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Bioorganic & Medicinal Chemistry 12 (2004) 1003–1010
Bioorganic & Medicinal Chemistry

Synthesis and blood-schizontocidal antimalarial activities of 2-substituted/2,5-disubstituted-8-quinolinamines and some of their amino acid conjugates☆

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Received 4 November 2003; accepted 11 December 2003

Abstract—Thirteen new analogues (**32**–**40**, **45**–**48**) of recently discovered potent blood-schizontocidal antimalarial agent, 2-*tert*-butylprimaquine (**2**) are synthesized and evaluated for in vivo antimalarial activities against drug-sensitive *P. berghei* strain and multi-drug resistant *P. yoelii nigeriensis* strain. Two of the amino acid conjugates (**47**–**48**) have exhibited potent antimalarial activities similar to that of **2** against both drug-sensitive and multi-drug resistant strains.

1. Introduction

Malaria, the ancient mosquito-borne disease that was rolled back by medical advances in the mid-20th century, is making a deadly comeback, with 600 million estimated clinical cases and approximately 3 million deaths per year.[1] Nearly all fatalities are caused by *Plasmodium falciparum*; one of the four species responsible for causing human malaria. The parasite's resistance to conventional antimalarial drugs such as chloroquine, the cheapest and most effective anti-malarial since the 1950s is growing at an alarming rate. Therefore, a re-look at the promising existing drugs that are effective against resistant strain and new efficient drugs are urgently needed.

2. Results and discussion

Primaquine (**1**) is known to be active against more of the life cycle stages of plasmodia than any other class of antimalarial drugs, but has minimal suppressive activity; that is, is ineffective as blood-schizontocide, and can't be used to cure infections caused by *P. falciparum*.[2] Usefulness of **1** is also restricted by toxic side effects including hemolytic lesions (caused by methemoglobin production), pronounced in the patients deficient in glucose-6-phosphate dehydrogenase. Our research efforts are directed towards improving blood-schizontocidal antimalarial activity of primaquine, and at the same time reducing the toxic manifestations that are traditionally associated with it. In this direction, we have recently reported that the placement of a bulky metabolically stable *tert*-butyl group at the C-2 position of quinoline ring in **1** results in tremendous improvement in the blood-schizontocidal antimalarial activity.[3] Antimalarial compound, 2-*tert*-butylprimaquine (**2**) (Fig. 1) exhibits potent in vivo antimalarial activities against both drug-sensitive strain (*P. berghei*), and multi-drug resistant strain (*P. yoelii nigeriensis*), and thus is a powerful blood-schizontocide. Furthermore, analogue **2** is discovered to be the first 8-quinolinamine completely devoid of methemoglobin (MetHb) toxicity associated with **1**. We had proposed that the extremely encouraging and remarkable antimalarial activities of the analogue **2** are emanated by blocking of a putative unwarranted metabolic degradation pathway of the primaquine at the C-2 position of the quinoline ring.[3] The most logical extension of this spectacular finding is to further explore the optimum size requirement of alkyl group at the C-2 position and other substituents in primaquine. Herein, we describe synthesis and antimalarial activities of nine additional 2-substituted and 2,5-di-

Keywords: Malaria; 8-Quinolinamines; 2-*tert*-Butylprimaquine; Blood-schizontocides.
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☆NIPER communication no. 245.

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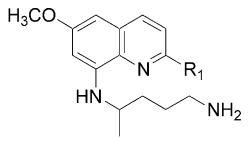
doi:10.1016/j.bmc.2003.12.029



1. R_1= H [primaquine]
2. R_1= $C(CH_3)_3$ [2-*tert*-butylprimaquine]

Figure 1.

substituted primaquine analogues (**32–40**) bearing other metabolically stable groups.

It is known that approximately 35–83% of primaquine gets metabolized to 4-(6-methoxy-quinolin-8-ylamino)-pentanoic acid in a primate model.[4] We have recently proposed that attachment of an amino acid residue may serve to protect the primary amino function of **1** against the aforementioned metabolic process.[5] This study demonstrates that a careful selection of amino acids and their attachment at the primary amino group led to a substantially increase in the antimalarial activities of 8-quinolinamines.[5] Keeping these observations in mind, the present work also reports synthesis and antimalarial activity evaluation of four amino acid conjugates (**45–48**) of 2-*tert*-butylprimaquine (**2**).

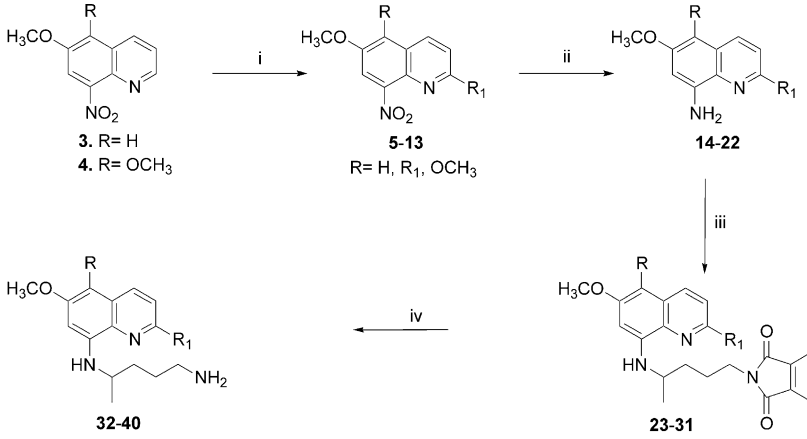
3. Chemistry

Commercially available 6-methoxy-8-nitroquinoline (**3**) and 5,6-dimethoxy-8-nitroquinoline (**4**) upon direct ring-alkylation via a silver catalyzed radical oxidative decarboxylation of appropriate alkylcarboxylic acid by ammonium persulfate in CH_3CN and 10%H_2SO_4 efficiently produced a mixture of mono and dialkylated 8-nitroquinolines (**5–13**), which were conveniently separated by column chromatography. The latter compounds (**5–13**) were converted to the requisite 2-substituted/2,5-disubstituted N^8-(4-amino-1-methylbutyl)-6-methoxy-8-quinolinamines (**32–40**) in three steps following previously published procedure (Scheme 1).[6]

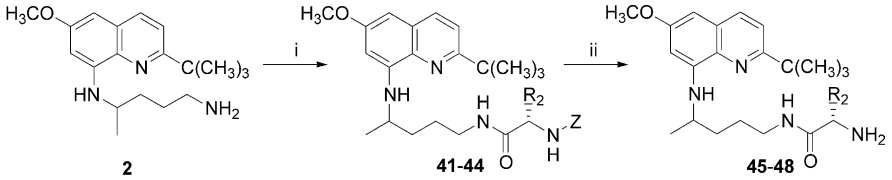
On the other hand, 2-*tert*-butylprimaquine (**2**) synthesized using procedure reported earlier,[3] upon condensation reaction with N-carbobenzyloxy (Z) protected L-amino acids in the presence of 1,3-dicyclohexylcarbodiimide (DCC) in CH_2Cl_2 for 4 h gave the required N^8-(4-amino-1-methylbutyl)-2-*tert*-butyl-6-methoxy-8-quinolinamine-Z-amino acid conjugates (**41–44**) in excellent yields. Hydrogenolysis of the latter compounds (**41–44**) at ambient temperature using 10% Pd-C catalyst in the presence of H_2 gas proceeded smoothly to produce the free amino acid conjugates that upon treatment with ethereal hydrogen chloride solution provided the N^8-(4-amino-1-methylbutyl)-2-*tert*-butyl-6-methoxy-8-quinolinamine-amino acid conjugates (**45–48**) as their hydrochloride salts (Scheme 2).

