Bisquinolines. 2. Antimalarial *N*,*N*-Bis(7-chloroquinolin-4-yl)heteroalkanediamines

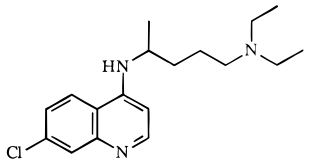
Jonathan L. Vennerstrom,*,† Arba L. Ager, Jr.,§ Arnulf Dorn,|| Steven L. Andersen,‡ Lucia Gerena,‡ Robert G. Ridley,|| and Wilbur K. Milhous‡

College of Pharmacy, University of Nebraska Medical Center, 600 South 42nd Street, Omaha, Nebraska 68198-6025, Department of Microbiology and Immunology, University of Miami School of Medicine, Miami, Florida 33136, Division of Experimental Therapeutics, Walter Reed Army Institute of Research, Washington, D.C., 20307-5100, and Pharma Division, Preclinical Research, F. Hoffmann-LaRoche Ltd., CH-4070 Basel, Switzerland

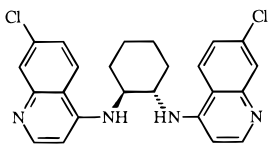
Received June 24, 1998

N,*N*-Bis(7-chloroquinolin-4-yl)heteroalkanediamines **1**−**11** were synthesized and screened against *Plasmodium falciparum* in vitro and *Plasmodium berghei* in vivo. These bisquinolines had IC_{50} values from 1 to 100 nM against *P. falciparum* in vitro. Six of the 11 bisquinolines were significantly more potent against the chloroquine-resistant W2 clone compared to the chloroquine-sensitive D6 clone. For bisquinolines **1**−**11** there was no relationship between the length of the bisquinoline heteroalkane bridge and antimalarial activity and no correlation between in vitro and in vivo antimalarial activities. Bisquinolines with alkyl ether and piperazine bridges were substantially more effective than bisquinolines with alkylamine bridges against *P. berghei* in vivo. Bisquinolines **1**−**10** were potent inhibitors of hematin polymerization with IC_{50} values falling in the narrow range of 5−20 μM, and there was a correlation between potency of inhibition of hematin polymerization and inhibition of parasite growth. Compared to alkane-bridged bisquinolines (Vennerstrom et al., 1992), none of these heteroalkane-bridged bisquinolines had sufficient antimalarial activity to warrant further investigation of the series.

Although bisquinolines such as piperaquine, hydroxypiperaquine, and dichloroquinazine are active against chloroquine-resistant *Plasmodium falciparum*, none have seen extensive clinical use in malaria chemotherapy.[1] Several years ago we initiated a reinvestigation of antimalarial bisquinolines when we disclosed antimalarial data for 13 new *N*,*N*-bis(7-chloroquinolin-4-yl)alkanediamines.[2] Six of the 13 bisquinolines had superior in vitro and in vivo antimalarial activity compared to chloroquine, and one, (±)-*trans*-N^1,N^2-bis-(7-chloroquinolin-4-yl)cyclohexane-1,2-diamine, was uniquely effective. This bisquinoline, WR 268,668 (Ro 48−6910), is a very effective inhibitor of hematin polymerization[3] and, like chloroquine and other antimalarial 4-aminoquinolines,[3,4] may exert its antimalarial properties by this mechanism. Although a subsequent study[5] revealed some cross-resistance between bisquinoline WR 268,668 and chloroquine in vitro, its *S*,*S* enantiomer (Ro 47−7737) underwent extensive preclinical evaluation at F. Hoffmann-LaRoche Ltd.,[6] but phototoxicity precluded its further development.



Chloroquine



WR 268,668 (Ro 48-6910)

Nevertheless, interest in antimalarial bisquinolines continues.[7−9] For example, Ismail et al.[8] disclosed the antimalarial properties of three analogues of WR 268,-668, while Cowman et al.[9] described the excellent activity of a novel bisquinolinemethanol which was regrettably too toxic for further investigation. Raynes et al.[7] prepared nine bisquinoline bisamides with low resistance indices, the most potent of which had IC_{50} values in the 100 nM range against *P. falciparum* in vitro. A subsequent investigation[10] revealed an excellent correlation between inhibition of parasite growth and inhibition of hematin polymerization for these nine bisquinolines.

