Synthesis and Antimalarial Activity of a Series of 2,4-Diamino-6-[(*N*-alkylanilino)methyl]quinazolines [1,2]

Leslie M. Werbel*, Edward F. Elslager, and Linda S. Newton

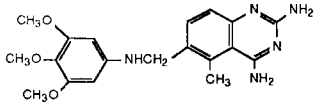
Chemistry Department, Warner-Lambert/Parke-Davis Pharmaceutical Research, Ann Arbor, Michigan, 48105

Received August 18, 1986

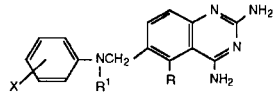
A series of 2,4-diamino-6-[(*N*-alkylanilino)methyl]quinazolines were prepared by bromination of 2,4-dibenzamido-6-methylquinazoline followed by treatment with secondary arylamines and deblocking with base. A variety of analogs demonstrated substantial activity against *Plasmodium berghei* infections in mice.

J. Heterocyclic Chem., **24**, 345 (1987).

The potent antimalarial and antitumor activity of nonclassical quinazoline dihydrofolate reductase inhibitors such as trimetrexate® (**1**) [3] made it of interest to examine the properties of some N6 substituted analogs. The synthesis and biological activity of a series of such analogs is presented herein.



1

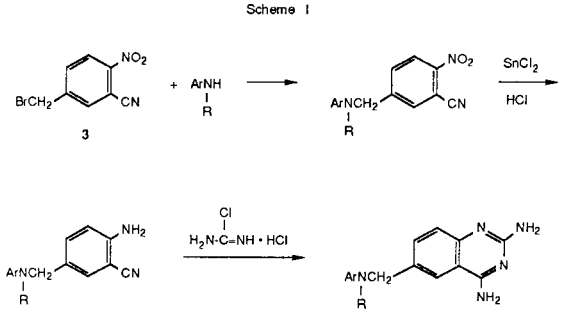


2

Chemistry.

The synthetic route to **1** and its analogs utilized reductive amination of a preformed 6-cyanoquinazoline and an aniline and was clearly not applicable to the desired *N*-substituted derivatives. Our initial plan therefore was to utilize Scheme I. This was frustrated however since all attempts to brominate the requisite nitrotoluene to provide

Scheme I

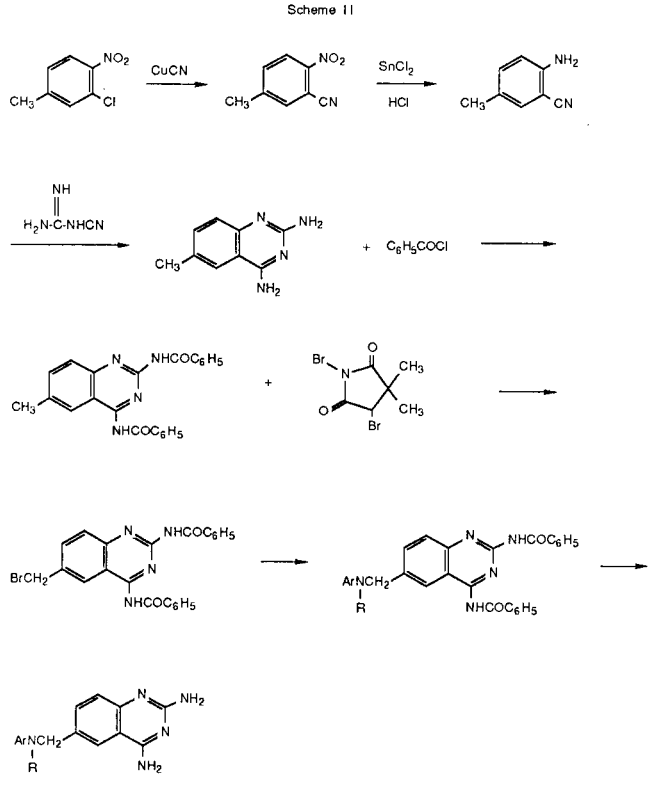


3

the necessary bromomethyl derivative **3** were unsuccessful. Direct alkylation of a corresponding NH analog such as the 3,4-dichloroanilino analog of **1** under a variety of conditions was also unsuccessful with spectral data indicating that alkylation was favored on the quinazoline ring rather than on the aniline NH.