4. Biological activity

The target compounds were evaluated for in vivo blood-schizontocidal antimalarial activity against *P. berghei* (sensitive strain) in a rodent model (Table 1). The details of the biological procedure are reported elsewhere.[7] Briefly; testing of synthesized analogues was conducted at various concentrations, orally, in mice (6 mice per group). The concentrations tested were 100, 50, 25 and 10 mg/kg/day×4 (oral). The compounds were administered on days 0–3 post infection. The results for the synthesized analogues were compared to a positive



Scheme 1. Reagents and conditions: (i) R_1CO_2H, $AgNO_3$, $(NH_4)_2S_2O_8$, 10% H_2SO_4, CH_3CN, 70 °C; (ii) Raney Ni, EtOH, H_2, 45 psi, 45 min; (iii) 2-(4-bromopentyl)-1,3-isoindolinedione, Et_3N, 120 °C, 24 h; (iv) $NH_2NH_2{\cdot}H_2O$, EtOH, reflux, 8 h.



Scheme 2. Reagents and conditions: (i) Z-L-R_2-OH, DCC, CH_2Cl_2, rt; (ii) H_2/Pd-C, 1 h, rt.

Table 1. In vivo blood-schizontocidal antimalarial activity of the N^8-(4-amino-1-methylbutyl)-2-substituted/2,5-disubstituted-6-methoxy-8-quinolinamines (**32–40**) and 2-*tert*-butylprimaquine conjugates (**45–48**) against *P. berghei* infection in mice (6 mice per group)

S. No.	R	R_1	R_2	Dose (mg/kg/day×4, oral)			
				10	25	50	100
32	H	$CH(CH_3)_2$	—	—	—	(3/6) Active	(6/6) Curative
33	$CH(CH_3)_2$	$CH(CH_3)_2$	—	—	—	—	(0/6) Inactive
34	H	*c*-C_5H_9	—	—	—	—	(0/6) Inactive
35	*c*-C_5H_9	*c*-C_5H_9	—	—	—	—	(0/6) Inactive
36	H	*c*-C_6H_{11}	—	—	—	—	(0/6) Inactive
37	*c*-C_6H_{11}	*c*-C_6H_{11}	—	—	—	—	(0/6) Inactive
38	OCH_3	$CH(CH_3)_2$	—	—	—	—	(2/6) Active
39	OCH_3	$C(CH_3)_3$	—	—	—	—	(3/6) Active
40	OCH_3	*c*-C_6H_{11}	—	—	—	—	(0/6) Inactive
45	—	—	CH_3	—	—	(4/6) Active	(6/6) Curative
46	—	—	$CH(CH_3)_2$	—	—	—	(0/6) Inactive
47	—	—	$(CH_2)_3NH_2$	(2/6) Active	(6/6) Curative	(6/6) Curative	(6/6) Curative
48	—	—	$(CH_2)_4NH_2$	(4/6) Active	(6/6) Curative	(6/6) Curative	(6/6) Curative
2-*tert*-Butylprimaquine (2)				(4/6) Active	(6/6) Curative	(6/6) Curative	(6/6) Curative
Primaquine				—	—	—	(0/6) Inactive

The term 'curative' indicates complete elimination of malaria parasites, and animals survive up to day D + 60. The term 'active' indicates that treated animals show negative parasitaemia up to D + 7. However, by D + 60, some mice die, and some survive with complete elimination of parasitaemia as indicated by numbers given in parentheses. The term 'inactive' indicates that the treated animals show positive parasitaemia either on D + 4 or D + 7 and usually die by D + 14.

control group of mice treated with chloroquine at a suppressive dose of 10 mg/kg/day×4 (oral). The results were also compared to a negative control group of mice where no treatment for the infection was administered, and in this case 100% mortality is observed within 6–8 days, with a mean survival time of 6.2 days.

All of the 2-substituted/2,5-disubstituted N^8-(4-amino-1-methylbutyl)-6-methoxy-8-quinolinamines (**32–40**) were found to be less effective compared to analogue **2** in the *P. berghei* test. The most effective compound **32** [R = H, $R_1 = CH(CH_3)_2$] produced 100% cures at the preliminary tested dose of 100 mg/kg, but exhibited only partial cures at 50 mg/kg. The observation of insignificant activity in the synthesized derivatives suggests that the *tert*-butyl group placed at C-2 position of the quinoline ring in primaquine is optimal for superior antimalarial activity. Likewise, placement of an additional alkyl substitution at C-5 position of the quinoline ring (2,5-dialkylated analogues **33**, **35** and **37**) led to diminished antimalarial activity. Finally, placement of 5-methoxy group in the quinoline ring (analogues **38–40**) resulted in compounds with complete loss of antimalarial activity. To our surprise, placement of an additional methoxy group at the C-5 position (analogue **38**) in 2-*tert*-butylprimaquine (**2**) also resulted in drastic reduction in antimalarial activity. The complete loss of activity in analogue **2** by virtue of an additional methoxy group in the ring clearly indicates that strategic positioning of 6-methoxy and 2-*tert*-butyl groups is required for superior antimalarial activities.

As observed by us earlier in the cases involving various 5-alkoxyprimaquine analogues,[5] valine adduct (**46**) of analogue **2** also did not show any antimalarial activity; whereas, alanine conjugate (**45**) exhibited 100% cures at 100 mg/kg, but was only partially curative at the next tested dose of 50 mg/kg. In contrast, ornithine adduct (**47**) and lysine adduct (**48**) exhibited potent antimalarial activities, and have produced partial cures at the lowest tested dosage of 10 mg/kg. Although analogue **48** was equi-potent to 2-*tert*-butylprimaquine (**2**), none of the synthesized amino acid conjugates were found superior to it.

Compounds exhibiting activity at 10 mg/kg/day were further selected for evaluation of antimalarial efficacy in *P. yoelii nigeriensis* (multi-drug resistant strain) using protocol similar to that for *P. berghei* test, and results are summarized in Table 2. Along the same lines with the results obtained for *P. berghei* test model, analogues **47** and **48** produced partial cures at the preliminary tested dose of 100 mg/kg. Analogue **48** showed antimalarial activity similar to that of 2-*tert*-butylprimaquine (**2**) and cured 2/6 mice at a dose of 50 mg/kg.

Table 2. In vivo blood-schizontocidal antimalarial activity of the 2-*tert*-butylprimaquine conjugates (**47–48**) against multi-drug resistant *P. yoelii nigeriensis* infection in mice (6 mice per group)

S. No.	R_2	Dose (mg/kg/day×4, oral)	
		50	100
47	$(CH_2)_3NH_2$	(0/6) Inactive	(3/6) Active
48	$(CH_2)_4NH_2$	(2/6) Active	(4/6) Active
2-*tert*-Butylprimaquine (2)		(4/6) Active	(6/6) Curative
Primaquine		—	(0/6) Inactive

The term 'curative' indicates complete elimination of malaria parasites, and animals survive up to day D + 60. The term 'active' indicates that treated animals show negative parasitaemia up to D + 7. However, by D + 60, some mice die, and some survive with complete elimination of parasitaemia as indicated by numbers given in parentheses. The term 'inactive' indicates that the treated animals show positive parasitaemia either on D + 4 or D + 7 and usually die by D + 14.

