω-diamine (**9**) (A = N or NH) (5.5 mmol) was added under N_2 to a solution of **8** (10 mmol) and triethylamine (11 mmol) in dry EtOH (25—60 ml). The mixture was stirred for 24 h at room temperature. After removal of the solvent, the residue was washed with ice-cooled EtOH. When **10** was an oil, the reaction mixture was extracted with CH_2Cl_2; the solvent layer was washed with H_2O until the pH was neutral, dried over Na_2SO_4 and evaporated under reduced pressure. Compounds **10** were used directly in the following procedure. A small sample was further purified either by recrystallization or flash chromatography. Yields and physicochemical data are listed in Table III.

Bis(4-Amino-5-hydroxy-6-methoxyquinazolines) (**11**). General Procedures: Method A: A solution of **10** (3 mmol) in TFA (30 ml) was refluxed under N_2 for 5 h. MeOH was added, the solution was evaporated *in vacuo*, the residue was triturated with H_2O and then the mixture was adjusted to pH 9 with concentrated NH_4OH. The precipitate was filtered off and washed with H_2O. In the case of the water-soluble compound (**11f**), after treatment with TFA, the residue was dissolved in water and stirred for 15 min with Amberlite 45 (OH) resin (40 ml). After filtration, H_2O was evaporated off under reduced pressure.

Method B: A solution of **10** (3 mmol) in dioxane–MeOH (1 : 1) was hydrogenated over 10% Pd–C (1.3 g) at room temperature and atmospheric pressure. After removal of the catalyst by filtration and washing of the filtrate with MeOH, the solvent was evaporated off under reduced pressure.

TABLE V. Reaction Conditions and Physicochemical Data for Bis(4-amino-6-methoxy-5,8-quinazolinediones) (**12**)

Compd. No.	Solvent[a] (ml)	Reaction time (h)	mp (°C) Recryst. solvent[b]	Yield (%)	IR (cm^{-1})[c] ν_{NH}	IR (cm^{-1})[c] ν_{CO}	IR (cm^{-1})[c] OCH_3	^{1}H-NMR δ (J=Hz)	Formula	Analysis (%) Calcd (Found) C	H	N
12a[d]	M–H_2O (200)	47	250 (D)	34	3325	1650	1135	3.77 (6H, s, OCH_3), 3.86 (4H, m, CH_2), 6.24 (2H, s, H_7), 8.71 (2H, s, H_2), 9.14 (2H, t, J=6, NH)	$C_{20}H_{16}N_6O_6\cdot$ 1/4 DMF	54.81 (55.07	3.93 4.46	19.26 18.74)
12b[e]	M–H_2O (150)	6	214 (T)	49	3320	1660 1680	1110	1.34 (6H, quint, J=6, CH_2), 1.66 (4H, quint, J=6, NH–CH_2–CH_2), 3.59 (4H, q, J=6, NH–CH_2), 3.86 (6H, s, OCH_3), 6.16 (2H, s, H_7), 8.71 (2H, s, H_2), 8.96 (2H, t, J=6, NH)	$C_{25}H_{26}N_6O_6$	59.29 (58.94	5.17 5.17	16.59 16.35)
12c[e]	AC–H_2O (200)	2.5	215 (A)	33	3350	1650 1660	1140	1.82 (4H, quint, J=6, NH–CH_2–CH_2), 2.25 (3H, s, N–CH_3), 2.44 (4H, t, J=6, N–(CH_3)–CH_2), 3.67 (4H, q, J=6, NH–CH_2), 3.84 (6H, s, OCH_3), 6.09 (2H, s, H_7), 8.89 (2H, s, H_2), 9.19 (2H, t, J=6, NH)	$C_{25}H_{27}N_7O_6\cdot$ $2H_2O$	53.85 (53.76	5.60 5.45	17.59 17.42)
12d[d]	M–H_2O (120)	24	247 (C–P)	66	3340	1650 1670	1125	1.0—2.9 (18H, m, N◯), 1.73 (4H, quint, J=6, NH–CH_2–CH_2), 2.34 (4H, t, J=6, CH_2N<), 3.60 (4H, q, J=6, NH–CH_2), 3.81 (6H, s, OCH_3), 6.38 (2H, s, H_7), 8.76 (2H, s, H_2), 9.19 (2H, t, J=6, NH)	$C_{34}H_{42}N_8O_6$	61.99 (61.93	6.43 6.39	17.01 17.30)
12e[e]	M–H_2O (100)	24	233 (D)	59	3300	1650	1135	3.67 (12H, m, CH_2), 3.80 (6H, s, OCH_3), 6.10 (2H, s, H_7), 8.81 (2H, s, H_2), 9.17 (2H, t, J=6, NH)	$C_{24}H_{24}N_6O_8$	54.96 (54.63	4.61 4.75	16.03 15.89)
12f[d]	M–H_2O (200)	3.5	96 (D–E)	25		1600	1120	3.34—3.91 (24H, m, CH_2), 3.76 (6H, s, OCH_3), 6.08 (2H, s, H_7), 8.13 (2H, s, H_2)	$C_{30}H_{34}N_6O_{10}$[f] H_2O			
12g[d]			235 (H_2O–M–AC)	94	3300	1650 1660	1110	2.0 (4H, quint, J=6, NH–CH_2–CH_2), 2.96 (6H, s, $N^+(CH_3)_2$), 3.29 (4H, t, J=6, N^+–CH_2), 3.60 (4H, q, NH–CH_2), 3.82 (6H, s, OCH_3), 6.31 (2H, s, H_7), 8.77 (2H, s, H_2), 9.07 (2H, t, J=6, NH)	$C_{26}H_{30}IN_7O_6\cdot$ $3/2\,H_2O$	45.25 (45.10	4.81 4.87	14.21 14.14)

