Exploration of a New Type of Antimalarial Compounds Based on Febrifugine

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Febrifugine (**1**), a quinazoline alkaloid, isolated from *Dichroa febrifuga* roots, shows powerful antimalarial activity against *Plasmodium falciparum*. The use of **1** as an antimalarial drug has been precluded because of side effects, such as diarrhea, vomiting, and liver toxicity. However, the potent antimalarial activity of **1** has stimulated medicinal chemists to pursue compounds derived from **1**, which may be valuable leads for novel drugs. In this study, we synthesized a new series of febrifugine derivatives formed by structural modifications at (i) the quinazoline ring, (ii) the linker, or (iii) the piperidine ring. Then, we evaluated their antimalarial activities. Thienopyrimidine analogue **15** exhibited a potent antimalarial activity and a high therapeutic selectivity both in vitro and in vivo, suggesting that **15** is a good antimalarial candidate.

Introduction

Malaria, which is caused by a protozoan parasite of the genus *Plasmodium*, is a major parasitic infection in many tropical and subtropical regions. Malaria affects 300−500 million patients worldwide and leads to more than 2 million deaths each year. Although malaria has been widely eradicated in many parts of the world, the number of patients continues to rise mainly due to the emergence of chloroquine-resistant and multiple-drug-resistant strains of malaria parasites. Thus, the discovery of new and effective antimalarial drugs is urgently needed.[1]

The roots of *Dichroa febrifuga*, a plant that belongs to the Saxifragaceae family, have been used as a traditional antimalarial drug in China. The quinazoline-type alkaloids, febrifugine **1** and its stereoisomer isofebrifugine **2**, have been identified as the active components of these roots (Chart 1).[2] Although **1** and **2** show powerful in vitro antimalarial activity against both chloroquine-sensitive *P. falciparum* FCR-3 and chloroquine-resistant *P.* falciparum K1, the in vivo activity of **1** against mouse malaria, *P. berghei*, is approximately 200 times more potent than that of **2**. Because of side effects, such as diarrhea, vomiting,[3] and liver toxicity,[4] **1** has been precluded as an antimalarial drug. However, the potent antimalarial activity of **1** has stimulated medicinal chemists to pursue derivatives of **1**, which may provide valuable leads for novel drugs. We have tried to create active febrifugine analogues.[5−7] However, the role of the structural components of **1** in its antimalarial activity has yet to be elucidated.

Therefore, we decided to synthesize febrifugine derivatives formed by structural modifications at (i) the quinazoline ring, (ii) the linker, or (iii) the piperidine ring (Figure 1). In this article, we report the syntheses of these febrifugine derivatives and evaluate their in vitro and in vivo antimalarial activities.

Results and Discussion

Syntheses of Febrifugine Analogues. (i) Quinazoline Ring. We synthesized quinazoline ring-modified derivatives **7**−**9** and **14**−**16** (Scheme 1). 6-Fluoro analogue **7** and 2-methyl analogue

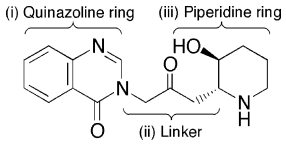
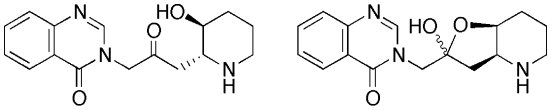


Figure 1. The three parts of febrifugine (**1**).

Chart 1. Structures of Febrifugine (**1**) and Isofebrifugine (**2**)



8 are based on febrifugine metabolites feb-A and -B,[5] respectively, which prevent metabolic oxidation. In compounds **9** and **14**−**16**, the quinazoline ring of **1** is replaced by purine, benzotriazine, thienopyrimidine, and quinoline rings, respectively. The antimalarial activity and cytotoxicity of the racemic analogues of **1** are due to the natural enantiomers because it was reported that both activities of the synthesized unnatural enantiomer of **1** were approximately 1/2000 and 1/100 of those of natural **1**, respectively.[8] Therefore, we synthesized **7**−**9** and **14**−**16** as racemic compounds.

Racemic **7**−**9** were synthesized by employing the synthetic strategy for **1** developed by Takeuchi et al.[9] The starting material, 3-hydroxypyridine, was used to produce hemiacetal **4** via compound **3**. A coupling reaction of **4** with the aromatic component of **7** provided compound **5a**, which was hydrogenolized to give **6a**. As in the isomerization of isofebrifugine (**2**) into febrifugine (**1**), the boiling of the MeOH solution of **6a** isomerized **6a** into **7**.[9b,10] Analogues **8** and **9** were synthesized in a similar manner using the corresponding heterocyclic component.