5. Conclusions

In summary, to further explore the most suitable alkyl group at the C-2 position in primaquine, we have synthesized additional analogues of the recently discovered 2-*tert*-butylprimaquine (**2**). The results of this study clearly established that the potent antimalarial activity displayed by **2** is attributed to the incorporation of 2-*tert*-butyl group in primaquine, and its replacement with other metabolically stable groups like adamantyl, isopropyl, cyclohexyl and cyclopentyl results in reduced antimalarial activity. Furthermore, in agreement with our earlier observations with 5-alkoxyprimaquines, lysine and ornithine conjugated derivatives (**47**–**48**) exhibited promising antimalarial effects similar to that of **2** against drug-sensitive and multi-drug-resistant malaria strains. However, incorporation of these amino acids failed to enhance the bio-efficacy of analogue **2**. Our efforts are currently directed towards exploration of other cationic L- and D-amino acids seeking further enhancement in the antimalarial activities of **2**, and results of this study will be published shortly.

6. Experimental

Melting points were recorded on Mettler DSC 851 or capillary melting point apparatus and are uncorrected. ^{1}H spectra were recorded on 300 MHz Bruker FT-NMR (Avance DPX300) spectrometer using tetramethylsilane as internal standard and the chemical shifts are reported in δ units. Mass spectra were recorded on either GCMS (Shimadzu QP 5000 spectrometer) auto sampler/direct injection (EI/CI) or HRMS (Finnigan Mat LCQ spectrometer) (APCI/ESI). Elemental analyses were recorded on Elementar Vario EL spectrometer. All chromatographic purification was performed with silica gel 60 (230–400 mesh), whereas all TLC (silica gel) development was performed on silica gel coated (Merck Kiesel 60 F_{254}, 0.2 mm thickness) sheets. All chemicals were purchased from Aldrich Chemical Ltd (Milwaukee, WI, USA). Solvents used for the chemical synthesis acquired from commercial sources were of analytical grade, and were used without further purification unless otherwise stated.

6.1. General method for the synthesis of 2-substituted/2,5-disubstituted-8-nitroquinolines (5–13)

A solution of 6-methoxy-8-nitroquinoline (**3**, 1 mmol) or 5,6-dimethoxy-8-nitroquinoline (**4**, 1 mmol) in CH_3CN (5 mL) was heated to 70 °C. Silver nitrate (0.6 mmol), requisite alkylcarboxylic acid (2.5 mmol), and 10% H_2SO_4 (10 mL) was added to the reaction mixture. A freshly prepared solution of ammonium persulfate (3 mmol) in water (10 mL) was added drop wise during 10 min. The heating source was removed and reaction proceeded with the evolution of carbon dioxide. After 10 min, reaction mixture was poured onto ice, and made alkaline by adding 30% NH_4OH solution. Extracted with $CHCl_3$ (4×50 mL), and combined extracts were washed with NaCl solution (2×10 mL). Dried over Na_2SO_4, and solvent removed in vacuo to afford oil, which upon column chromatography over silica gel (230–400 mesh) afforded 2-substituted/2,5-disubstituted-8-nitroquinolines (**5**–**13**).

6.1.1. 2-Isopropyl-6-methoxy-8-nitroquinoline (5). Yield: 47%; mp 73–74 °C; IR (KBr) 1540, 1388 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 8.01 (d, 1H, J = 8.6 Hz), 7.60 (s, 1H), 7.40 (d, 1H, J = 8.6 Hz), 7.26 (s, 1H), 3.95 (s, 3H), 3.19 (m, 1H), 1.43 (m, 6H); EIMS m/z 246 (M^+); analysis for $C_{13}H_{14}N_2O_3$ (246.3), calcd, C, 63.40; H, 5.73; N, 11.38; found, C, 63.41; H, 5.79; N, 11.65.

6.1.2. 2,5-Diisopropyl-6-methoxy-8-nitroquinoline (6). Yield: 12%; oil; IR (KBr) 1545, 1395 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 8.09 (d, 1H, J = 8.4 Hz), 7.47 (d, 1H, J = 8.4 Hz), 7.19 (s, 1H), 3.99 (s, 3H), 3.22 (m, 2H), 1.52 (m, 6H), 1.48 (m, 6H); EIMS m/z 288 (M^+); analysis for $C_{16}H_{20}N_2O_3$ (288.3), calcd, C, 66.65; H, 6.99; N, 9.72; found, C, 66.45; H, 6.74; N, 9.78.

6.1.3. 2-Cyclopentyl-6-methoxy-8-nitroquinoline (7). Yield: 52%; mp 85–86 °C; IR (KBr) 1535, 1215 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 8.10 (d, 1H, J = 8.4 Hz), 7.63 (s, 1H), 7.45 (d, 1H, J = 8.4 Hz), 7.21 (s, 1H), 3.98 (s, 3H), 3.15 (m, 1H), 1.64 (m, 8H); EIMS m/z 272 (M^+); analysis for $C_{15}H_{16}N_2O_3$ (272.3), calcd, C, 66.16; H, 5.92; N, 10.29; found, C, 66.44; H, 5.73; N, 10.37.

6.1.4. 2,5-Dicyclopentyl-6-methoxy-8-nitroquinoline (8). Yield: 10%; oil; IR (KBr) 1545, 1388 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 8.09 (d, 1H, J = 8.1 Hz), 7.47 (d, 1H, J = 8.1 Hz), 7.17 (s, 1H), 3.98 (s, 3H), 2.82 (m, 2H), 1.74 (m, 16H); EIMS m/z 340 (M^+); analysis for $C_{20}H_{24}N_2O_3$ (340.4), calcd, C, 70.56; H, 7.11; N, 8.23; found, C, 70.44; H, 7.07; N, 8.23.

6.1.5. 2-Cyclohexyl-6-methoxy-8-nitroquinoline (9). Yield: 43%; mp 88–89 °C; IR (KBr) 1533, 1386 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 8.14 (d, 1H, J = 8.4 Hz), 7.67 (s, 1H), 7.41 (d, 1H, J = 8.4 Hz), 7.23 (s, 1H), 3.99 (s, 3H), 2.82 (m, 1H), 1.67 (m, 10H); EIMS m/z 286 (M^+); analysis for $C_{16}H_{18}N_2O_3$ (286.3), calcd, C, 67.12; H, 6.34; N, 9.78; found, C, 66.98; H, 6.52; N, 9.63.

6.1.6. 2,5-Dicyclohexyl-6-methoxy-8-nitroquinoline (10). Yield: 11%; oil; IR (KBr) 1537, 1382 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 8.09 (d, 1H, J = 8.2 Hz), 7.49 (d, 1H, J = 8.2 Hz), 7.12 (s, 1H), 3.94 (s, 3H), 2.87 (m, 2H), 1.77 (m, 20H); EIMS m/z 368 (M^+); analysis for $C_{22}H_{28}N_2O_3$ (368.5), calcd, C, 71.71; H, 7.66; N, 7.60; found, C, 72.04; H, 7.59; N, 7.79.

6.1.7. 2-Isopropyl-5,6-dimethoxy-8-nitroquinoline (11). Yield: 55%; mp 91–92 °C; IR (KBr) 1565, 1340 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 7.87 (d, 1H, J = 8.8 Hz), 7.65 (s, 1H), 6.86 (d, 1H, J = 8.8 Hz), 4.08 (s, 3H), 4.02 (s, 3H), 3.12 (m, 1H), 1.25 (m, 6H); EIMS m/z 276 (M^+); analysis for $C_{14}H_{16}N_2O_4$ (276.3), calcd, C, 60.86; H, 5.84; N, 10.14; found, C, 60.77; H, 5.69; N, 9.98.

6.1.8. 2-*tert*-Butyl-5,6-dimethoxy-8-nitroquinoline (12). Yield: 65%; mp 79–80 °C; IR (KBr) 1528, 1349 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 8.05 (d, 1H, J = 8.9 Hz), 7.89 (s, 1H), 7.60 (d, 1H, J = 8.9 Hz), 4.07 (s, 3H), 4.02 (s, 3H),

1.42 (s, 9H); ESIMS *m/z* 291 (M+1); analysis for $C_{15}H_{18}N_2O_4$ (290.3), calcd, C, 60.06; H, 6.25; N, 9.65; found, C, 60.25; H, 6.59; N, 9.79.