In this work, we describe the synthesis of a series of 11 *N*,*N*-bis(7-chloroquinolin-4-yl)heteroalkanediamine bisquinolines and examine the effects of incorporating oxygen and nitrogen atoms in the alkane bridge on antimalarial activity and inhibition of hematin polymerization.

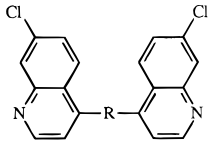
Chemistry

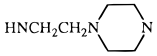
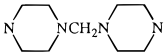
Bisquinolines **1**−**10** were obtained in yields of 12−86% (Table 1) via a displacement reaction with 4,7-dichloroquinoline, heteroalkanediamine, and triethylamine in a 2:1:2 molar ratio in refluxing *N*-methylpyrrolidinone.[2,11] Bisquinolines **1**−**10** were isolated by adding water and ethyl ether or ethyl acetate to the cooled reaction mixtures which initiated product precipitation and dissolved any unreacted starting materials. In some cases, crystallization was required to obtain an analytically pure sample (Table 1). Bisquinoline **11** was obtained by treatment of 7-chloro-4-(piperazin-1-yl)quinoline (**12**)[12] with aqueous formaldehyde. For only bisquinoline **9** was flash column chromatography required for purification. For the alkyl ether-bridged bisquinolines **1**−**4**, melting point decreased as the length of the bridge increased; a similar

† University of Nebraska Medical Center.
‡ Walter Reed Army Institute of Research.
§ University of Miami.
|| F. Hoffmann-LaRoche Ltd.

10.1021/jm9803828 CCC: $15.00 © 1998 American Chemical Society
Published on Web 09/25/1998

Table 1. Heteroalkane-Bridged Bisquinolines **1**−**11**



compd	R	mp (°C)	yield (%)	crystallization solvent
1	$HN(CH_2)_2O(CH_2)_2NH$	257−262 dec	33	
2	$HN(CH_2)_2O(CH_2)_2O(CH_2)_2NH$	181−182	79	CH_3CN
3	$HN(CH_2)_3O(CH_2)_4O(CH_2)_3NH$	156−166[a]	86	
4	$HN(CH_2)_3O(CH_2)_2O(CH_2)_2O(CH_2)_3NH$	99−106	23	CH_3CN
5	$HN(CH_2)_2NH(CH_2)_2NH$	248−253 dec	33	EtOH
6	$HN(CH_2)_3NH(CH_2)_2NH$	255−258 dec	37	MeOH
7	$HN(CH_2)_3NH(CH_2)_3NH$	199−201[b]	26	EtOH
8	$HN(CH_2)_3NCH_3(CH_2)_3NH$	162−167[c]	50	
9	$HN(CH_2)_6NH(CH_2)_6NH$	134−137	12	CH_3CN
10		216−218	54	
11		237−239	65	$CH_3CN/CHCl_3$

[a] Lit.[19] mp 124−126 °C. [b] Softening at 76−85 °C. [c] Softening at 115−118 °C.

Table 2. Antimalarial Activity and Inhibition of Hematin Polymerization for Bisquinolines **1**−**11**