However, the syntheses of **14**−**16** were carried out in a different manner from those of **7**−**9** because the coupling reaction or catalytic hydrogenation was unsuccessful. Compound **3** was converted into Boc-derivative **10** in two steps. After the MOM-etherification of **10**, the epoxidation by 3-chloroperbenzoic acid afforded **11**. A coupling reaction with the aromatic component of **14** and subsequent oxidation gave **12a**. The acidic deprotection of **12a** and refluxing the residue in MeOH gave a mixture of **13a** and **14**. The isomerization of **13a** into **14** also

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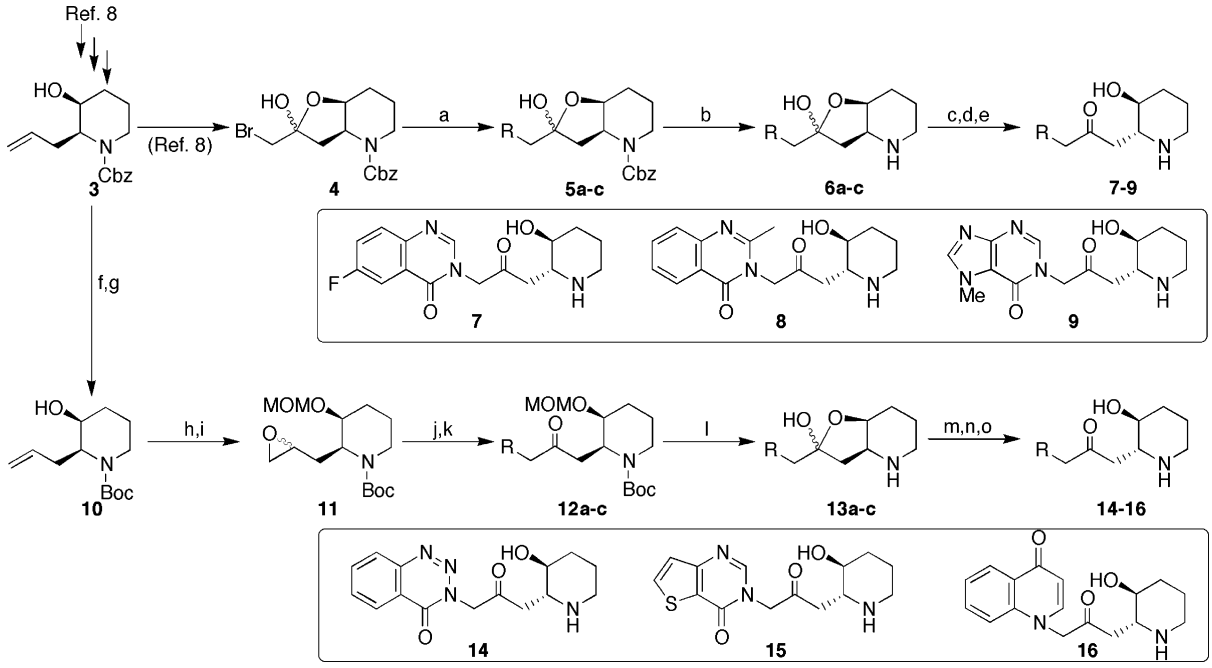
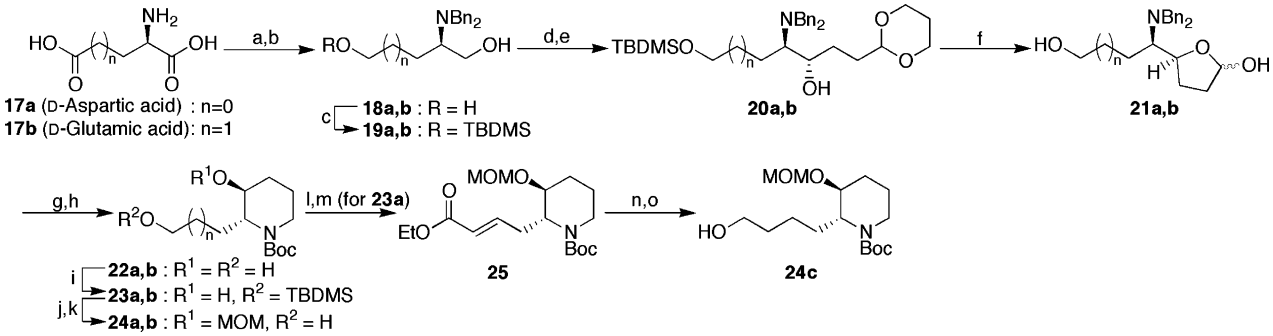
Scheme 1. Synthesis of **7**−**9** and **14**−**16**[a]



[a] Reagents and conditions: (a) corresponding aromatic compound, K_2CO_3, DMF, rt; (b) H_2 (1 atm), $Pd(OH)_2$/C, MeOH, rt (65% (**6a**), 80% (**6b**) and 78% (**6c**) (2 steps)); (c) MeOH, reflux; (d) Boc_2O, *i*Pr_2EtN, CH_2Cl_2, rt; (e) 10% HCl−MeOH, rt (48% (**7**), 42% (**8**) and 40% (**9**) (3 steps)); (f) 6 M HCl, reflux; (g) Boc_2O, 5 M NaOH, rt (61% (2 steps)); (h) MOMCl, *i*Pr_2EtN, CH_2Cl_2, rt; (i) *m*CPBA, CH_2Cl_2, $NaHCO_3$, 0 °C (44% (2 steps)); (j) corresponding aromatic compound, KH, DMF, rt; (k) Dess−Martin periodinane, CH_2Cl_2, rt (71% (**12a**), 31% (**12b**) and 39% (**12c**) (2 steps)); (l) 10% HCl-MeOH, rt (97% (**13a**), 98% (**13b**) and 95% (**13c**)); (m) MeOH, reflux; (n) Boc_2O, *i*Pr_2EtN, CH_2Cl_2, rt; (o) 10% HCl−MeOH, rt (41% (**10**), 28% (**11**) and 30% (**12**) (3 steps)).