6.1.9. 2-Cyclohexyl-5,6-dimethoxy-8-nitroquinoline (13). Yield: 57%; mp 84–85 °C; IR (KBr) 1540, 1375 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.94 (d, 1H, *J* = 8.6 Hz), 7.75 (s, 1H), 6.93 (d, 1H, *J* = 8.6 Hz), 3.98 (s, 3H), 3.97 (s, 3H), 2.92 (m, 1H), 1.25 (s, 10H); EIMS *m/z* 316 (M^+); analysis for $C_{17}H_{20}N_2O_4$ (316.3), calcd, C, 64.54; H, 6.37; N, 8.86; found, C, 64.30; H, 6.39; N, 8.88.

6.2. General method for the synthesis of 2-substituted/2,5-disubstituted-6-methoxy-8-quinolinamines (14–22)

A solution of 2-substituted/2,5-disubstituted-6-methoxy-8-nitroquinoline (**5–13**, 5 mmol) in 95% ethyl alcohol (15 mL) was hydrogenated over wet raney nickel (T_1 grade) at 45 psi in a parr hydrogenator for 45 min. Catalyst was removed by filtration, and filtrate was evaporated under vacuum to afford 2-substituted/2,5-disubstituted-6-methoxy-8-quinolinamine (**14–22**) as dark colored oil.

6.2.1. 2-Isopropyl-6-methoxy-8-quinolinamine (14). Yield: 95%; oil; IR (KBr) 3330 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.84 (d, 1H, *J* = 8.6 Hz), 7.41 (d, 1H, *J* = 8.6 Hz), 6.54 (s, 1H), 6.43 (s, 1H), 5.13 (bs, 2H), 4.22 (s, 3H), 3.07 (m, 1H), 1.39 (m, 6H); EIMS *m/z* 216 (M^+); analysis for $C_{13}H_{16}N_2O$ (216.3), calcd, C, 72.19; H, 7.46; N, 12.95; found, C, 72.12; H, 7.37; N, 13.09.

6.2.2. 2,5-Diisopropyl-6-methoxy-8-quinolinamine (15). Yield: 88%; oil; IR (KBr) 3333 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.67 (d, 1H, *J* = 8.5 Hz), 6.90 (d, 1H, *J* = 8.5 Hz), 6.77 (s, 1H), 5.17 (bs, 2H), 4.15 (s, 3H), 3.17 (m, 2H), 1.42 (m, 6H), 1.37 (m, 6H); EIMS *m/z* 258 (M^+); analysis for $C_{16}H_{22}N_2O$ (258.4), calcd, C, 74.38; H, 8.58; N, 10.84; found, C, 74.37; H, 8.79; N, 10.57.

6.2.3. 2-Cyclopentyl-6-methoxy-8-quinolinamine (16). Yield: 92%; oil; IR (KBr) 2958, 2928 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.75 (d, 1H, *J* = 8.4 Hz), 6.88 (d, 1H, *J* = 8.4 Hz), 6.60 (s, 1H), 6.48 (s, 1H), 5.09 (bs, 2H), 3.89 (s, 3H), 3.57 (m, 1H), 1.88 (m, 8H); EIMS *m/z* 242 (M^+); analysis for $C_{15}H_{18}N_2O$ (242.3), calcd, C, 74.35; H, 7.49; N, 11.56; found, C, 74.39; H, 7.67; N, 11.88.

6.2.4. 2,5-Dicyclopentyl-6-methoxy-8-quinolinamine (17). Yield: 90%; oil; IR (KBr) 2930 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.92 (d, 1H, *J* = 8.3 Hz), 7.23 (d, 1H, *J* = 8.3 Hz), 6.57 (s, 1H), 5.09 (bs, 2H), 3.89 (s, 3H), 3.29 (m, 2H), 1.94 (m, 16H); EIMS *m/z* 310 (M^+); analysis for $C_{20}H_{26}N_2O$ (310.4), calcd, C, 77.38; H, 8.44; N, 9.02; found, C, 77.47; H, 8.71; N, 8.79.

6.2.5. 2-Cyclohexyl-6-methoxy-8-quinolinamine (18). Yield: 82%; oil; IR (KBr) 3008, 2926 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.80 (d, 1H, *J* = 8.4 Hz), 6.91 (d, 1H, *J* = 8.4 Hz), 6.66 (s, 1H), 6.50 (s, 1H), 5.10 (bs, 2H), 3.89 (s, 3H) 3.10 (m, 1H), 1.69 (m, 10H); EIMS *m/z* 256 (M^+); analysis for $C_{16}H_{20}N_2O$ (256.3), calcd, C, 74.97; H, 7.86; N, 10.93; found, C, 75.21; H, 8.03; N, 9.77.

6.2.6. 2,5-Dicyclohexyl-6-methoxy-8-quinolinamine (19). Yield: 88%; oil; IR (KBr) 3030 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.95 (d, 1H, *J* = 8.5 Hz), 7.28 (d, 1H, *J* = 8.5 Hz), 6.58 (s, 1H), 5.14 (bs, 2H), 3.84 (s, 3H) 3.23 (m, 2H), 1.77 (m, 20H); EIMS *m/z* 338 (M^+); analysis for $C_{22}H_{30}N_2O$ (338.5), calcd, C, 78.06; H, 8.93; N, 8.28; found, C, 78.32; H, 9.05; N, 7.99.

6.2.7. 2-Isopropyl-5,6-dimethoxy-8-quinolinamine (20). Yield: 93%; oil; IR (KBr) 3355 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.70 (d, 1H, *J* = 9.2 Hz), 7.23 (d, 1H, *J* = 9.2 Hz), 6.82 (s, 1H), 5.71 (bs, 2H), 4.14 (s, 3H), 4.12 (s, 3H), 3.10 (m, 1H), 1.20 (m, 6H); EIMS *m/z* 246 (M^+); analysis for $C_{14}H_{18}N_2O_2$ (246.3), calcd, C, 68.27; H, 7.37; N, 11.37; found, C, 68.53; H, 7.05; N, 11.44.

6.2.8. 2-*tert*-Butyl-5,6-dimethoxy-8-quinolinamine (21). Yield: 97%; oil; IR (KBr) 3461 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.32 (d, 1H, *J* = 8.85 Hz), 7.52 (d, 1H, *J* = 8.85 Hz), 6.81 (s, 1H), 5.1 (bs, 2H), 3.95 (s, 3H), 3.88 (s, 3H), 1.51 (s, 9H); ESIMS *m/z* 261 (M+1); analysis for $C_{15}H_{20}N_2O_2$ (260.3), calcd, C, 69.20; H, 7.74; N, 10.76; found, C, 68.97; H, 7.73; N, 10.55.

6.2.9. 2-Cyclohexyl-5,6-dimethoxy-8-quinolinamine (22). Yield: 80%; oil; IR (KBr) 3230 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.33 (d, 1H, *J* = 9.0 Hz), 7.51 (d, 1H, *J* = 9.0 Hz), 6.81 (s, 1H), 5.17 (bs, 2H), 3.95 (s, 3H), 3.88 (s, 3H), 3.1 (m, 1H), 1.45 (m, 10H); EIMS *m/z* 286 (M^+); analysis for $C_{17}H_{22}N_2O_2$ (286.4), calcd, C, 71.30; H, 7.74; N, 9.78; found, C, 71.55; H, 7.79; N, 10.12.