compd	hematin polymerization IC_{50} (μM)	*P. falciparum* IC_{50} (nM) D6, W2	number of *P. berghei*-infected mice dead/day died[a] 40 mg/kg[b]	160 mg/kg	640 mg/kg
1	20 ± 1	16, 89	1/7 3/8 1/10	1/8 2/9 1/10 1/18	2/13 1/14 1/24 1/29[c]
2	15 ± 1	2.5, 9.9	2/7 2/9 1/13	3/8 1/9 1/C	1/T 1/6 1/13 1/15 1/C
3	19 ± 1	22, 21	1/7 1/8 2/9 1/10	2/10 1/11 1/12 1/26	3/12 2/13
4	20 ± 1	24, 48	1/8 3/9 1/11	2/12 1/13 2/14	1/14 1/19 3/C[c]
5	20 ± 1	30, 8.9	4/6 1/7	3/6 1/7 1/11	3/6 2/7
6	16 ± 2	26, 7.0	4/6 1/7	5/6	5/6
7	14 ± 1	86, 26	2/6 3/7	1/6 3/7 1/8	3/8 1/10 1/12
8	8 ± 1	5.9, 2.8	2/8 2/9 1/11	3/13 2/18	1/12 1/13 1/14 1/16 1/33
9	7 ± 1	11, 5.4	4/6 1/7	2/6 3/7	5/7
10	5 ± 1	1.2, 9.0	1/8 1/9 1/12 2/13	3/13 1/14 1/17 1/C	1/16 1/18 1/20 1/22 1/C
11	>2500	>1000, >1000	1/6 4/7	3/9 1/10 1/16	1/14 1/22 3/C
chloroquine[d]	80 ± 3	6.5, 99	1/12 2/14 1/15 1/16	1/16 1/18 1/20 1/23 1/C	3/T 1/25 1/C

[a] Controls died on day 6 or 7 postinfection, survival beyond 60 days was considered curative (C), and deaths from 0−2 days posttreatment were attributed to toxicity (T). [b] Compounds were administered in a single sc dose 3 days postinfection, $n = 5$. [c] Skin lesions observed at site of injection. [d] In vivo data for chloroquine was obtained from multiple experiments at WRAIR.

trend was observed for the secondary alkylamine-bridged bisquinolines **5**−**7** and **9**.

Antimalarial Activity

These bisquinolines had IC_{50} values from 1 to 100 nM against *P. falciparum* in vitro (Table 2). Only bisquinolines **1**, **2**, and **10** were cross-resistant with chloroquine, but in each case the potency difference between the D6 and W2 clones was less than that observed for chloroquine. Bisquinolines **3** and **4** were roughly equipotent against the two *P. falciparum* clones, while the remaining compounds were more active against the chloroquine-resistant W2 clone. For bisquinolines **1**−**11**, there was a poor correlation between in vitro and in vivo antimalarial activities, especially evident for bisquinoline **11**. It was also clear that those bisquinolines with alkyl ether (**1**−**4**) and piperazine (**10**, **11**) bridges were substantially more effective than bisquinolines with alkylamine (**5**−**9**) bridges against *Plasmodium berghei* in vivo. There was no relationship between the length of the bisquinoline heteroalkane bridge and antimalarial activity. Skin lesions at the site of injection were observed for bisquinolines **1** and **4** at the 640 mg/kg dose.

Inhibition of Hematin Polymerization

Bisquinolines **1**−**10** were potent inhibitors of hematin polymerization with IC_{50} values falling in the narrow range of 5−20 μM (Table 2). The complete lack of inhibition of hematin polymerization for bisquinoline **11** corresponded to its complete lack of activity against *P. falciparum* in vitro. We next analyzed our data to assess if the potency of inhibition of hematin polymerization by these bisquinolines correlated with the potency of inhibition of parasite growth in culture. As it is probable that resistance to 4-aminoquinolines (and perhaps bis(4-aminoquinolines)) is associated with reduced drug permeability,[13] we took the lowest IC_{50} value against the D6 and W2 parasite clones for a given bisquinoline to use in the analysis, assuming that drug uptake was optimal for that clone. For bisquinolines **1**−**10** we observed a modest correlation ($r = 0.61$, $p = 0.06$) between the potency of inhibition of hematin polymerization and inhibition of parasite growth.[14]