We discovered that if the quinazoline amino groups were first protected, bromination of the 6-methyl group could be affected with 1,3-dibromo-5,5-dimethylhydantoin. Treatment with secondary arylamines then provided the desired protected products which could be deprotected with brief treatment with sodium methoxide in methanol. In this manner, as depicted in Scheme II, analogs **2a-r** were prepared (Table I).

Scheme II



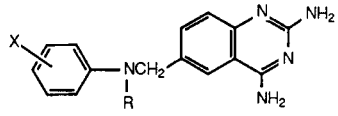
Suppressive Antimalarial Screening in Mice.

The 2,4-diamino-6-[(*N*-alkylanilino)methyl]quinazolines **2a-r** were tested against a normal drug-sensitive strain of *P. berghei* in mice by the parenteral route [5,6].

The compounds were dissolved or suspended in sesame or peanut oil and were administered to mice in a single subcutaneous dose 72 hours postinfection. The antimalarial activities are summarized in Table II. Clearly the presence of a small alkyl group on the 6-nitrogen allows retention of curative activity (compare for example **2a** with the parent **4** as well as the reverse isomer **3**). On the other

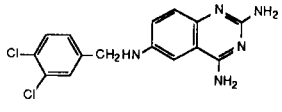
Table I

Properties of 2,4-Diamino-6-[(*N*-alkylanilino)methyl]quinazolines

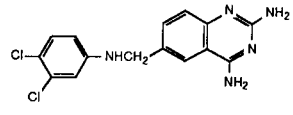


Compound No.	X	R	Method	mp °C	Yield %	Purification Solvent	Formula
2a	3,4-Cl_2	CH_3	A	236-239	19	DMF-NH_4OH	$C_{16}H_{15}Cl_2N_5 \cdot C_3H_7NO$
b	4-Br	CH_3	A	235-236	39	DMF-NH_4OH	$C_{16}H_{16}BrN_5 \cdot 0.7C_3H_7NO$
c	4-Cl	C_2H_5	A	125-127	68	HOAc-NH_4OH	$C_{17}H_{18}ClN_5 \cdot 1.0C_2H_4O_2 \cdot 1.3H_2O$
d	4-Br	C_2H_5	B	200(dec)	65	EtOH-Et_2O	$C_{17}H_{18}BrN_5 \cdot 1.8HCl \cdot 1.2H_2O$
e	3,4-Cl_2	C_3H_7	B	238-240	8.5	EtOH-Et_2O	$C_{18}H_{19}Cl_2N_5 \cdot 1.66HCl$
f	3,4-Cl_2	$CH(CH_3)_2$	B	248-250	62	1) EtOH-Et_2O 2) H_2O-NH_4OH	$C_{18}H_{19}Cl_2N_5 \cdot 0.1H_2O$
g	4-F	CH_3	C	200-202	26	CH_3CN	$C_{16}H_{16}FN_5$
h	3-CF_3	CH_3	B	190-192	19	20% HOAc	$C_{17}H_{16}F_3N_5 \cdot 1.1C_2H_4O_2 \cdot 0.6H_2O$
i	3-Cl	CH_3	B	222-225	29	MeOH-H_2O	$C_{16}H_{16}ClN_5$
j	4-OCH_3	C_2H_5	B	209-211	26	CH_3CN	$C_{18}H_{21}N_5O$
k	4-CH_3	C_2H_5	B	180-183	21	CH_3CN	$C_{18}H_{21}N_5 \cdot 0.25H_2O$
l	4-Cl	CH_3	B	180-190	10	20% HOAc	$C_{16}H_{16}ClN_5 \cdot 1H_2O \cdot 1.33C_2H_4O_2$
m	2-Cl,4-CH_3	CH_3	B	178-179	18	CH_3CN	$C_{17}H_{18}ClN_3 \cdot 0.9H_2O$
n	2,5-Cl_2	CH_3	B	105-109	16	20% HOAc	$C_{16}H_{15}Cl_2N_5 \cdot 1.1C_2H_4O_2 \cdot 0.8H_2O$
o	3,4-Cl_2	C_2H_5	B	232-234	14	CH_3CN	$C_{17}H_{17}Cl_2N_5$
p	3-CF_3	C_2H_5	B	168-171	17	toluene	$C_{18}H_{18}F_3N_5$
q	4-Cl	$CH(CH_3)_2$	B	216-218	23	MeOH	$C_{18}H_{20}ClN_5 \cdot 0.15H_2O$
r	3,5-$(CF_3)_2$	CH_3	B	232-233	12	MeOH	$C_{18}H_{15}F_6N_5$