6.3. General method for the synthesis of 2-[4-(2-substituted/2,5-disubstituted-6-methoxy-8-quinolylamino)-pentyl]-1,3-isoindolinediones (23–31)

A mixture of 2-substituted/2,5-disubstituted-6-methoxy-8-quinolinamine (**14–22**, 4 mmol), 2-(4-bromopentyl)-1,3-isoindolinedione (4 mmol) and triethylamine (4 mmol) was heated at 120 °C with stirring for 4 h. An additional quantity of 2-(4-bromopentyl)-1,3-isoindolinedione (6 mmol) and triethylamine (4 mmol) was added, and stirring continued with heating for another 4 h. A third aliquot of 2-(4-bromopentyl)-1,3-isoindolinedione (4 mmol) and triethylamine (4 mmol) was added, and the reaction mixture stirred at 120 °C for additional 16 h. The dark brown reaction mixture was diluted with ethyl acetate (75 mL) and filtered. The filtrate was basified with 2N NaOH solution and extracted with ethyl acetate (3×50 mL). The combined extracts were washed with water (15 mL), dried over Na_2SO_4, and concentrated to afford dark colored residue. Flash column chromatography on silica gel using EtOAc/hexanes (20:80) provided 2-[4-(2-substituted/2,5-disubstituted-6-methoxy-8-quinolylamino)pentyl]-1,3-isoindolinediones (**23–31**) as colorless or pale yellow viscous oil.

6.3.1. 2-[4-(2-Isopropyl-6-methoxy-8-quinolylamino)pentyl]-1,3-isoindolinedione (23). Yield: 73%; oil; IR (KBr) 3433, 1709 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.88 (d, 1H, *J* = 8.3 Hz), 7.83 (m, 4H), 7.41 (d, 1H, *J* = 8.3 Hz), 6.27 (s, 1H), 6.24 (s, 1H), 4.81 (bs, 1H), 3.86 (s, 3H), 3.52 (m,

3H), 2.93 (m, 1H), 1.70 (m, 2H), 1.46 (m, 2H), 1.32 (m, 6H), 0.98 (d, 3H, $J=6.5$ Hz); EIMS m/z 431(M^+); analysis for $C_{26}H_{29}N_3O_3$ (431.5), calcd, C, 72.37; H, 6.77; N, 9.74; found, C, 72.44; H, 6.89; N, 9.93.

6.3.2. 2-[4-(2,5-Diisopropyl-6-methoxy-8-quinolylamino)-pentyl]-1,3-isoindolinedione (24). Yield: 77%; oil; IR (KBr) 3400, 1715 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.12 (d, 1H, $J=7.5$ Hz), 7.80 (m, 4H), 7.35 (d, 1H, $J=7.5$ Hz), 6.87 (s, 1H), 4.82 (bs, 1H), 3.87 (s, 3H), 3.53 (m, 3H), 3.10 (m, 2H,), 1.73 (m, 2H), 1.49 (m, 2H), 1.39 (m, 6H), 1.32 (m, 6H), 0.98 (d, 3H, $J=6.6$ Hz); EIMS m/z 473 (M^+); analysis for $C_{29}H_{35}N_3O_3$ (473.6), calcd, C, 73.54; H, 7.45; N, 8.87; found, C, 73.52; H, 7.49; N, 8.55.

6.3.3. 2-[4-(2-Cyclopentyl-6-methoxy-8-quinolylamino)-pentyl]-1,3-isoindolinedione (25). Yield: 74%; oil; IR (KBr) 1712 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.90 (d, 1H, $J=8.3$ Hz), 7.81 (m, 4H), 7.45 (d, 1H, $J=8.3$ Hz), 6.33 (s, 1H), 6.28 (s, 1H), 5.12 (bs, 1H), 3.89 (s, 3H), 3.69 (m, 3H), 3.46 (m, 1H), 2.02 (m, 4H), 1.69 (m, 8H), 1.22 (d, 3H, $J=6.9$ Hz); EIMS m/z 457 (M^+); analysis for $C_{28}H_{31}N_3O_3$ (457.6), calcd, C, 73.50; H, 6.83; N, 9.18; found, C, 73.57; H, 6.87; N, 8.91.

6.3.4. 2-[4-(2,5-Dicyclopentyl-6-methoxy-8-quinolylamino)-pentyl]-1,3-isoindolinedione (26). Yield: 89%; oil; IR (KBr) 1715 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.95 (d, 1H, $J=7.9$ Hz), 7.85 (m, 4H), 7.27 (d, 1H, $J=7.9$ Hz), 6.69 (s, 1H), 5.12 (bs, 1H), 3.89 (s, 3H), 3.65 (m, 3H), 3.44 (m, 2H), 2.07 (m, 4H), 1.62 (m, 16H), 1.21 (d, 3H, $J=6.9$ Hz); EIMS m/z 525 (M^+); analysis for $C_{33}H_{39}N_3O_3$ (525.7), calcd, C, 75.40; H, 7.48; N, 7.99; found, C, 75.07; H, 7.63; N, 8.11.

6.3.5. 2-[4-(2-Cyclohexyl-6-methoxy-8-quinolylamino)pentyl]-1,3-isoindolinedione (27). Yield: 63%; oil; IR (KBr) 3387, 1713 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.91 (d, 1H, $J=8.3$ Hz), 7.79 (m, 4H), 7.49 (d, 1H, $J=8.3$ Hz), 6.37 (s, 1H), 6.30 (s, 1H), 5.15 (bs, 1H), 3.89 (s, 3H), 3.68 (t, 2H), 3.32 (m, 1H), 3.18 (m, 1H), 2.22 (m, 4H), 1.50 (m, 10H), 1.10 (d, 3H, $J=6.9$ Hz); EIMS m/z 471 (M^+); analysis for $C_{29}H_{33}N_3O_3$ (471.6), calcd, C, 73.86; H, 7.05; N, 8.91; found, C, 73.52; H, 7.13; N, 8.63.

6.3.6. 2-[4-(2,5-Dicyclohexyl-6-methoxy-8-quinolylamino)-pentyl]-1,3-isoindolinedione (28). Yield: 88%; oil; IR (KBr) 3360, 1713 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.88 (d, 1H, $J=7.6$ Hz), 7.80 (m, 4H), 7.23 (d, 1H, $J=7.6$ Hz), 6.67 (s, 1H), 5.22 (bs, 1H), 3.88 (s, 3H), 3.65 (m, 2H), 3.33 (m, 1H), 3.15 (m, 2H), 2.22 (m, 4H), 1.65 (m, 20H), 1.17 (d, 3H, $J=6.7$ Hz); EIMS m/z 553 (M^+); analysis for $C_{35}H_{43}N_3O_3$ (553.7), calcd, C, 75.92; H, 7.83; N, 7.59; found, C, 75.97; H, 7.80; N, 7.48.

6.3.7. 2-[4-(2-Isopropyl-5,6-dimethoxy-8-quinolylamino)-pentyl]-1,3-isoindolinedione (29). Yield: 79%; oil; IR (KBr) 3260, 1720 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.10 (d, 1H, $J=8.0$ Hz), 7.82 (m, 4H), 7.37 (d, 1H, $J=8.0$ Hz), 6.84 (s, 1H), 4.81 (bs, 1H), 3.97 (s, 3H), 3.86 (s, 3H), 3.52 (m, 3H), 3.20 (m, 1H), 1.70 (m, 2H), 1.46 (m, 2H), 1.32 (m, 6H), 0.98 (d, 3H, $J=6.7$ Hz); ESIMS m/z 462 ($M+1$); analysis for $C_{27}H_{31}N_3O_4$ (461.6), calcd, C, 70.26; H, 6.77; N, 9.10; found, C, 70.34; H, 7.05; N, 8.93.