Scheme 2. Synthesis of the Piperidine and Linker Parts of **28**−**30**[a]



[a] Reagents and conditions: (a) BnBr, K_2CO_3, NaOH, MeOH−H_2O (1:1), reflux; (b) $LiAlH_4$, THF, 0 °C (34% (**18a**) and 62% (**18b**) (2 steps)); (c) TBDMSCl, imidazole, CH_2Cl_2, 0 °C (42% (**19a**) and 56% (**19b**)); (d) $(COCl)_2$, DMSO, Et_3N, CH_2Cl_2, −78 °C; (e) (1,3-dioxan-2-ylethyl)magnesium bromide, THF, −78 °C (56% (**20a**) and 59% (**20b**) (2 steps)); (f) 2M HCl, acetone, rt; (g) H_2 (1 atm), $Pd(OH)_2$/C, MeOH, rt; (h) Boc_2O, Et_3N, MeOH, rt (66% (**22a**) and 33% (**22b**) (3 steps)); (i) TBDMSCl, imidazole CH_2Cl_2, 0 °C (82% (**23a**) and 55% (**23b**)); (j) MOMCl, *i*Pr_2EtN, DME, 50 °C; (k) TBAF, THF, 0 °C (83% (**24a**) and 93% (**24b**) (2 steps)); (l) Dess−Martin periodinane, CH_2Cl_2, 0 °C; (m) triethyl phosphonoacetate, NaH, toluene, 0 °C (76% (2 steps)); (n) H_2 (1 atm), $Pd(OH)_2$/C, THF, rt; (o) DIBAL-H, toluene, −78 °C (84% (2 steps)).

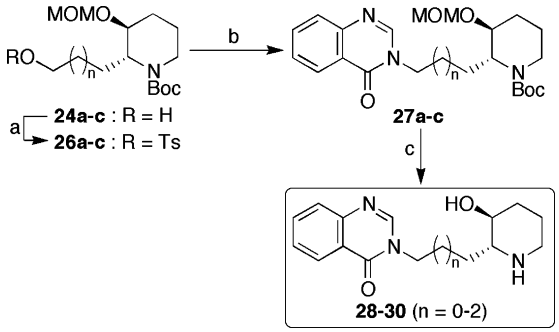
occurred. Analogues **15** and **16** were synthesized in a similar manner from **3**.

(ii) Linker Part. To evaluate the role of the C-2′ carbonyl group in antimalarial activity, we synthesized chiral compound **29** (Scheme 3), which is a decarbonylated derivative of febrifugine **1**. The piperidine and linker parts of **29** were synthesized by using *N*,*N*-dibenzylamino aldehyde[11] as a chiral building block (Scheme 2). D-Glutamic acid (**17b**) was converted into amino alcohol **18b** by a reaction with benzyl bromide and a reduction using lithium aluminum hydride. Compound **18b** was reacted with *tert*-butyldimethylsilyl chloride to yield **19b**.[12] The Swern oxidation of **19b** gave *N*,*N*-dibenzylamino aldehyde, which was then reacted with (1,3-dioxan-2-ylethyl)magnesium bromide[13,14] to diastereoselectively give **20b**. The hydrolysis of the acetal **20b** gave hemiacetal **21b**. Catalytic hydrogenation followed by *N*-protection afforded **22b**. Three successive reactions of **22b** gave mono-MOM ether **24b**, which contained the piperidine and linker parts of **29**. In Scheme 2, tosylation of **24b** afforded **26b**, which was coupled with 4-hydroxyquinazoline to produce **27b**. Finally, compound **27b** was deprotected under acidic conditions, which resulted in a decarbonylated febrifugine **29** as a hydrochloride.

In the same manner, analogues **28** and **30**, which contain shorter and longer linkers, respectively, were synthesized. D-Aspartic acid (**17a**) was used as the starting material to generate **24a** (Scheme 2). The oxidation of **24a** followed by the Horner−Emmons reaction with triethyl phosphonoacetate yielded **25**. After the catalytic hydrogenation of **25**, the reduction by diisobutylaluminum hydride gave **24c**. Compounds **24a** and **24c** were converted into **28** and **30**, respectively (Scheme 3).