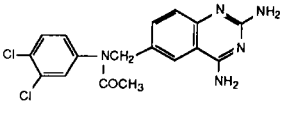
hand a small acyl group *i.e.* **5** reduces activity. In addition the data suggest that a moderately larger alkyl group substantively increases potency — for example **2f, 2j, 2o.**



3



4



5

Conclusion.

This work was not carried further when the potent antimalarial activity of the 6-arylsulfonyl analogs [7] was discovered, however, further pursuit of structures related to the 2,4-diamino-6-[(*N*-alkylanilino)methyl]quinazolines may be justified.

EXPERIMENTAL

Melting points (corrected) were taken on a Thomas-Hoover capillary melting point apparatus. Spectral data (ir and nmr) were obtained on all compounds to verify structure and the presence of solvate.

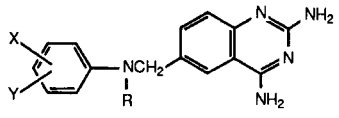
N,N'-[6-(Bromomethyl)-2,4-quinazolinediyl]bisbenzamide.

5-Methyl-2-nitrobenzonitrile.

A mixture of 125.0 g (0.729 mole) of 1-chloro-5-methyl-2-nitrobenzene, 130.7 g (1.46 mole) of cuprous cyanide, and 484 ml of 1-methyl-2-pyrrolidinone was heated at 160° for 5 hours. The hot reaction mixture was poured into 2 ℓ of pre-cooled chloroform. The solid that formed was collected by filtration and digested in 1 ℓ of chloroform. This suspension was filtered and the filtrate was combined with the first chloroform filtrate, treated with charcoal, and filtered through Celite. The solvent was removed *in vacuo* and the residue was poured into 2 ℓ of water and stirred

Table II

Parenteral Effects of 2,4-Diamino-6-[(*N*-alkylanilino)methyl]quinazolines Against Trophozoite-Induced *P. Berghei* in Mice



			ΔMST, T or C [a] after Mg/Kg					
No.	X,Y	R	640	320	160	80	40	20
2a	3,4-Cl_2	CH_3	5C	5C	5C	2C	10.6	5.3
b	4-Br	CH_3	3C	3C	3C	2C	1C	3.9
c	4-Cl	C_2H_5	3C	3C	3C	12.1	5.8	2.1
d	4-Br	C_2H_5	4C	4C	2C	11.1	7.9	5.9
e	3,4-Cl_2	$(CH_2)_2CH_3$	3C	2C	14.7	12.1	10.9	5.1
f	3,4-Cl_2	$CH(CH_3)_2$	5C	5C	3C	3C	2C	4.9
g	4-F	CH_3	5C	5C	5C	4C	8.0	5.3
h	3-CF_3	CH_3		10.9	8.5	7.5	1.7	0.7
i	3-Cl	CH_3	5C	5C	2C	1C	1C	5.1
j	4-OCH_3	C_2H_5			5C	4C	2C	4.7
k	4-CH_3	C_2H_5	5C	5C	4C	2C	4.5	4.0
l	4-Cl	CH_3		5C	4C	3C	2C	4.8
m	2-Cl,4-CH_3	CH_3	5C	4C	3C	2C	5.2	3.0
n	2,5-Cl_2	CH_3	12.7	3.1	2.7	0.9	0.7	—
o	3,4-Cl_2	Et	5C	5C	5C	3C	2C	—
p	3-CF_3	C_2H_5	—	4C	2C	7.7	3.7	—
q	4-Cl	$CH(CH_3)_2$	5C	5C	4C	1C	6.5	—
r	3,5-$(CF_3)_2$	CH_3	5C	5C	5C	12.1	9.1	—
3	—	—	5C	5C	3C	12.9	7.1	2.5
4	—	—	5C	5C	3C	1C	11.4	5.8
5	—	—	5T	1C, 4T	11.7	8.3	3.7	0.9