Discussion

On balance, incorporation of nitrogen and oxygen atoms in the bisquinoline bridge did not improve antimalarial activity in these bisquinolines. If one compares

the activity of each of these heteroalkane-bridged bisquinolines with their alkane-bridged bisquinoline counterparts described earlier,[2] only for ether-bridged bisquinolines **2**−**4** was in vivo antimalarial efficacy enhanced, and only for bisquinoline **9** was in vitro antimalarial potency increased. The ether functional groups in **2**−**4** may have endowed these bisquinolines with increased water solubility and perhaps better absorption compared to their purely alkane-bridged analogues.[2] For the remaining heteroalkane-bridged bisquinolines, activity was no better or in some cases was substantially lower. This loss of in vivo antimalarial activity was especially notable for bisquinolines **5**−**9** with amine functional groups in the connecting bridge. We suspect that this structural feature probably allows for rapid N-dealkylation metabolism which would convert these bisquinolines to monoquinolines. Bisquinolines **5**−**9** would thus offer no advantages over chloroquine and other monoquinolines, as monoquinoline metabolic products are uniformly less active than the parent drug, particularly against drug-resistant parasites.[15]

The hypothesis that bisquinolines[3,10] inhibit parasite growth via inhibition of hematin polymerization is supported by our data. However, this also requires that these diprotic and triprotic bisquinolines, like the diprotic weak base chloroquine,[16] concentrate to micromolar levels in the food vacuole, the organelle in which hematin polymerization takes place. Whether these bisquinolines meet this criterion is not known. Recent data[17] implies that if differences in bisquinoline accumulation were to be considered, the correlation between inhibition of parasite growth and inhibition of hematin polymerization would likely improve.

We hypothesize that these bisquinolines, like bisquinoline Ro 48−6910 (WR 268,668), inhibit hematin polymerization by binding to hematin μ-oxo dimer, shifting the equilibrium between hematin monomer and hematin μ-oxo dimer, thereby decreasing hematin monomer incorporation into hemozoin.[18] However, bisquinoline−hematin μ-oxo binding may have other consequences which may or may not contribute to their antimalarial properties. One example is inhibition of both glutathione- and glutathione/hydrogen peroxide-mediated iron release from hematin and inhibition of hematin-dependent lipid peroxidation by bisquinoline **3** as shown by Scott et al.[19] Another is inhibition of polyamine transport[20] as was observed for bisquinoline N^1,N^4-bis(7-chloroquinolin-4-yl)-1,4-diaminobutane.[2]

In summary, compared to alkane-bridged bisquinolines,[2] none of these heteroalkane-bridged bisquinolines had sufficient antimalarial activity to warrant further investigation of the series. However one promising outcome was that 6 of the 11 bisquinolines were significantly more potent against the chloroquine-resistant W2 clone compared to the chloroquine-sensitive D6 clone, an unusual occurence for 4-aminoquinolines of any type. This work also adds further evidence to support the hypothesis that bisquinolines inhibit parasite growth via inhibition of hematin polymerization.

Experimental Section

Melting points were taken with a Mel-Temp capillary apparatus. Except where noted, IR spectra were run as KBr disks on a Perkin Elmer 1420 spectrophotometer. NMR spectra were obtained with either Varian XL-300 or Bruker AC-200 spectrometers using deuteriated dimethyl sulfoxide for **1**−**9** and deuteriated chloroform for **10** and **11** with TMS as an internal standard. It was not possible to obtain ^{13}C NMR spectra for **1** due to its low solubility in DMSO. Microanalyses were performed by M-H-W Laboratories, Phoenix, AZ. The purity of **1**−**11** was confirmed with silica gel (75:24:1 $CHCl_3$−CH_3OH−NH_4OH) or alumina (95:5 $CHCl_3$−CH_3OH) TLC. The heteroalkanediamines were obtained from Aldrich Chemical Co., E. I. DuPont De Nemours and Co. Inc., BASF Corp., and the Texaco Chemical Co. 7-Azatridecane-1,13-diamine (DuPont BHMT amine) was purified by vacuum distillation. All other reagents were obtained from Aldrich Chemical Co.

Synthesis of 1−10. A solution of 4,7-dichloroquinoline (10 mmol, 1.98 g), triethylamine (10 mmol, 1.01 g), and diaminoheteroalkane (5 mmol) in *N*-methylpyrrolidinone (10 mL) was subject to 10 purge cycles using a Firestone valve and then heated to reflux for 2−6 h under a slight positive N_2 pressure. After the reaction mixture cooled to room temperature, ether or ethyl acetate (15 mL) and water (15 mL) were added with stirring, and the resulting solid was filtered and washed with water and ethyl acetate or ether to provide **1**−**10**. In some cases, cooling of this two-phase mixture was required to induce precipitation of product.