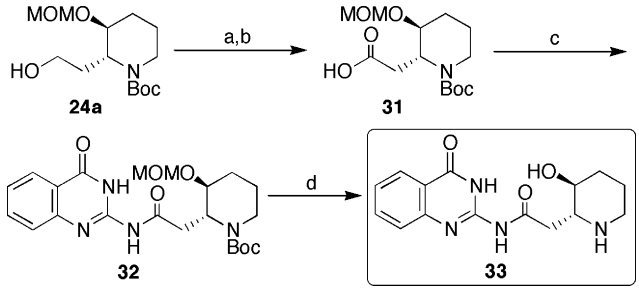
We synthesized compound **33**, which has an amide bond for the linkage between the quinazoline ring and the linker (Scheme 4). The successive oxidation of **24b** by pyridinium chlorochromate and sodium hypochlorite gave carboxylic acid **31**. A

Scheme 3. Synthesis of **28**−**30**[a]



[a] Reagents and conditions: (a) *p*TsCl, pyridine, rt (69% (**26a**), 80% (**26b**) and 78% (**26c**)); (b) 4-hydroxyquinazoline, K_2CO_3, DMF, 50 °C (30% (**27a**), 72% (**27b**) and 84% (**27c**)); (c) 10% HCl-MeOH, 50 °C (90% (**28a**), 88% (**28b**) and 93%(**28c**)).

Scheme 4. Synthesis of **33**[a]



[a] Reagents and conditions: (a) Dess−Martin periodinane, CH_2Cl_2, 0 °C; (b) $NaClO_2$, 2-methyl-2-butene, NaH_2PO_4, *t*BuOH, H_2O, rt (83% (2 steps)); (c) 2-amino-4(*3H*)-quinazolinone, HATU, *i*Pr_2EtN, DMF, CH_2Cl_2, rt (38%); (d) 10% HCl-MeOH, rt (45%).

coupling reaction of **31** with 2-amino-4(*3H*)-quinazolinone[15] provided compound **32**, which was deprotected under acidic conditions to afford **33**.

(iii) Piperidine Ring. Piperidine ring-opened analogue **41** was synthesized as shown in Scheme 5. The Swern oxidation of amino alcohol **19a**, which was prepared from D-aspartic acid **17a**, and a subsequent reaction with ethylmagnesium bromide diastereoselectively gave **34**. Four successive reactions, O-protection by MOM group, debenzylation, N-protection by a Boc group, and deprotection of the TBDMS group, afforded **37**. The Dess−Martin oxidation of **37** gave the aldehyde, which was treated with trimethylsulfoxonium iodide and sodium hydride to yield epoxide **38**. After N-methylation of **38**, a coupling reaction with 4-hydroxyquinazoline and subsequent oxidation gave **40**. Deprotection of **40** under 10% hydrogen chloride in MeOH, formed the unstable hydrochloride of **41**. The treatment of **40** with trifluoroacetic acid afforded the stable trifluoroacetate of **41**. ^{1}H and ^{13}C NMR spectra showed that compound **41** exists as a hemiacetal form.

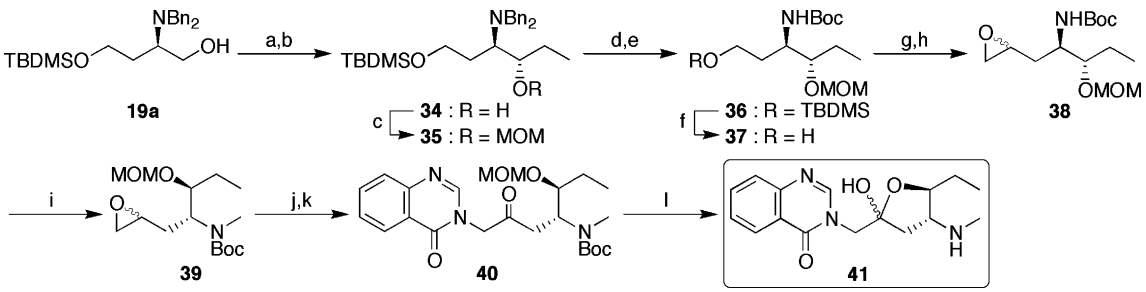
The position of the linkage between the piperidine ring and linker is changed in febrifugine analogues **46** and **50**. To synthesize **46** (Scheme 6), the hydroxy group of 3-hydroxypiperidine **42** was protected as TIPS ether by three sequential reactions to give **43**. The treatment of **43** with epibromohydrin afforded **44**, which was coupled with 4-hydroxyquinazoline to yield **45**. The oxidation of **45** and subsequent deprotection gave racemic **46** as the hydrochloride.

To synthesize **50**, the *N*-Boc derivative of **42** was treated with 2,2-dimethyl-1,3-dioxolan-4-ylmethyl tosylate to give **47**. After deprotection under acidic conditions and N-reprotection of **47**, the epoxidation by *N*-tosylimidazole afforded **48**. Finally, three successive reactions, a coupling with 4-hydroxyquinaoline, Dess−Martin oxidation, and acidic deprotection, yielded racemic **50** as the hydrochloride (Scheme 7).