[a] ΔMST is the mean survival time (days) of treated mice (MSTT) minus the mean survival time (days) of control mice (MSTC). In the present study, the MSTC ranged from 6.1 to 6.3 days. T signifies the number of toxic deaths, occurring on day 2-5 after injection, that are attributed to drug action. Compounds are arbitrarily considered to be "active" when they produce at least a 100% increase in the mean survival time of treated mice. C indicates the number of mice surviving at 60 days post-infection and termed "cured"; data to establish parasitological cure based on subinoculation are unavailable. Each entry at each dose level represents results with a five-animal group.

Table III

Analytical Data

Compound No.	Carbon Calcd. (Found)	Hydrogen Calcd. (Found)	Nitrogen Calcd. (Found)	Other Calcd. (Found)
2a	54.16 (54.16)	5.26 (5.01)	19.94 (20.02)	
b	53.10 (53.35)	5.15 (5.29)	19.50 (19.82)	
c	55.49 (55.11)	6.02 (6.00)	17.03 (16.93)	H_2O 5.69 (5.38)
d	44.44 (44.63)	4.86 (4.52)	15.24 (14.99)	Cl 13.88 (14.30) H_2O 4.70 (4.51)
e	49.50 (49.29)	4.76 (4.74)	16.03 (16.10)	Cl 29.70 (29.57)
f	57.20 (57.47)	5.12 (5.06)	18.53 (18.23)	H_2O 0.48 (0.63)
g	64.63 (64.56)	5.42 (5.57)	23.55 (23.79)	
h	54.36 (54.31)	5.12 (5.45)	16.51 (16.43)	H_2O 2.54 (2.64)
i	61.24 (61.37)	5.14 (5.36)	22.32 (22.03)	
j	66.85 (66.76)	6.55 (6.60)	21.65 (21.65)	
k	69.32 (69.05)	6.94 (6.92)	22.46 (22.79)	H_2O 1.44 (1.14)
l	54.44 (54.18)	5.71 (5.71)	17.01 (16.70)	H_2O 4.37 (4.51)
m	59.35 (59.24)	5.80 (5.60)	20.35 (20.27)	H_2O 4.71 (4.78)
n	50.99 (50.62)	4.93 (5.02)	16.34 (16.48)	H_2O 3.36 (3.58)
o	56.36 (56.19)	4.73 (5.01)	19.33 (19.28)	
p	59.82 (59.60)	5.02 (5.00)	19.38 (19.44)	
q	62.75 (62.61)	5.93 (6.15)	20.33 (20.39)	H_2O 0.78 (0.30)
r	52.05 (51.86)	3.64 (3.95)	16.86 (17.02)	

well overnight. The liquid was decanted and the oily solid was triturated twice more with water. After the third trituration, the water was decanted and the resulting solid was dissolved in chloroform. The water layer was separated and the chloroform layer was dried over magnesium sulfate. The solvent was removed *in vacuo* leaving a liquid residue that solidified after cooling to room temperature. Recrystallization from 95% ethanol yielded 44.5 g (38%) of the desired product, mp 83-83.5°.

A crude sample was recrystallized two times from carbon tetrachloride to give material with melting point of 88-89° (lit [4] mp 92°).