6.3.8. 2-[4-(2-*tert*-Butyl-5,6-dimethoxy-8-quinolylamino)-pentyl]-1,3-isoindolinedione (30). Yield: 66%; oil; IR (KBr) 3382, 1712 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.20 (d, 1H, $J=9.0$ Hz), 7.82 (m, 4H), 7.46 (d, 1H, $J=9.0$ Hz), 6.38 (s, 1H), 6.02 (bs, 1H), 3.96 (s, 3H), 3.84 (s, 3H), 3.69 (t, 2H), 3.64 (m, 1H), 1.73 (m, 4H) 1.41 (s, 9H), 1.31 (d, 3H, $J=6.9$ Hz); ESIMS m/z 476 ($M+1$); analysis for $C_{28}H_{33}N_3O_4$ (475.6), calcd, C, 70.71; H, 6.99; N, 8.84; found, C, 70.66; H, 6.89; N, 8.81.

6.3.9. 2-[4-(2-Cyclohexyl-5,6-dimethoxy-8-quinolylamino)-pentyl]-1,3-isoindolinedione (31). Yield: 70%; oil; IR (KBr) 3350, 1720 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.22 (d, 1H, $J=9.2$ Hz), 7.75 (m, 4H), 7.46 (d, 1H, $J=9.2$ Hz), 6.39 (s, 1H), 6.0 (bs, 1H), 3.96 (s, 3H), 3.84 (s, 3H), 3.69 (t, 2H), 3.64 (m, 1H), 3.20 (m, 1H), 1.75 (m, 4H) 1.41 (m, 10H), 1.35 (d, 3H, $J=6.7$ Hz); EIMS m/z 501 (M^+); analysis for $C_{30}H_{35}N_3O_4$ (501.6), calcd, C, 71.83; H, 7.03; N, 8.38; found, C, 71.91; H, 7.38; N, 8.38.

6.4. General method for the synthesis of N^8-(4-amino-1-methylbutyl)-2-substituted/2,5-disubstituted-6-methoxy-8-quinolinamines (32–40)

A solution of 2-[4-(2-substituted/2,5-disubstituted-6-methoxy-8-quinolylamino)pentyl]-1,3-isoindolinedione (**23–31**, 5 mmol) in 95% ethyl alcohol (25 mL), and hydrazine hydrate (100 mmol) was heated under reflux for 8 h. Solvent was removed under reduced pressure, and the residue was taken up in water (25 mL), and basified with 8N NaOH solution. Extracted with $CHCl_3$ (4×20 mL), and washed once with water (10 mL). Combined organic extracts were dried over Na_2SO_4, and concentrated under reduced pressure to yield N^8-(4-amino-1-methylbutyl)-2-substituted/2,5-disubstituted-6-methoxy-8-quinolinamines (**32–40**) as oil. Treatment with ethereal hydrochloric acid solution provided **32–40** as their hydrochloride salts.

6.4.1. N^8-(4-Amino-1-methylbutyl)-2-isopropyl-6-methoxy-8-quinolinamine ·2HCl (32). Yield: 61%; mp (salt) 99–102 °C (dec); IR (KBr) 3422 cm^{-1}; ^1H NMR (free base, $CDCl_3$) δ 7.94 (d, 1H, $J=8.6$ Hz), 7.42 (d, 1H, $J=8.6$ Hz), 6.30 (s, 1H), 6.25 (s, 1H), 6.10 (bs, 1H), 3.49 (s, 3H), 3.25 (m, 4H), 2.71 (m, 4H), 1.90 (bs, 2H), 1.52 (m, 6H), 1.15 (d, 3H, $J=6.3$ Hz); EIMS m/z 301 (M^+); analysis for $C_{18}H_{29}Cl_2N_3O$ (374.4), calcd, C, 57.75; H, 7.81; N, 11.22; found, C, 57.88; H, 7.67; N, 11.15.

6.4.2. N^8-(4-Amino-1-methylbutyl)-2,5-diisopropyl-6-methoxy-8-quinolinamine ·2HCl (33). Yield: 79%; mp (salt) 104–106 °C (dec); IR (KBr) 3430; ^1H NMR (free base, $CDCl_3$) δ 8.15 (d, 1H, $J=8.1$ Hz), 7.65 (m, 1H, $J=8.1$ Hz), 7.18 (s, 1H), 6.15 (bs, 1H), 3.72 (m, 1H), 3.45 (s, 3H), 3.29 (m, 4H), 2.73 (m, 4H), 1.95 (bs, 2H), 1.55 (m, 6H), 1.54 (m, 6H), 1.19 (d, 3H, $J=6.7$ Hz); EIMS m/z 343 (M^+); analysis for $C_{21}H_{35}Cl_2N_3O$ (416.4), calcd, C, 60.57; H, 8.47; N, 10.09; found, C, 60.45; H, 8.33; N, 10.37.

6.4.3. N^8-(4-Amino-1-methylbutyl)-2-cyclopentyl-6-methoxy-8-quinolinamine ·2HCl (34). Yield: 62%; mp (salt) 90–92 °C (dec); IR (KBr) 3294, 2953 cm^{-1}; ^1H NMR (free base, $CDCl_3$) δ 7.97 (d, 1H, $J = 8.2$ Hz), 7.45 (d, 1H, $J = 8.2$ Hz), 6.32 (s, 1H), 6.29 (s, 1H), 6.08 (bs, 1H), 3.89 (s, 3H), 3.71 (m, 1H) 3.48 (m, 2H) 3.12 (m, 1H), 2.64 (m, 4H), 1.93 (bs, 2H), 1.75 (m, 8H), 1.11 (d, 3H, $J = 6.9$ Hz); EIMS m/z 327 (M^+); analysis for $C_{20}H_{31}Cl_2N_3O$ (400.4), calcd, C, 60.00; H, 7.80; N, 10.49; found, C, 60.12; H, 7.72; N, 10.45.

6.4.4. N^8-(4-Amino-1-methylbutyl)-2,5-dicyclopentyl-6-methoxy-8-quinolinamine ·2HCl (35). Yield: 75%; mp (salt) 110–112 °C (dec); IR (KBr) 3300, 2955 cm^{-1}; ^1H NMR (free base, $CDCl_3$) δ 7.98 (d, 1H, $J = 8.1$ Hz), 7.22 (d, 1H, $J = 8.1$ Hz), 6.61 (s, 1H), 6.10 (bs, 1H), 3.89 (s, 3H), 3.71 (m, 1H) 3.45 (m, 2H) 3.15 (m, 2H), 2.65 (m, 4H), 1.91 (bs, 2H), 1.79 (m, 16H), 1.20 (d, 3H, $J = 6.4$ Hz); EIMS m/z 395 (M^+); analysis for $C_{25}H_{39}Cl_2N_3O$ (468.5), calcd, C, 64.09; H, 8.39; N, 8.97; found, C, 63.87; H, 8.44; N, 8.77.

6.4.5. N^8-(4-Amino-1-methylbutyl)-2-cyclohexyl-6-methoxy-8-quinolinamine ·2HCl (36). Yield: 59%; mp (salt) 101–104 °C (dec); IR (KBr) 3417 cm^{-1}; ^1H NMR (free base, $CDCl_3$) δ 7.92 (d, 1H, $J = 8.4$ Hz), 7.44 (d, 1H, $J = 8.4$ Hz), 6.36 (s, 1H), 6.28 (s, 1H), 6.05 (bs, 1H), 3.91 (s, 3H), 3.73 (m, 1H) 3.48 (m, 2H) 3.12 (m, 1H), 2.64 (m, 4H), 1.88 (bs, 2H), 1.75 (m, 10H), 1.11 (d, 3H, $J = 6.5$ Hz); EIMS m/z 341 (M^+); analysis for $C_{21}H_{33}Cl_2N_3O$ (414.4), calcd, C, 60.86; H, 8.03; N, 10.14; found, C, 60.66; H, 8.15; N, 10.33.