Antimalarial Activity in Vitro of the Synthetic Analogues of Febrifugine. The in vitro antimalarial activity of febrifugine analogues **7**−**9**, **14**−**16**, **28**−**30**, **33**, **41**, **46**, and **50** against *P. falciparum* and the cytotoxicity against mouse L929 cells were evaluated (Table 1). Compounds **7**−**9**, **14**, and **15** displayed antimalarial activity (EC_{50} <0.5 μg/mL). The antimalarial activity of 6-fluoro analogue **7** and thienopyrimidine ring-containing analogue **15** were equivalent to that of febrifugine (**1**). These results suggest that a suitable method to create more active derivatives is to modify the quinazoline ring of **1**. On the contrary, compound **16** did not display any activity, indicating that the nitrogen atom at position 3 in the quinazoline ring is crucial to activity. The antimalarial activity of decarbonylated derivative **29** was 1/15 of that of **1**. Therefore, the carbonyl group at C-2" was effective but not critical to activity. Compounds **28** and **30**, which have two and four carbon linkers, respectively, did not display activity, suggesting that a three carbon linker is important for potent antimalarial activity. In addition, compounds **33**, **41**, **46**, and **50** did not exhibit activity. The piperidine ring of febrifugine **1** should be maintained to create useful derivatives.

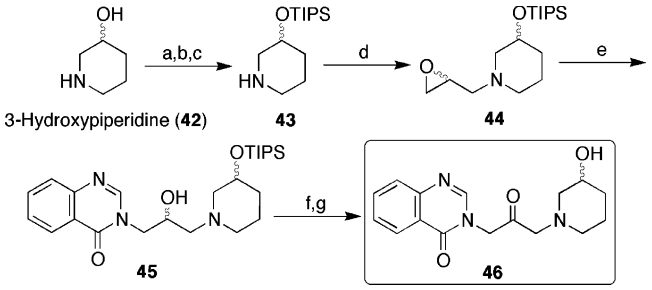
Antimalarial Activity in Vivo of the Synthetic Analogues of Febrifugine. The in vivo antimalarial activities against rodent malaria *P. berghei* and the acute toxicity in mice was examined for **7**, **15**, and **29**, which were more potent in vitro than chloroquine (Table 2). Because **7** was highly toxic in mice (LD_{50} <2.5 mg/kg), the in vivo antimalarial activity of **7** was not evaluated. The antimalarial activity (ED_{50} 2.95 mg/kg) of **15** was comparable to that of chloroquine (ED_{50} 1.53 mg/kg), a clinically used medicine. The ED_{50} value of **15** was also similar to the reported value of artemisinin derivatives (ED_{50} 3−10 mg/kg).[16] In addition, the toxicity of **15** was much weaker (LD_{50}

Scheme 5. Synthesis of **41**[a]



[a] Reagents and conditions: (a) $(COCl)_2$, DMSO, Et_3N, CH_2Cl_2, −78 °C; (b) EtMgBr, THF, −78 °C (66% (2 steps)); (c) MOMCl, *i*Pr_2EtN, CH_2Cl_2, rt (79%); (d) H_2 (1 atm), $Pd(OH)_2$/C, MeOH, rt; (e) Boc_2O, Et_3N, MeOH, 0 °C (98% (2 steps)); (f) TBAF, THF, rt (67%); (g) Dess−Martin periodinane, CH_2Cl_2, rt; (h) trimethylsulfoxonium iodide, NaH, DMSO, rt (75% (2 steps)); (i) MeI, NaH, DMF, 0 °C (91%); (j) 4-hydroxyquinazoline, NaH, DMF, 80 °C; (k) Dess−Martin periodinane, CH_2Cl_2, rt (41% (2 steps)); (l) TFA, CH_2Cl_2, rt (44%).

Scheme 6. Synthesis of **46**[a]



[a] Reagents and conditions: (a) CbzCl, Et_3N, CH_2Cl_2, 0 °C (96%); (b) TIPSCl, imidazole, DMF, 0 °C; (c) H_2 (1 atm), $Pd(OH)_2/C$, MeOH, rt (71% (2 steps)); (d) epibromohydrin, K_2CO_3, DMF, rt (71%); (e) 4-hydroxyquinazoline, NaH, DMF, 80 °C (68%); (f) $(COCl)_2$, DMSO, Et_3N, CH_2Cl_2, −78 °C (72%); (g) 10% HCl-MeOH, 70 °C (80%).