***N*^{1},*N*^{5}-Bis(7-chloroquinolin-4-yl)-3-oxapentane-1,5-diamine (1):** ^{1}H NMR δ 3.38−3.61 (m, 4H), 3.73 (t, J = 5.7 Hz, 4H), 6.50 (d, J = 5.4 Hz, 2H), 7.32 (t, J = 5.1 Hz, 2H), 7.43 (dd, J = 9.0, 2.1 Hz, 2H), 7.80 (d, J = 2.1 Hz, 2H), 8.25 (d, J = 9.1 Hz, 2H), 8.37 (d, J = 5.4 Hz, 2H). Anal. ($C_{22}H_{20}Cl_2N_4O$) C, H, N.

***N*^{1},*N*^{8}-Bis(7-chloroquinolin-4-yl)-3,6-dioxaoctane-1,8-diamine (2):** ^{1}H NMR δ 3.41 (dt, J = 5.6, 5.4 Hz, 4H), 3.58 (s, 4H), 3.66 (t, J = 5.6 Hz, 4H), 6.47 (d, J = 5.4 Hz, 2H), 7.32 (t, J = 5.4 Hz, 2H), 7.42 (dd, J = 9.0, 2.2 Hz, 2H), 7.77 (d, J = 2.2 Hz, 2H), 8.24 (d, J = 9.0 Hz, 2H), 8.37 (d, J = 5.4 Hz, 2H); ^{13}C NMR δ 42.23, 68.11, 69.74, 98.68, 117.38, 123.94, 123.98, 127.48, 133.32, 149.05, 149.99, 151.79. Anal. ($C_{24}H_{24}Cl_2N_4O_2$) C, H, N.

***N*^{1},*N*^{12}-Bis(7-chloroquinolin-4-yl)-4,9-dioxadodecane-1,12-diamine (3):** ^{1}H NMR δ 1.57 (br s, 4H), 1.77−2.03 (m, 4H), 3.18−3.73 (m, 12H), 6.46 (d, J = 5.5 Hz, 2H), 7.19−7.39 (m, 2H), 7.44 (dd, J = 9.0, 2.0 Hz, 2H), 7.79 (d J = 1.8 Hz, 2H), 8.26 (d, J = 9.0 Hz, 2H), 8.39 (d, J = 5.3 Hz, 2H); ^{13}C NMR δ 26.08, 28.21, 39.58, 67.67, 69.90, 98.50, 117.41, 123.96, 127.45, 133.31, 149.04, 150.08, 151.84. Anal. ($C_{28}H_{32}Cl_2N_4O_2$) C, H, N.

***N*^{1},*N*^{13}-Bis(7-chloroquinolin-4-yl)-4,7,10-trioxatridecane-1,13-diamine (4):** ^{1}H NMR δ 1.84−2.11 (m, 4H), 3.16−3.47 (m, 4H), 3.48−3.83 (m, 12H), 5.93−6.15 (m, 2H), 6.31 (d, J = 5.4 Hz, 2H), 7.31 (dd, J = 9.0, 2.1 Hz, 2H), 7.70 (d, J = 9.0 Hz, 2H), 7.92 (d, J = 2.1 Hz, 2H), 8.48 (d, J = 5.4 Hz, 2H); ^{13}C NMR δ 28.10, 42.27, 70.31, 70.46, 70.74, 98.60, 117.34, 121.60, 124.95, 128.55, 134.63, 149.09, 150.03, 152.05. Anal. ($C_{28}H_{32}Cl_2N_4O_3$) C, H, N.