Anal. Calcd. for $C_8H_6N_2O_2$: C, 59.26; H, 3.73; N, 17.27. Found: C, 59.41; H, 3.71; N, 17.30.

2-Amino-5-methylbenzonitrile.

To a cooled solution of 120 g (0.52 mole) of stannous chloride dihydrate and 300 ml of concentrated hydrochloric acid was added 24.3 g (0.15 mole) of 5-methyl-2-nitrobenzonitrile. An ice bath was applied intermittently to maintain the temperature below 40°. After 15 minutes, when the dilution of a sample from the reaction mixture with water gave no precipitate, the reaction flask was cooled in an ice-salt bath for 3 hours. The solid which precipitated was filtered, dried briefly on the filter and added to 600 ml of water. Addition of 50% sodium hydroxide until the mixture was weakly basic resulted in the formation of a white solid which was collected and dried *in vacuo* to give 10.1 g (51%) of the product which was shown to be homogeneous by thin layer chromatography.

A sample was dissolved in dilute hydrochloric acid and reprecipitated with 50% sodium hydroxide to give material with mp 60°.

Anal. Calcd. for $C_8H_8N_2$: C, 72.71; H, 6.10; N, 21.19. Found: C, 72.42; H, 6.12; N, 21.02.

6-Methyl-2,4-quinazolinediamine.

2-Amino-5-methylbenzonitrile (12.4 g, 0.093 mole) and 7.8 g (0.093 mole) of cyanoguanidine were heated under reflux in 93 ml of 1*N* hydrochloric acid for 1.5 hours. The reaction mixture was diluted to 217 ml with water and 93 ml of 1*N* hydrochloric acid was added. The hot solution was filtered and made strongly basic with 165 ml of 2*N* sodium hydroxide. The yellow solid that formed was collected by filtration and air dried. The solid was then suspended in 150 ml of water and 25 ml of 88% formic acid was added with stirring. The resulting white solid was collected by filtration, dissolved in 970 ml of water, and made strongly basic with ammonium hydroxide. The beige solid that formed was collected by filtration and dried to give 9.3 g (58%) of the desired product, mp 258-259.5°.

Anal. Calcd. for $C_9H_{10}N_4$: C, 62.05; H, 5.78; N, 32.16. Found: C, 61.77; H, 5.80; N, 32.55.

N,N'-(6-Methyl-2,4-quinazolinediyl)bisbenzamide.

To a refluxing mixture of 20.7 g (0.12 mole) of 6-methyl-2,4-quinazolinediamine, 83 ml (0.59 mole) of triethylamine, and 600 ml of 1,4-dioxane, was added a solution of 33 ml (0.30 mole) of benzoyl chloride in 30 ml of 1,4-dioxane. The reaction was heated under reflux for 20 minutes, filtered hot and the solid triethylamine hydrochloride was washed with 50 ml of hot 1,4-dioxane. The filtrates were allowed to stand for 2 days, and the solid product was filtered to give 32.5 g, mp 175-180°. A second crop of 10.9 g, mp 160-185°, was obtained by removing the dioxane *in vacuo*, and crystallizing the residue from anhydrous ethanol. Both crops were combined and recrystallized from anhydrous ethanol to give 33.4 g (72%) of the desired product, mp 187-189°.

N,N'-[6-(Bromomethyl)-2,4-quinazolinediyl]bisbenzamide.

A refluxing mixture of 19.1 g (0.05 mole) of *N,N'*-(6-methyl-2,4-quinazolinediyl)bisbenzamide, 7.6 g (0.026 mole) of 1,3-dibromo-5,5-dimethyl-2,4-imidazolidinedione and 1.4 g of benzoylperoxide in 1 ℓ of carbon tetrachloride was irradiated with a high intensity lamp (600W-120V). After 10 minutes an almost clear reddish-brown solution was obtained; after 10 minutes more the color faded, a yellow solid appeared, and the mixture was negative to starch-iodide test paper. The reaction was filtered hot, and the solid was washed first with ether, then with 50 ml of water, and finally with ether again. The solid was dried *in vacuo* and then ground with a mortar and pestle first with ether and then with a water-ether mixture. The solid was collected and dried *in vacuo* to give 11.3 g of product, mp 198-215°. A second crop was obtained by cooling the carbon tetrachloride reaction filtrate and treating in the same manner to give 4.9 g of product, mp 185-198°. The total yield of both crops was 16.2 g (70%) and the nmr spectrum (in trifluoroacetic acid) showed both crops to be quite pure except for traces of unchanged starting material and 5,5-dimethyl-2,4-imidazolidinedione, a by product of the reaction, and the material was used as is.