Table 1. Antimalarial Activities of Synthesized Febrifugine Derivatives against *P. falciparum* in Vitro

	antimalarial activity		cytotoxicity[c]	
	FCR-3[a]	K1[b]		
compound	EC_{50} (μg/mL)		EC_{50} (μg/mL)	selectivity[d]
febrifugine (**1**)	0.00142	0.00140	0.167	118
7	0.000730	0.000916	0.0790	108
8	0.116	0.190	15.6	134
9	0.506	0.943	>100	>198
14	0.154	N.T.[e]	16.4	106
15	0.00306	0.00319	0.563	184
16	>1	>1	>100	
28	>1	>1	>100	
29	0.0225	0.0236	4.45	198
30	>1	>1	>100	
33	>1	>1	>100	
41	>1	>1	6.44	<6.44
46	>1	>1	>100	
50	>1	>1	>100	
chloroquine	0.0475	0.375	18.6	392
artemisinin	0.0179	0.0117	>100	>5590

[a] Against *P. falciparum* FCR-3 (chloroquine-sensitive strain). [b] Against *P. falciparum* K1 (chloroquine-resistant strain). [c] Against mouse L929 cells. [d] Selectivity = EC_{50} for L929 cells/EC_{50} for *P. falciparum* FCR-3. [e] N.T., not tested.

88 mg/kg) than that of **1**, and the therapeutic index of **15** was higher than that of **1**. These results suggest that **15** may be a good antimalarial candidate. Although no mice died after receiving 320 mg/kg of **29** in the acute toxicity test, its antimalarial activity was moderate (ED_{50} 22.5 mg/kg).

In conclusion, we synthesized and evaluated a new series of febrifugine derivatives. Among them, thienopyrimidine analogue **15** exhibited potent antimalarial activity and a high therapeutic selectivity both in vitro and in vivo. Further studies on **15**, such as a metabolic analysis and the elucidation of the action mechanism, are necessary to develop a novel antimalarial drug.

Table 2. Antimalarial Activities against *P. berghei* in Vivo and Acute Toxicities of Synthesized Febrifugine Derivatives **7**, **15**, and **29**[a]

	antimalarial activity[b]	acute toxicity[c]	
compound	ED_{50} (mg/kg)	LD_{50} (mg/kg)	therapeutic index[d]
febrifugine (**1**)	0.41	7.1	17
7	N.T.[e]	<2.5	
15	2.95	88	30
29	22.5	>320	>14
chloroquine	1.53	N.T.[e]	

[a] All compounds were administrated by po. [b] Against *P. berghei* (rodent malaria). [c] Toxicity in mice. [d] Therapeutic index = LD_{50} in mice/ED_{50} for *P. berghei* in mice. [e] N.T., not tested.

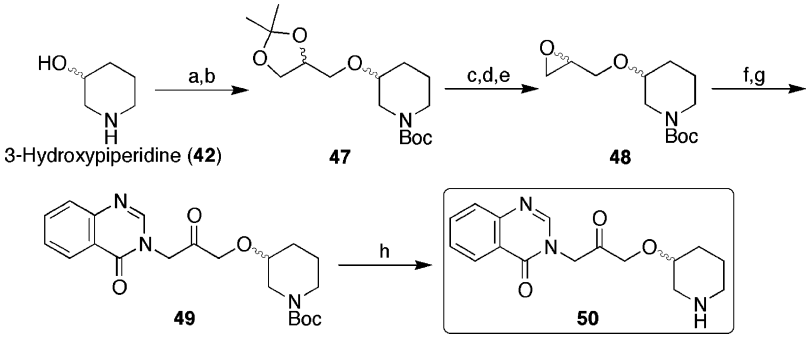
Experimental Section

General Methods. Starting materials were either commercially available or prepared as reported previously in the literature. Analytical TLC was performed on silica gel 60 F_{254} (Merck) and Aluminumoxid 150 F_{254} neutral (Type T, Merck). Column chromatography was carried out on silica gel 60 (70−230 mesh, Merck), and aluminum oxide 150 basic (type T, Merck). NMR spectra were recorded on JEOL JNM ECA-600, ECP-500, and AL-400. Mass spectra were measured on JEOL JMS AX-500 and AX-700. Elemental analyses were performed by Yanaco CHN Corder MT-6.

6-Fluoro-3-[(3a*S,7a*S**)-2-hydroxyoctahydrofuro[3,2-*b*]pyridin-2-ylmethyl]-3*H*-quinazolin-4-one (6a).** To a solution of **4**[9] (579 mg, 1.57 mmol) in DMF (6 mL) were added 6-fluoro-3*H*-quinazolin-4-one[17] (262 mg, 1.59 mmol) and K_2CO_3 (220 mg, 1.59 mmol) at room temperature. After stirring for 3 h, the mixture was poured into brine and extracted three times with EtOAc. The organic layer was washed with brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (49:1) to give crude **5a**. 20% $Pd(OH)_2$ on carbon (150 mg) was added to the MeOH solution (12 mL) of this crude product. After stirring for 1 h under a hydrogen atmosphere, the mixture was filtered and concentrated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (9:1) to give **6a** (314 mg, 65% (2 steps)). Using a similar procedure, compounds **6b** (yield: 80%) and **6c** (78%) were prepared from 2-methyl-3*H*-quinazolin-4-one and **7**-methylhypoxanthine, respectively.