6.4.6. N^8-(4-Amino-1-methylbutyl)-2,5-dicyclohexyl-6-methoxy-8-quinolinamine ·2HCl (37). Yield: 75%; mp (salt) 88-90 °C (dec); IR (KBr) 3400 cm^{-1}; ^1H NMR (free base, $CDCl_3$) δ 7.93 (d, 1H, $J = 7.8$ Hz), 7.28 (d, 1H, $J = 7.8$ Hz), 6.75 (s, 1H), 6.15 (bs, 1H), 3.95 (s, 3H), 3.71 (m, 1H) 3.42 (m, 2H) 3.05 (m, 2H), 2.62 (m, 4H), 1.97 (bs, 2H), 1.78 (m, 20H), 1.15 (d, 3H, $J = 6.7$ Hz); EIMS m/z 423 (M^+); analysis for $C_{27}H_{43}Cl_2N_3O$ (496.4), calcd, C, 65.31; H, 8.73; N, 8.46; found, C, 65.45; H, 8.32; N, 8.44.

6.4.7. N^8-(4-Amino-1-methylbutyl)-2-isopropyl-5,6-dimethoxy-8-quinolinamine ·2HCl (38). Yield: 95%; mp (salt) 112–114 °C (dec); IR (KBr) 3330, 2900 cm^{-1}; ^1H NMR (free base, $CDCl_3$) δ 8.18 (d, 1H, $J = 7.8$ Hz), 7.35 (d, 1H, $J = 7.8$ Hz), 6.87 (s, 1H), 6.03 (bs, 1H), 3.95 (s, 3H), 3.82 (s, 3H), 3.55 (m, 4H), 1.97 (bs, 2H), 1.70 (m, 2H), 1.46 (m, 2H), 1.32 (m, 6H), 0.98 (d, 3H, $J = 6.8$ Hz); ESIMS m/z 331 ($M+1$); analysis for $C_{19}H_{31}Cl_2N_3O_2$ (404.4), calcd, C, 56.43; H, 7.73; N, 10.37; found, C, 56.47; H, 7.59; N, 10.07.

6.4.8. N^8-(4-Amino-1-methylbutyl)-2-*tert*-butyl-5,6-dimethoxy-8-quinolinamine ·2HCl (39). Yield: 96%; mp (salt) 122–125–102 °C (dec); IR (KBr) 3388, 2958 cm^{-1}; ^1H NMR (free base, $CDCl_3$) δ 8.23 (d, 1H, $J = 9.0$ Hz), 7.48 (d, 1H, $J = 9.0$ Hz), 6.40 (s, 1H), 6.03 (bs, 1H), 3.97 (s, 3H), 3.86 (s, 3H), 3.62 (m, 1H), 2.78 (t, 2H), 2.01 (bs, 2H), 1.78 (m, 4H), 1.42 (s, 9H), 1.33 (d, 3H, $J = 6.7$ Hz); EIMS m/z 345 (M^+); analysis for $C_{20}H_{33}Cl_2N_3O_2$ (418.4), calcd, C, 57.41; H, 7.95; N, 10.04; found, C, 57.33; H, 7.99; N, 10.23.

6.4.9. N^8-(4-Amino-1-methylbutyl)-2-cyclohexyl-5,6-dimethoxy-8-quinolinamine ·2HCl (40). Yield: 67%; mp (salt) 117–119 °C (dec); IR (KBr) 3310 cm^{-1}; ^1H NMR (free base, $CDCl_3$) δ 8.21 (d, 1H, $J = 9.1$ Hz), 7.49 (d, 1H, $J = 9.1$ Hz), 6.41 (s, 1H), 6.03 (bs, 1H), 3.97 (s, 3H), 3.86 (s, 3H), 3.62 (m, 1H), 2.78 (t, 2H), 1.91 (bs, 2H), 1.78 (m, 5H), 1.42 (m, 10H), 1.31 (d, 3H, $J = 6.9$ Hz); EIMS m/z 368 (M^+); analysis for $C_{22}H_{35}Cl_2N_3O_2$ (444.4), calcd, C, 59.45; H, 7.94; N, 9.45; found, C, 59.64; H, 8.24; N, 9.17.

6.5. General method for the synthesis of N^8-(4-amino-1-methylbutyl)-2-*tert*-butyl-6-methoxy-8-quinolinamine-Z-amino acid conjugates (41–44)

To an ice cooled stirred solution of N^8-(4-amino-1-methylbutyl)-2-*tert*-butyl-6-methoxy-8-quinolinamine (**2**, 1 mmol) and *Z*-L-amino acid (1.1 mmol) in CH_2Cl_2 (15 mL), DCC (1.1 mmol) was added. Reaction mixture was allowed to attain room temperature and stirring was continued for another 4 h. The reaction mixture was kept in refrigerator overnight and the separated 1,3-dicyclohexylurea (DCU) was filtered. Filtrate was washed with saturated sodium bicarbonate solution (3×5 mL) followed by water (2×5 mL), and dried over Na_2SO_4. The solvent was removed under reduced pressure to afford the crude product, which was purified by flash column chromatography on silica gel (230–400 mesh) using CH_3OH:$CHCl_3$ (2:98) to afford the N^8-(4-amino-1-methylbutyl)-2-*tert*-butyl-6-methoxy-8-quinolinamine-Z-amino acid conjugates (**41–44**) as viscous oil.

6.5.1. {1-[4-(2-*tert*-Butyl-6-methoxy-quinolin-8-ylamino)-pentylcarbamoyl]-ethyl}-carbamic acid benzyl ester (41). Yield: 93%; oil; IR (KBr) 3293, 1647 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.88 (d, 1H, $J = 8.5$ Hz), 7.44 (d, 1H, $J = 8.5$ Hz), 7.33 (m, 5H), 6.32 (s, 1H), 6.27 (s, 1H), 5.88 (bs, 1H), 5.34 (bs, 1H), 5.08 (s, 2H), 3.87 (m, 4H), 3.59 (m, 1H), 3.30 (m, 2H), 1.66 (m, 2H), 1.42 (s, 9H), 1.31 (d, 3H, $J = 6.7$ Hz), 0.91 (m, 6H); ESIMS m/z 521 ($M+1$); analysis for $C_{30}H_{40}N_4O_4$ (520.7), calcd, C, 69.20; H, 7.74; N, 10.76; found, C, 69.34; H, 7.85; N, 10.79.

6.5.2. {1-[4-(2-*tert*-Butyl-6-methoxy-quinolin-8-ylamino)-pentylcarbamoyl]-2-methyl-propyl}-carbamic acid benzyl ester (42). Yield: 92%; oil; IR (KBr) 3394, 1712, 1655 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.87 (d, 1H, $J = 8.5$ Hz), 7.43 (d, 1H, $J = 8.5$ Hz), 7.22 (m, 5H), 6.31 (s, 1H), 6.25 (s, 1H), 6.20 (bs, 1H), 5.18 (s, 2H), 5.04 (bs, 1H), 4.13 (m, 2H), 3.87 (s, 3H), 3.60 (m, 2H), 3.30 (m, 4H), 1.68 (m, 4H), 1.42 (s, 9H), 1.31 (d, 3H, $J = 6.7$ Hz), 1.12 (m, 6H); ESIMS m/z 549 ($M+1$); analysis for $C_{29}H_{46}N_4O_4$ (548.7), calcd, C, 70.04; H, 8.08; N, 10.21; found, C, 70.17; H, 8.36; N, 10.37.