2,4-Diamino-6-[(*N*-alkylanilino)methyl]quinazolines. Method A.

6-[[(4-Bromophenyl)methylamino]methyl]-2,4-quinazolinediamine, 0.7 f. wt. of *N,N*-Dimethylformamide of Crystallization.

A mixture of 4.61 g (0.01 mole) of *N,N'*-[6-(bromomethyl)-2,4-quinazolinediyl]bisbenzamide, 1.86 g (0.01 mole) of 4-bromobenzenamine, and 1.30 g (0.01 mole) of potassium carbonate in 150 ml of *N,N*-dimethylformamide was heated at 75° with stirring for 4 hours. The reaction was cooled, diluted with chloroform and extracted several times with water. The chloroform was dried over magnesium sulfate and the solvent was removed *in vacuo*. The resulting oil was triturated with water to give a semi-solid; the water was decanted, and the semi-solid digested with ethanol to give 3.9 g of crude *N,N'*-[6-[[(4-bromophenyl)methylamino]methyl]-2,4-quinazolinediyl]bisbenzamide. The crude dibenzamide, 3.9 g, (7.0 mmoles) and 0.83 g (14.5 mmoles) of sodium methoxide in 200 ml of methanol was heated under reflux for 1.5 hours. The solvent was removed *in vacuo* and the resulting oil was digested with 15 ml of 1*N* sodium methoxide and 30 ml of water. The resulting yellow solid was collected by filtration, washed with water and dried *in vacuo* to give 2.6 g of crude product, mp 227-235°. This material was recrystallized from 20% acetic acid to give 2.2 g of the product as its acetate salt. The free base was generated by dissolving the solid in *N,N*-dimethylformamide and pouring the solution into concentrated ammonium hydroxide. The solid was collected by filtration and dried at 100° *in vacuo* for 5 hours to give 1.4 g (39%) of the product containing 0.7 mole of *N,N*-dimethylformamide of crystallization, mp 235-236°.

The nmr spectrum confirms the presence of *N,N*-dimethylformamide.

Method B.

6-[[(4-Chlorophenyl)(1-methylethyl)amino]methyl]-2,4-quinazolinediamine, 0.15 Hydrate.

To a mixture of 1.69 g (0.01 mole) of 4-chloro-*N*-(1-methylethyl)benzenamine, 1.38 g (0.01 mole) of potassium carbonate, and 20 g of dimethylsulfone at 145° was added 4.61 g (0.01 mole) of crude *N,N'*-[6-(bromomethyl)-2,4-quinazolinediyl]bisbenzamide. The reaction was heated at 140-145° for 45 minutes and then poured into 500 ml of chloroform. A dark solid was removed by filtration and the solution was washed three times with warm water, dried over magnesium sulfate, and the solvent was removed *in vacuo*. To the residue was added 20 ml of benzene-ethyl acetate (4:1) and a white solid (dimethylsulfone) was removed by filtration. Upon standing a second solid precipitated and was collected to give 1.88 g of crude *N,N'*-[6-[[(4-chlorophenyl)(1-methylethyl)amino]methyl]-2,4-quinazolinediyl]bisbenzamide. The filtrate was eluted through a silica gel column with benzene-ethyl acetate (4:1) to give another 0.54 g of the bisbenzamide. The 1.88 g crop was digested with methanol to give 1.55 g of material which was combined with the 0.54 g crop. This 2.09 g of the bisbenzamide was heated under reflux in 150 ml of methanol containing 0.45 g of sodium methoxide for 1.25 hours. The solvent was evaporated to give an oily residue which formed a yellow solid upon trituration with water. This solid was collected, dried *in vacuo*, and stirred with 7 ml of methanol to give a cream colored solid. This solid was collected and dried under high vacuum at 140° to give 0.82 g (23%) of the title compound, mp 216-218°.