6-Fluoro-3-[3-[(2*R,3*S**)-3-hydroxypiperidin-2-yl]-2-oxopropyl]-3*H*-quinazolin-4-one (7).** Compound **6a** (386 mg, 1.21 mmol) was dissolved in MeOH (20 mL). After refluxing for 3 h, the reaction mixture was concentrated. To a solution of this residue in CH_2Cl_2 (12 mL) were added triethylamine (190 μL, 1.32 mmol) and di-*tert*-butyl dicarbonate (526 mg, 2.41 mmol) at room temperature. After stirring for 1.5 h, the mixture was poured into

Scheme 7. Synthesis of **50**[a]



[a] Reagents and conditions: (a) Boc_2O, Et_3N, MeOH, rt (95%); (b) 2,2-dimethyl-1,3-dioxolan-4-ylmethyl *p*-toluenesulfonate, NaH, DMF, 80 °C (57%); (c) 10% HCl-MeOH, 70 °C; (d) Boc_2O, Et_3N, CH_2Cl_2, 0 °C (80% (2 steps)); (e) *N*-tosylimidazole, NaH, THF, rt (32%); (f) 4-hydroxyquinazoline, NaH, DMF, 80 °C; (g) Dess−Martin periodinane, CH_2Cl_2, rt (67% (2 steps)); (h) 10% HCl-MeOH, 70 °C (89%).

0.2 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with EtOAc to give the Boc-derivative of **7**.

To a solution of the Boc-derivative was added 10% HCl−MeOH (20 mL) at room temperature. After stirring for 2 h, the mixture was evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (4:1) to give **7** (73.5 mg, 48% from **6a**) as a dihydrochloride. Using a similar procedure, compounds **8** (yield: 42%) and **9** (40%) were prepared from **6b** and **6c**, respectively.

***tert*-Butyl (2*S**,3*S**)-2-allyl-3-hydroxypiperidine-1-carboxylate (10).** Compound **3** (4.39 g, 16.0 mmol) was dissolved in 6 M HCl (32 mL). After refluxing for 1.5 h. the solution was cooled to 0 °C. Then, 5 M NaOH (56 mL) and di-*tert*-butyl dicarbonate (6.96 g, 31.9 mmol) were added at room temperature. After stirring for 1.5 h, the mixture was poured into 0.1 M HCl, and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (2:1) to give **10** (2.36 g, 61%).

***tert*-Butyl (2*S**,3*S**)-3-methoxymethoxy-2-oxiranylmethylpiperidine-1-carboxylate (11).** To a solution of **10** (2.45 g, 10.1 mmol) in CH_2Cl_2 (40 mL) were added *N*,*N*-diisopropylethylamine (7.06 mL, 40.6 mmol) and chloromethyl methyl ether (3.85 mL, 50.7 mmol) at room temperature. After stirring for 7 h, the mixture was poured into 0.2 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated to give the crude MOM ether.

To a solution of this crude product in CH_2Cl_2 (50 mL) and water (50 mL) were added $NaHCO_3$ (1.89 g, 22.5 mmol) and *m*-chloroperbenzoic acid (75%) (3.45 g, 15.0 mmol) at 0 °C. After stirring for 48 h, the mixture was poured into 10% sodium thiosulfate solution and extracted three times with EtOAc. The organic layer was washed with 0.5 M NaOH and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (4:1) to give **11** (1.14 g, 44%).

3-[3-[(2*S,3*S**)-1-*tert*-Butyloxycarbonyl-3-methoxymethoxypiperidin-2-yl]-2-oxopropyl]-3*H*-benzo[*d*][1,2,3]triazin-4-one (12a).** To a solution of 3*H*-benzo[*d*][1,2,3]triazin-4-one (68.0 mg, 0.462 mmol) in DMF (4.0 mL) was added potassium hydride (30% oil suspension) (62.0 mg, 0.462 mmol) at 0 °C. After 30 min, compound **11** (116 mg, 0.385 mmol) dissolved in DMF (1.0 mL) was added to the solution. After stirring for 24 h at 80 °C, the mixture was poured into water and extracted three times with EtOAc. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (39:1) to give a mixture of 3-[3-[(2*S**,3*S**)-1-*tert*-butyloxycarbonyl-3-methoxymethoxypiperidin-2-yl]-2-hydroxypropyl]-3*H*-benzo[*d*][1,2,3]-triazin-4-one and 3*H*-benzo[*d*][1,2,3]triazin-4-one.

The mixture was dissolved in CH_2Cl_2 (2.0 mL). Then, 15% Dess−Martin periodinane solution in CH_2Cl_2 (1.50 mL, 0.535 mmol) was added to this solution at room temperature. After stirring for 3 h, the mixture was poured into 1 M NaOH solution and extracted three times with EtOAc. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (2:1) to give **12a** (122 mg, 71% (2 steps)). Using a similar procedure, compounds **12b** (yield: 31%) and **12c** (39%) were prepared from 3*H*-thieno[3,2-*d*]pyrimidin-4-one[18] and 4-hydroxyquinoline, respectively.