***N*^{1},*N*^{5}-Bis(7-chloroquinolin-4-yl)-2-azapentane-1,5-diamine (5):** ^{1}H NMR δ 2.88 (t, J = 6.3 Hz, 4H), 3.04−3.64 (m, 4H), 6.47 (d, J = 5.5 Hz, 2H), 7.11−7.35 (m, 2H), 7.39 (dd, J = 8.9, 2.1 Hz, 2H), 7.77 (d, J = 2.1 Hz, 2H), 8.20 (d, J = 9.0 Hz, 2H), 8.36 (d, J = 5.4 Hz, 2H); ^{13}C NMR δ 42.63, 47.07, 98.70, 117.43, 123.99, 127.50, 133.37, 149.06, 150.15, 151.83. Anal. ($C_{22}H_{21}Cl_2N_5$) C, H, N.

***N*^{1},*N*^{6}-Bis(7-chloroquinolin-4-yl)-3-azahexane-1,6-diamine (6):** ^{1}H NMR δ 1.78−1.92 (m, 2H), 2.72 (t, J = 6.5 Hz, 2H), 2.86 (t, J = 6.5 Hz, 2H), 3.26−3.49 (m, 4H), 6.47 (d, J = 5.6 Hz, 1H), 6.53 (d, J = 5.5 Hz, 1H), 7.22−7.32 (m, 1H), 7.39 (dd, J = 9.0, 2.3 Hz, 1H), 7.44 (dd, J = 9.0, 2.2 Hz, 1H), 7.47−7.58 (m, 1H), 7.78 (d, J = 2.2 Hz, 1H), 7.80 (d, J = 2.2 Hz, 1H), 8.21 (d, J = 9.0 Hz, 1H), 8.25 (d, J = 9.1 Hz, 1H), 8.38 (d, J = 5.4 Hz, 1H), 8.41 (d, J = 5.4 Hz, 1H); ^{13}C NMR δ 28.10, 41.00, 42.55, 47.03, 47.38, 98.52, 98.66, 117.41, 123.94, 127.46, 133.26, 133.32, 149.05, 150.08, 150.13, 151.87. Anal. ($C_{23}H_{23}Cl_2N_5$) C, H, N.

***N*[1],*N*[5]-Bis(7-chloroquinolin-4-yl)-4-azaheptane-1,7-diamine (7):** ^{1}H NMR δ 1.72−1.95 (m, 4H), 2.68 (t, J = 6.5 Hz, 4H), 3.33 (t, J = 6.5 Hz, 4H), 6.45 (d, J = 5.5 Hz, 2H), 7.43, (dd, J = 9.2, 2.0 Hz, 2H), 7.52 (br s, 2H), 7.79 (d, J = 2.2 Hz, 2H), 8.23 (d, J = 9.0 Hz, 2H), 8.38 (d, J = 5.4 Hz, 2H); ^{13}C NMR δ 27.97, 41.06, 47.37, 98.53, 117.41, 123.92, 123.94, 127.49, 133.29, 149.05, 150.07, 151.86. Anal. ($C_{24}H_{25}Cl_2N_5$) C, H, N.

***N*[1],*N*[7]-Bis(7-chloroquinolin-4-yl)-*N*[4]-methyl-4-azaheptane-1,7-diamine (8):** ^{1}H NMR δ 1.70−1.96 (m, 4H), 2.23 (s, 3H), 2.46 (t, J = 6.7 Hz, 4H), 3.16−3.40 (m, 4H), 6.40 (d, J = 5.5 Hz, 2H), 7.43 (dd, J = 9.0, 2.3 Hz, 2H), 7.49 (d, J = 2.2 Hz, 2H), 8.21 (d, J = 9.0 Hz, 2H), 8.36 (d, J = 5.4 Hz, 2H); ^{13}C NMR δ 25.44, 40.93, 41.86, 55.08, 98.49, 117.41, 123.89, 123.97, 127.48, 133.29, 149.03, 150.05, 151.84. Anal. ($C_{25}H_{27}Cl_2N_5$) C, H, N.