6.5.3. {4-Benzyloxycarbonylamino-1-[4(2-*tert*-butyl-6-methoxy-quinolin-8-ylamino)-pentyl-carbamoyl]-butyl}-carbamic acid benzyl ester (43). Yield: 98%; oil; IR (KBr) 3430, 1714, 1666 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.85

(d, 1H, $J=8.6$ Hz), 7.42 (d, 1H, $J=8.6$ Hz), 7.34 (m, 10H), 6.44 (bs, 1H), 6.29 (s, 1H), 6.24 (s, 1H), 6.12 (bs, 1H), 5.56 (bs, 1H), 5.08 (m, 4H), 4.36 (m, 1H), 4.23 (m, 1H), 3.88 (s, 3H), 3.72 (m, 4H), 3.56 (m, 1H), 3.21 (m, 4H), 1.55 (m, 4H), 1.42 (s, 9H), 1.26 (d, 3H, $J=6.5$ Hz); APCIMS m/z 698 (M + 1); analysis for $C_{40}H_{51}N_5O_6$ (697.9), calcd, C, 68.84; H, 7.37; N, 10.04; found, C, 68.88; H, 7.22; N, 9.85.

6.5.4. {5-Benzyloxycarbonylamino-1-[4-(2-*tert*-butyl-6-methoxy-quinolin-8-ylamino)-pentyl-carbamoyl]-pentyl}-carbamic acid benzyl ester (44). Yield: 91%; oil; IR (KBr) 3304, 1719, 1690 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.86 (d, 1H, $J=8.6$ Hz), 7.43 (d, 1H, $J=8.6$ Hz), 7.31 (m, 10H), 6.31 (s, 1H), 6.25 (s, 1H), 6.15 (bs, 1H), 5.50 (bs, 1H), 5.07 (s, 4H), 4.82 (bs, 1H), 4.13 (m, 1H), 3.85 (s, 3H), 3.58 (m, 2H), 3.27 (m, 2H), 3.12 (m, 2H), 1.67 (m, 10H), 1.41 (s, 9H), 1.25 (d, 3H, $J=6.8$ Hz); ESIMS m/z 712 (M + 1); analysis for $C_{41}H_{53}N_5O_6$ (711.9), calcd, C, 69.17; H, 7.50; N, 9.84; found, C, 69.34; H, 7.44; N, 9.94.

6.6. General method for the synthesis of N^8-(4-amino-1-methylbutyl)-2-*tert*-butyl-6-methoxy-8-quinolinamine-amino acid conjugates (45–48)

To a mixture of Z-amino acid linked 2-*tert*-butylprimaquine derivatives (41–44, 1 mmol), glacial acetic acid (1 mL) and 10% Pd-C (200 mg) in methanol (30 mL) was bubbled a slow stream of hydrogen gas for 1 h. The catalyst was removed by filtration, and filtrate was concentrated in vacuum to afford the product as oil, which upon treatment with a solution of ethereal hydrogen chloride provided the amino acid conjugates as hydrochloride salt.[8]

6.6.1. N^1-[4-(2-*tert*-Butyl-6-methoxy-8-quinolylamino)-pentyl]-(2*S*)-2-aminopropanamide ·2HCl (45). Yield: 98%; mp (salt) 89–91 °C (dec.); IR (KBr) 3018, 1710 cm^{-1}; ^1H NMR (free base, $CDCl_3$) δ 7.86 (d, 1H, $J=7.8$ Hz), 7.65 (s, 1H, $J=7.8$ Hz), 7.15 (s, 1H), 6.81 (s, 1H), 6.20 (bs, 1H), 5.48 (bs, 1H), 4.30 (bs, 2H), 3.99 (m, 1H), 3.87 (s, 3H), 3.71 (m, 1H), 3.26 (m, 2H), 1.84 (m, 4H), 1.50 (s, 9H), 1.25 (m, 6H); ESIMS m/z 387 (M + 1); analysis for $C_{22}H_{36}Cl_2N_4O_2$ (459.5), calcd, C, 57.51; H, 7.90; N, 12.19; found, C, 57.58; H, 7.68; N, 12.02.

6.6.2. N^1-[4-(2-*tert*-Butyl-6-methoxy-8-quinolylamino)-pentyl]-(2*S*)-2-amino-3-methyl-butan-amide 2HCl (46). Yield: 94%; mp (salt) 97–99 °C (dec.); IR (KBr) 3243, 1663 cm^{-1}; ^1H NMR (free base, $CDCl_3$) δ 7.86 (d, 1H, $J=8.5$ Hz), 7.61 (bs, 1H), 7.43 (d, 1H, $J=8.5$ Hz), 6.31 (s, 1H), 6.26 (s, 1H), 4.80 (bs, 1H), 4.30 (bs, 2H), 3.87 (s, 3H), 3.62 (m, 1H) 3.35 (m, 2H), 2.72 (m, 2H), 1.68 (m, 4H), 1.41 (s, 9H), 1.29 (d, 3H, $J=6.5$ Hz), 0.945 (m, 6H); ESIMS m/z 414 (M + 1); analysis for $C_{24}H_{40}Cl_2N_4O_2$ (487.5), calcd, C, 59.13; H, 8.27; N, 11.49; found, C, 59.27; H, 8.39; N, 11.67.

6.6.3. N^1-[4-(2-*tert*-Butyl-6-methoxy-8-quinolylamino)-pentyl]-(2*S*)-2,5-diaminopentamide ·3HCl (47). Yield: 99%; mp (salt) 100–101 °C (dec.); IR (KBr) 3018 cm^{-1}; ^1H NMR (free base, $CDCl_3$) δ 7.85 (d, 1H, $J=7.9$ Hz), 7.42 (d, 1H, $J=7.9$ Hz), 6.30 (s, 1H), 6.23 (s, 1H), 3.85 (s, 3H), 3.58 (m, 1H), 3.44 (m, 1H), 3.25 (m, 4H), 2.95 (m, 4H), 1.91 (m, 10H), 1.40 (s, 9H), 1.26 (d, 3H, $J=6.6$ Hz); ESIMS m/z 429 (M + 1); analysis for $C_{24}H_{42}Cl_3N_5O_2$ (539.0), calcd, C, 53.48; H, 7.85; N, 12.99; found, C, 53.35; H, 7.97; N, 12.78.

6.6.4. N^1-[4-(2-*tert*-Butyl-6-methoxy-8-quinolylamino)-pentyl]-(2*S*)-2-6-diaminohexanamide ·3HCl (48). Yield: 98%; mp (salt) 107–109 °C (dec.); IR (KBr) 3435, 1667 cm^{-1}; ^1H NMR (free base, $CDCl_3$) δ 7.85 (d, 1H, $J=8.5$ Hz), 7.42 (d, 1H, $J=8.5$ Hz), 6.29 (s, 1H), 6.24 (s, 1H), 5.64 (bs, 4H), 3.89 (s, 3H), 3.68 (m, 1H), 3.24 (m, 4H), 2.87 (m, 1H), 1.88 (m, 8H), 1.63 (m, 4H), 1.41 (s, 9H), 1.24 (d, 3H, $J=6.7$ Hz); ESIMS m/z 444 (M + 1); analysis for $C_{25}H_{44}Cl_3N_5O_2$ (553.0), calcd, C, 54.30; H, 8.02; N, 12.66; found, C, 54.68; H, 8.11; N, 12.47.

Acknowledgements

Generous financial support from the Council of Scientific and Industrial Research (CSIR), New Delhi, India [Grant no. 01(1555)/98/EMR-II] for the synthesis of primaquine derivatives is gratefully acknowledged.

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