a) Abbreviations areas: M–H_2O (methanol–H_2O 6:4), AC–H_2O (acetone–H_2O 6:4). *b*) Abbreviations for solvents are: A (acetonitrile), AC (acetone), C (dichloromethane), D (DMF), E (ethyl ether), M (methanol), P (petroleum ether), T (THF). IR spectra were measured by the KBr disc method. *d*) ^{1}H-NMR spectra were measured in DMSO-d_6. *e*) ^{1}H-NMR spectra were measured in $CDCl_3$. *f*) MS m/z: 656 (M+18).

Compounds **11** were not purified because of their instability in air. Yields and physicochemical data are listed in Table IV.

Bis(4-Amino-6-methoxy-5,8-quinazolinediones) (**12a—f**). General Procedure: Potassium nitrosodisulfonate (2.15 g, 8 mmol) was added with stirring to a solution of **11** (1 mmol) and monobasic potassium phosphate (313 mg, 2.3 mmol) in MeOH–H_2O (6:4) or in acetone–H_2O (6:4). The mixture was stirred for several hours. The reaction was monitored by TLC. After removal of the organic solvent, when necessary, the mixture was adjusted to pH 7 with 10% aqueous $NaHCO_3$ solution, then extracted with CH_2Cl_2. The organic layer was dried on Na_2SO_4 and evaporated off under reduced pressure to give the corresponding quinone (**12**), which was purified by recrystallization. In the case of the extremely water-soluble quinone (**12f**), the reaction mixture was evaporated to dryness, and extracted with CH_2Cl_2 at 20 °C. After removal of the solvent, the residue was dissolved in H_2O and stirred for 1 min with Amberlite 45 (OH) resin (12 ml). After filtration, H_2O was evaporated off under reduced pressure to give extremely hygroscopic **12f**, which was purified by recrystallization from dry solvents. Elemental analysis was not attempted for **12f**, but the molecular formula was confirmed by MS measurement. Yields and physicochemical data are listed in Table V.

4,4′-(5-Azonia-1,9-diaza-5,5-dimethylnonane-1,9-diyl)bis(6-methoxy-5,8-quinazolinedione) Iodide (**12g**): Iodomethane (3 ml) was added to an ice-cooled solution of **12c** (150 mg, 0.3 mmol) in acetone (3 ml). The mixture was stirred for an additional 30 min at 0 °C, then for 27 h at room temperature. After evaporation to dryness, **12g** was recrystallized (Table V).

Bis[4-Amino-6,7-bis(1-aziridinyl)-5,8-quinazolinediones] (**14**). General Procedure: Compound **12** (100 mg) was added under N_2 to either ice-cooled aziridine or an ice-cooled solution of aziridine in MeOH. The mixture was stirred under the conditions (time and temperature) indicated in Table VI. The mixture was evaporated to dryness at 20 °C, and extracted with CH_2Cl_2, then the extract was washed with H_2O until the pH was neutral. After removal of the solvent, the crude purple solid ought to be immediately purified by flash chromatography. The fractions have to be evaporated as thoroughly as possible. The quinones (**14**) were purified by recrystallization. Except for **14e**, elemental analyses were not correct, but the molecular formulae were confirmed by MS measurements: MS *m/z*: 541 $[(M+H)^+]$ and 611 $[(M+H)^+]$ for **14a** and **14b** respectively; for **14c** see Synthesis. Yields and physico-chemical data are listed in Table VI.