***N*[1],*N*[13]-Bis(7-chloroquinolin-4-yl)-7-azatridecane-1,13-diamine (9).** The crude reaction product was purified by column chromatography using neutral alumina eluting successively with 99:1 and then 99:5 $CHCl_3$−CH_3OH. Bisquinoline **9** was collected during elution with the 99:5 solvent mixture: ^{1}H NMR δ 1.34 (br s, 12H), 1.60−1.67 (m, 4H), 2.42−2.46 (m, 4H), 3.12−3.27 (m, 4H), 6.44 (d, J = 5.4 Hz, 2H), 7.29 (t, J = 5.1 Hz, 2H), 7.44 (dd, J = 9.0, 2.4 Hz, 2H), 7.78 (d, J = 2.4 Hz, 2H), 8.28 (d, J = 9.0 Hz, 2H), 8.38 (d, J = 5.4 Hz, 2H); ^{13}C NMR δ 26.61, 26.67, 27.77, 29.59, 42.36, 49.40, 98.55, 117.43, 123.89, 124.08, 127.45, 133.29, 149.10, 150.05, 151.87. Anal. ($C_{30}H_{37}Cl_2N_5$) C, H, N.

***N*[1]-[(7-Chloroquinolin-4-yl)aminoethyl]-*N*[4]-(7-chloroquinolin-4-yl)piperazine (10):** ^{1}H NMR δ 2.70−2.86 (m, 6H), 3.11−3.27 (br s, 4H), 3.40−3.56 (m, 2H), 6.55 (d, J = 5.5 Hz, 1H), 6.98 (d, J = 5.1 Hz, 1H), 7.30 (t, J = 5.1 Hz, 1H), 7.48 (dd, J = 9.0, 2.2 Hz, 1H), 7.55 (dd, J = 9.1, 2.1 Hz, 1H), 7.84 (d, J = 2.2 Hz, 1H), 8.00 (d, J = 2.2 Hz, 1H), 8.02 (d, J = 8.8 Hz, 1H), 8.28 (d, J = 9.1 Hz, 1H), 8.45 (d, J = 5.4 Hz, 1H), 8.71 (d, J = 5.0 Hz, 1H); ^{13}C NMR δ 51.78, 52.65, 53.65, 98.76, 109.38, 117.44, 121.36, 123.96, 124.14, 125.72, 126.02, 127.55, 128.07, 133.42, 133.55, 149.08, 149.62, 149.99, 151.97, 152.16, 156.25. Anal. ($C_{24}H_{23}Cl_2N_5 \cdot 0.5H_2O$) C, H, N.

Bis[4-(7-chloroquinolin-4-yl)piperazin-1-yl]methane (11). Aqueous formaldehyde (37%) (1.5 mmol, 0.045 g) was added to a solution of 7-chloro-4-(piperazin-1-yl)quinoline (1.0 mmol, 0.248 g) in MeOH (1 mL) and the mixture stirred for 5 h at room temperature. $CHCl_3$ (15 mL) was added, and the solution was dried over Na_2SO_4. Removal of solvent in vacuo provided a viscous oil which was dissolved in hot $CHCl_3$ (1 mL). To this solution was added CH_3N (10 mL) which initiated crystallization of **11** (0.164 g, 65%) as white needles: ^{1}H NMR ($CDCl_3$) δ 2.85 (t, J = 5.4 Hz, 8H), 3.28 (t, J = 5.4 Hz, 8H), 3.19 (s, 2H), 6.85 (d, J = 5.4 Hz, 2H), 7.42 (dd, J = 9.0, 1.8 Hz, 2H), 7.97 (d, J = 9.0 Hz, 2H), 8.04 (d, J = 1.8 Hz, 2H), 8.72 (d, J = 5.4 Hz, 2H); ^{13}C NMR ($CDCl_3$) δ 51.38 (t), 52.24 (t), 80.74 (t), 108.96 (d), 121.97 (s), 125.22 (d), 126.03 (d), 128.90 (d), 134.81 (s), 150.19 (s), 151.92 (d), 157.02 (s). Anal. ($C_{27}H_{28}N_6Cl_2$) C, H, N.