Method C.

6-[[(4-Fluorophenyl)methylamino]methyl]-2,4-quinazolinediamine (**2**).

To a slurry of 0.42 g (0.01 mole) of sodium hydride (57% mineral oil dispersion) in 50 ml of toluene was added 1.25 g (0.01 mole) of 4-fluoro-*N*-methylbenzenamine. The mixture was stirred for 15 minutes at room temperature, 4.61 g (0.01 mole) of *N,N'*-[6-(bromomethyl)-2,4-quinazolinediyl]bisbenzamide was added and the mixture was heated at 75° for 1 hour. A second slurry of 0.42 g (0.01 mole) of sodium hydride, 50 ml of toluene, and 1.25 g (0.01 mole) of 4-fluoro-*N*-methylbenzenamine was prepared and added to the reaction mixture, and the mixture was heated again at 75° for 1 hour. The reaction mixture was filtered, the toluene removed *in vacuo* and 100 ml of chloroform added to the residue. This solution was filtered, the solvent reduced *in vacuo* to 15 ml, and applied to a silica gel column and eluted with chloroform. Those fractions which contained only a single spot by thin layer chromatography (silica-ethyl acetate, R_f 0.17) were combined and the solvent was removed *in vacuo* to give 0.85 g of pure *N,N'*-[6-[[(4-fluorophenyl)methylamino]methyl]-2,4-quinazolinediyl]bisbenzamide. Those fractions which contained an impurity (Si-ethyl acetate, R_f 0.44) were combined to give another 1.72 g of impure intermediate. The impure material and 0.38 g (0.007 mole) of sodium methoxide in 100 ml of methanol was heated under reflux for 45 minutes. The reaction was cooled, the solvent was removed *in vacuo*, 100 ml of water was added, and the resulting solid was collected by filtration and dried *in vacuo* to give 0.67 g, mp 176-190°. The 0.85 g of pure intermediate was treated similarly to provide an additional 0.26 g of material, mp 155-175°. Both crops were very similar by thin layer chromatography and were combined and recrystallized from acetonitrile to give 0.75 g (26%) of the product, mp 200-202°.

Acknowledgements.

The authors thank Mr. C. E. Childs and associates for microanalytical

data, and Dr. J. M. Vandenbelt and coworkers for determination of the spectral data.

REFERENCES AND NOTES

[1] This is paper **62** of a series on antimalarial agents. For paper **61** see S. J. Kesten, J. Johnson and L. M. Werbel, *J. Med. Chem.*, in press.

[2] This investigation was supported by U.S. Army Medical Research and Development Command Contract DADA17-72-C-2077. This is contribution 1795 to the U.S. Army Drug Development Program.

[3] E. F. Elslager, J. L. Johnson, and L. M. Werbel, *J. Med. Chem.*, **26**, 1763 (1983).

[4] E. Macovski and J. Georgescu, *Bull. Sect. Sci. Acad. Roumaine*, **28**, 354 (1946); *Chem. Abstr.*, **43**, 3805e (1949).

[5] T. S. Osdene, P. B. Russell, and L. Rane, *J. Med. Chem.*, **10**, 431 (1967).

[6] The parenteral antimalarial screening in mice was carried out by Dr. Leo Rane of the University of Miami, and test results were provided through the courtesy of Drs. T. R. Sweeney, E. A. Steck and Mr. M. Musallam of the Walter Reed Army Institute of Research.

[7] E. F. Elslager, M. P. Hutt, P. Jacob, J. Johnson, B. Temporelli, L. M. Werbel, and D. F. Worth, *J. Med. Chem.*, **22**, 1247 (1979).