Synthesis of 3,3′-(3,6-Dioxaoctamethylene)bis(6-methoxy-4,5,8(3*H*)-quinazolinetrione) (25)——3,3′-(3,6-Dioxaoctamethylene)bis(5-benzyloxy-6-methoxy-4(3*H*)-quinazolinone) (**23**): Compound **7** (2.82 g, 10 mmol) in dry *N*,*N*-dimethylformamide (DMF) (20 ml) was added with stirring under N_2 to sodium hydride (300 mg, 12.5 mmol) in dry DMF (5 ml). After 20 min, 1,8-dichloro-3,6-dioxaoctane (**22**) (2.04 g, 5.5 mmol) in dry DMF (7 ml) was added at 20 °C. The mixture was heated at 50 °C for 15 h, then the solvent was eliminated *in vacuo*. The residue was extracted with CH_2Cl_2, and the extract was washed with H_2O until the pH was neutral, then evaporated under reduced pressure to afford **23** (3.86 g, 57%), mp 127 °C. IR (KBr): 1660 (ν_{CO}), 1120 (OCH_3) cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 3.4 (4H, s, O–CH_2–CH_2–O), 3.65 (4H, t, J=6 Hz, NCH_2CH_2), 3.9 (6H, s, OCH_3), 4.05 (4H, t, J=6 Hz, NCH_2), 7.1 (2H, d, J=8 Hz, H_7), 7.3 (2H, d, J=8 Hz, H_8), 7.9 (2H, s, H_2), 11.7 (2H, s, OH). *Anal.* Calcd for $C_{38}H_{38}N_4O_8$: C, 67.24; H, 5.64; N, 8.26. Found: C, 67.25; H, 5.57; N, 8.25.

3,3′-(3,6-Dioxaoctamethylene)bis(5-hydroxy-6-methoxy-4(3*H*)-quinazolinone) (**24**): The debenzylation of **23** was carried out by the same procedure as used for **11** (method A) to give **24** in 91% yield, mp 146 °C (toluene). IR (KBr): 3050 (ν_{OH}), 1650 (ν_{CO}) cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 3.5 (4H, s, O–CH_2–CH_2–O), 3.65 (4H, t, J=6 Hz, N–CH_2–CH_2), 3.9 (6H, s, OCH_3), 4.05 (4H, t, J=6 Hz, NCH_2), 7.1 (2H, d, J=8 Hz, H_7), 7.3 (2H, d, J=8 Hz, H_8), 7.9 (2H, s, H_2), 11.7 (2H, s, OH). *Anal.* Calcd for $C_{24}H_{26}N_4O_8$: C, 57.82; H, 5.26; N, 11.24. Found: C, 58.14; H, 5.37; N, 10.94.

3,3′-(3,6-Dioxaoctamethylene)bis(6-methoxy-4,5,8(3*H*)-quinazolinetrione) (**25**): Potassium nitrosodisulfonate (2.68 g, 10 mmol) was added under N_2 to a suspension of **24** (4.98 g, 1 mmol) and dibasic sodium phosphate (4.58 g, 12.8 mmol) in H_2O (110 ml). The mixture was stirred for 24 h, then the precipitate was filtered off, and washed with H_2O. A second crop was isolated by extraction of the filtrate with CH_2Cl_2. The quinone **25** was purified by recrystallization from DMF (1 g, 20%), mp 288—290 °C. IR (KBr): 1750 (ν_{CO} quinone), 1660 (ν_{CO} in the 4-position), 1120 (OCH_3) cm^{-1}. ^{1}H-NMR (DMSO-d_6) δ: 3.46 (4H, s, O–CH_2–CH_2–O), 3.59 (4H, t, J=6 Hz, N–CH_2–CH_2), 3.79 (6H, s, OCH_3), 4.10 (4H, t, J=6 Hz, N–CH_2), 6.24 (2H, s, H_7), 8.73 (2H, s, H_2). *Anal.* Calcd for $C_{24}H_{22}N_4O_{10}$: C, 54.75; H, 4.21; N, 10.64. Found: C, 54.47; H, 4.13; N, 10.61.