7-Chloro-4-(piperazin-1-yl)quinoline (12). A solution of 4,7-dichloroquinoline (0.03 mol, 5.94 g) and piperazine (0.3 mol, 25.84 g) in 2-ethoxyethanol (30 mL) was refluxed under Ar for 24 h. The mixture was cooled and then reheated to distill off the solvent and excess piperazine. Water (100 mL) and 1 N KOH were added, and the aqueous layer was extracted twice with 150-mL portions of a 1:1:1 mixture of EtOAc−Et_2O−CH_2Cl_2. The combined organic layers were washed with 75 mL of brine and dried over K_2CO_3. Solvent removal and prolonged evacuation under low pressure provided **12** (6.68 g, 90%) as a pale-yellow crystalline solid. Recrystallization from ether afforded an off-white crystalline solid: mp 113−115 °C (lit.[12] mp 113−115 °C); ^{1}H NMR ($CDCl_3$) δ 3.18 (s, 8H), 6.83 (d, J = 5.4 Hz, 1H), 7.41 (dd, J = 9.0, 1.8 Hz, 1H), 7.96 (d, J = 9.0 Hz, 1H), 8.03 (d, J = 1.8 Hz, 2H), 8.72 (d, J = 5.4 Hz, 2H); ^{13}C NMR ($CDCl_3$) δ 46.01, 53.49, 108.85, 121.86, 125.16, 125.97, 128.97, 134.70, 150.08, 151.87, 157.29.

Antimalarial Screens. In vitro activity against *P. falciparum* was determined using a modification of the semiautomated microdilution technique of Desjardins et al.[21] and Milhous et al.[22] Two *P. falciparum* malaria parasite clones,[23] designated as Sierra Leone (D6) and Indochina (W2), were used in susceptibility testing. The former is resistant to mefloquine and the latter to chloroquine, pyrimethamine, sulfadoxine, and quinine. Test compounds were dissolved in dimethyl sulfoxide and solutions serially diluted with culture media. Erythrocytes with 0.25−0.5% parsitemia were added to each well of a 96-well microdilution plate to give a final hematocrit of 1.5%. Inhibition of uptake of tritiated hypoxanthine was used as an index of antimalarial activity. Results are reported as IC_{50} (ng/mL) values.

In vivo activity against *P. berghei* was obtained against a drug-sensitive strain of *P. berghei* (strain KBG 173).[24] Each test compound was administered sc to five male mice per dilution in a single subcutaneous dose 3 days after infection. Untreated mice survived on average 6.2 days. Compounds were classified as curative (C) when one or more test animals lived 60 days postinfection. Deaths from 0−2 days posttreatment were attributed to toxicity (T).

Hematin Polymerization Assay. Hematin polymerization experiments were performed as described by Dorn et al.[3,4c] with *P. falciparum* K1 trophozoite acetonitrile extract to initiate the reaction. Bisquinolines were added to the reaction mixtures as DMSO solutions up to a maximum DMSO concentration of 10%. The values of triplicate assays were plotted semilogarithmically (DeltaGraph Pro 3.5 and CA-Cricket Graph III 1.5.2) and the IC_{50} values (μM) calculated graphically ± SD.

Acknowledgment. This work was funded in part by DHHS/NIH/NIAID Grant 1 R15 AI28012-01 to J. L. Vennerstrom and Army Contract DAMD 17-91-1010 to A. L. Ager. 3-Azahexane-1,6-diamine, 7-azatridecane-1,13-diamine, and 4-azaheptane-1,7-diamine samples were graciously provided by Grace A. Pulci, Jeff Weeks, Sally McCoach, Frank E. Herkes, and Robert Smiley of E. I. DuPont De Nemours and Co. Inc. Bis(3-aminopropyl)methylamine, 3-azapentane-1,5-diamine, 4,9-dioxadodecane-1,12-diamine, 3-azahexane-1,6-diamine, and 4,7,10-trioxatridecane-1,13-diamine were graciously provided by Gordon Ross, John Banger, and Robert J. Steffl of BASF Corp. *N*-Aminoethylpiperazine and 3,6-dioxaoctane-1,8-diamine were graciously provided by Gordon Neville, Howard P. Klein, and Phillip B. Valkovich of Texaco Chemical Co.

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JM9803828