3,3′-(3,6-Dioxaoctamethylene)bis[8-(4-*N*,*N*-dimethylaminophenylimino)-6-methoxy-4,5-quinazolinedione] (**27**): **24** (150 mg, 0.3 mmol) in suspension in EtOH (18 ml) was added to recently prepared silver chloride (760 mg, 5.3 mmol) and Na_2CO_3 (365 mg, 3.44 mmol) in H_2O (12 ml). Then *N*,*N*-dimethyl-1,4-benzenediamine dihydrochloride (**26**) (126 mg, 0.6 mmol) in H_2O (9 ml) was added under stirring, which was continued for 4.5 h. The precipitate was filtered off, washed with H_2O, dried, then extracted with CH_2Cl_2. A second crop was obtained by extraction of the filtrate with CH_2Cl_2. The whole organic layer was evaporated to give a dark-blue solid, which was recrystallized from CH_2Cl_2–petroleum ether to give **27** (130 mg, 53%), mp 169—175 °C. IR (KBr): 1690 (ν_{CO} quinone and $\nu_{C=N}$), 1600 (ν_{CO} in position 4), 1140 (OCH_3) cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 3.0 (6H, s, $N(CH_3)_2$), 3.43 (4H, s, O–CH_2–CH_2–O), 3.67 (10H, m, NH–CH_2CH_2), 4.15 (4H, t, J=6 Hz, N–CH_2), 6.62 (2H, s, H_7), 6.67—7.09 (8H, 2d, J=8 Hz, C_6H_4), 8.51 (2H, s, H_2). *Anal.* Calcd for $C_{40}H_{42}N_8O_8 \cdot 3H_2O$: C, 58.81; H, 5.92; N, 13.72. Found: C, 58.91; H, 5.81; N,

TABLE VI. Reaction Conditions and Physicochemical Data for Bis[4-amino-6,7-bis(1-aziridinyl)-5,8-quinazolinediones] (**14**)

Compd. No.	Aziridine (ml) [MeOH (ml)]	Reaction conditions			mp (°C) Recryst. solvent[a]	Yield (%)	IR (cm^{-1})[b]		^{1}H-NMR δ (J=Hz)	Formula	Analysis (%) Calcd (Found)		
		θ (°C)	Time (h)	Chromato. solvent[a]			ν_{NH}	ν_{CO}			C	H	N
14a[c]	3	20	24	B–M (91:9)	200	11	3300	1660	2.31 (8H, s, N$\underline{\text{H}}_2$), 2.35 (8H, s, N$\underline{\text{H}}_2$), 3.91 (4H, m, CH_2), 8.77 (2H, s, H_2), 9.24 (2H, t, J=6, NH)	$C_{26}H_{24}N_{10}O_4$			
14b[d]	3, 3	0	9	B–M (95:5)	147 (B–P)	7	3320	1665	1.33 (6H, quint, J=6, CH_2), 1.64 (4H, quint, J=6, $NCH_2C\underline{H}_2$), 2.29 (8H, s, N$\underline{\text{H}}_2$), 2.34 (8H, s, N$\underline{\text{H}}_2$), 8.75 (2H, s, H_2), 8.98 (2H, q, J=6, NH)	$C_{31}H_{34}N_{10}O_4$			
14c[d]	3 [1]	0	6	C–M (90:10)	160 (C–P)	7	3300	1655	1.82 (4H, quint, J=6, NH–CH_2–$C\underline{H}_2$), 2.25 (3H, s, NCH_3), 2.33 (16H, m, N$\underline{\text{H}}_2$), 2.42 (4H, t, J=6, $CH_2N\underline{\text{H}}_2$), 3.65 (4H, q, J=6, NH–CH_2), 8.73 (2H, s, H_2), 9.20 (2H, t, J=6, NH)	$C_{31}H_{35}N_{11}O_4$			
14e[d]	.4	20	5	EA–M (85:15)	123 (C–P)	20	3000	1650	2.31 (16H, s, N$\underline{\text{H}}_2$), 3.64 (4H, s, $OC\underline{H}_2$-$C\underline{H}_2O$), 3.69 (4H, q, J=6, NH–$C\underline{H}_2$), 3.77 (4H, t, J=6, NH–CH_2–$C\underline{H}_2O$), 8.67 (2H, s, H_2), 9.15 (2H, t, J=6, NH)	$C_{30}H_{32}N_{10}O_{16}\cdot 1/2\,CH_2Cl_2$	54.58 (54.73	4.92 4.98	20.88 20.75)

a) Abbreviations of solvents are: EA (ethyl acetate), B (benzene), C (dichloromethane), M (methanol), P (petroleum ether). *b*) IR spectra were measured by the KBr disc method. *c*) ^{1}H-NMR spectra was measured in DMSO-d_6. *d*) ^{1}H-NMR spectra were measured in $CDCl_3$.

13.36.

Synthesis of 1,1′-Bis(3-aminopropyl)-4,4′-bipiperidine (18)——1,1′-Bis[3-(*N*-Phthalimido)propyl]-4,4′-bipiperidine (**17**): The 4,4′-bipiperidine (**16**) (10 g, 41.5 mmol) was added to a suspension of *N*-(3-bromopropyl)phthalimide (**15**) (22.24 g, 83 mmol) and K_2CO_3 (24 g, 174 mmol) in dry acetone (1 l). The mixture was refluxed under N_2 for 15 h. After concentration to half the original volume, the precipitate was filtered off then extracted with CH_2Cl_2. The organic layer was evaporated to dryness to give crude **17** (18.9 g, 84%), which was used without further purification. Recrystallization from EtOH gave an analytical sample melting at 178 °C. IR (KBr): 1700 (ν_{CO}) cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 0.84—2.77 (18H, m, N◯), 1.78 (4H, J=6 Hz, quint, N–CH_2–CH_2), 2.29 (4H, t, J=6 Hz, CH_2–N◯), 3.66 (4H, t, J=6 Hz, N–CH_2), 7.64 (4H, dd, J=6 Hz, Ar-H), 7.87 (4H, dd, J=8 Hz, Ar-H). *Anal.* Calcd for $C_{32}H_{38}N_4O_4 \cdot 1/4H_2O$: C, 70.24; H, 7.09; N, 10.24. Found: C, 69.89; H, 7.23; N, 10.19.

1,1′-Bis(3-aminopropyl)-4,4′-bipiperidine (**18**): A mixture of **17** (10 g, 185 mmol), hydrazine hydrate (20 ml, 412 mmol) in EtOH (200 ml) was refluxed for 3 h. After removal of the solvent, excess hydrazine hydrate was eliminated by addition of EtOH and evaporation, five times. The residue was dissolved in EtOH (200 ml), and the mixture was adjusted to pH 1 with HCl then refluxed for 0.5 h. After cooling, the white precipitate was collected by filtration, and washed with H_2O. The H_2O was evaporated down to 150 ml, and the solution was adjusted to pH 12.5—13 with NaOH pellets, then evaporated to dryness. The solid was dried over KOH pellets, then extracted with benzene. Evaporation of the solvent gave 4.85 g (93%) of crude **18**. IR (KBr): 3380, 3270 (ν_{NH_2}) cm^{-1}. ^{1}H-NMR (DMSO-d_6–TFA) δ: 1.18—3.62 (18H, m, N◯), 1.95 (4H, quint, J=6 Hz, N–CH_2–CH_2), 2.87 (8H, m, N–CH_2), 7.80 (4H, t, J=6 Hz, NH_2). Compound **18** was purified by transformation to its tetrahydrochloride, mp 300 °C (MeOH–ethyl acetate). *Anal.* Calcd for $C_{16}H_{38}N_4 \cdot 4HCl \cdot 1/4H_2O$: C, 43.94; H, 8.99; N, 12.81. Found: C, 43.97; H, 8.99; N, 13.22.

b) Pharmacology——Growth Inhibition of L1210 Cells in Culture: The experimental protocol has been reported.[4] The cells were exposed to increasing concentrations of drugs dissolved in dimethyl sulfoxide (DMSO) (1% final concentration) and incubated at 37°C without agitation in a 5% CO_2 atmosphere. Cells either in the presence of a heterocyclic quinone or in its absence (control) were counted in triplicate after 48 h of culturing with a Coultronics counter. The cytotoxic activity of the compounds was measured by determining the drug concentration which decreased the growth rate of L1210 cells to 50% of that of control cells. The IC_{50} was estimated from equations obtained by plotting the logarithm of the drug concentration *versus* the probit of the percentage inhibition of the growth.

Antitumor Activity: The compounds were studied on P388 lymphocytic leukemia. Experiments were performed on 2-month-old CD2F1 mice (Balb C × DBA/2). The antitumor tests were performed using the protocols and evaluation criteria established by the NCI. P388 leukemia cells (10^6) were inoculated i.p. on day 0 in mice. Treatments were given i.p. on day 1. The drugs were inoculated as suspensions in 0.4% hydroxypropylcellulose (Klucel J. F. Hercules Co.) solution in water. The mortality was checked daily for 30 d. Antitumor activity was expressed as T/C%; T is the median survival time of treated animals and C is the median survival time of control animals. Antitumor activity is considered significant when T/C × 100 ≥ 125%. T/C value ≤ 85% indicates toxicity.

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(**15**) to give the corresponding 1,1′-bis[3-(*N*-phthalimido)propyl]-4,4′-bipiperidine (**17**) followed by removal of the phthaloyl group with hydrazine hydrate in the usual way.

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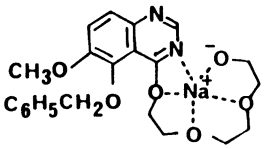

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