3-[(3a*S,7a*S**)-2-Hydroxyoctahydrofuro[3,2-*b*]pyridin-2-ylmethyl]-3*H*-benzo-[*d*][1,2,3]triazin-4-one (13a).** To a solution of the **12a** (92.1 mg, 0.206 mmol) was added 10% HCl−MeOH (3.0 mL) at room temperature. After stirring for 2 h, the mixture was evaporated. The residue was chromatographed over aluminum oxide eluted with $CHCl_3$−MeOH (4:1) to give **13a** (59.8 mg, 97%). Using a similar procedure, compounds **13b** (yield: 98%) and **13c** (95%) were prepared from **12b** and **12c**, respectively.

3-[3-[(2*R,3*S**)-3-Hydroxypiperidin-2-yl]-2-oxopropyl]-3*H*-benzo[*d*][1,2,3]-triazin-4-one (14).** Compound **6a** (45.1 mg, 0.149 mmol) was dissolved in MeOH (2.0 mL). After refluxing for 3 h, the reaction mixture was concentrated. To a solution of this residue in CH_2Cl_2 (3.0 mL) were added triethylamine (30 μL, 0.215 mmol) and di-*tert*-butyl dicarbonate (47.0 mg, 0.215 mmol) at room temperature. After stirring for 1.5 h, the mixture was poured into 0.2 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with EtOAc to give the Boc-derivative of **14**.

To a solution of the Boc-derivative was added 10% HCl−MeOH (2.0 mL) at room temperature. After stirring for 2 h, the mixture was evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (4:1) to give **14** (18.6 mg, 41% from **13a**) as a dihydrochloride. Using a similar procedure, compounds **15** (yield: 28%) and **16** (30%) were prepared from **13b** and **13c**, respectively.

(3*S*,4*R*)-6-(*tert*-Butyldimethylsilyloxy)-4-(dibenzylamino)-1-([1,3]-dioxan-2-yl)-hexan-3-ol (20a). Compound **19a** was prepared from D-aspartic acid (**17a**) according to the method reported by Gmeiner et al.[12] To a solution of oxalyl chloride (2.56 mL, 29.5 mmol) in CH_2Cl_2 (100 mL) were added DMSO (4.18 mL, 58.9 mmol) and **19a** (7.85 g, 19.6 mmol) dissolved in CH_2Cl_2 (20 mL) at −78 °C. After stirring for 1 h, triethylamine (13.7 mL, 98.2 mmol) was added to this mixture and stirred at 0 °C. The mixture was poured into 1 M HCl and extracted three times with Et_2O. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated to give the crude aldehyde of (2*R*)-4-(*tert*-butyldimethylsilyloxy)-2-(dibenzylamino)butanal. This crude aldehyde was dissolved in THF (35 mL). Then, 0.5 M (1,3-dioxan-2-ylethyl)magnesium bromide in THF (45.5 mL) was added to this solution at −50 °C. After stirring for 2 h, the mixture was allowed to warm up to room temperature and poured into a saturated NH_4Cl solution. This mixture was extracted three times with Et_2O. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (4:1) to give **20a** (5.61 g, 56% (2 steps)). Using a similar procedure, compound **20b** (yield: 59%) was prepared from **19b**.

***tert*-Butyl (2*R*,3*S*)-3-hydroxy-2-(2-hydroxyethyl)piperidine-1-carboxylate (22a).** Compound **20a** (5.61 g, 10.9 mmol) was dissolved in 2 M HCl (25 mL) and acetone (25 mL) at room temperature. After stirring for 4 h, the solution was poured into a 1 M NaOH solution and extracted three times with EtOAc. The organic layer was washed with a saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated to give the crude product of **21a**.

This crude product and 20% $Pd(OH)_2$ on carbon (760 mg) in MeOH (50 mL) were stirred at room temperature for 1 h under hydrogen atmosphere. After filtration, the filtrate was evaporated to give an oily residue. The residue was dissolved in MeOH (30 mL). Triethylamine (2.96 mL, 21.3 mmol) and di-*tert*-butyl dicarbonate (3.48 g, 16.0 mmol) were added to this solution at room temperature. After stirring for 1 h, the mixture was poured into 0.2 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with EtOAc to give **22a** (1.03 g, 66% (3 steps)). Using a similar procedure, compound **22b** (yield: 33%) was prepared from **20b**.

***tert*-Butyl (2*R*,3*S*)-2-(2-*tert*-butyldimethylsilyloxyethyl)-3-hydroxypiperidine-1-carboxylate (23a).** To a solution of **22a** (1.29 g, 5.23 mmol) in CH_2Cl_2 (15 mL) were added imidazole (712 mg, 10.5 mmol) and *tert*-butyldimethylsilyl chloride (800 mg, 5.31 mmol) at 0 °C. After stirring for 1 h, the mixture was poured into water and extracted three times with EtOAc. The organic layer was

washed with brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (4:1) to give **23a** (1.54 g, 82%). Using a similar procedure, compound **23b** (yield: 55%) was prepared from **22b**.

***tert*-Butyl (2*R*,3*S*)-2-(2-hydroxyethyl)-3-methoxymethoxypiperidine-1-carboxylate (24a).** To a solution of **23a** (1.54 g, 4.30 mmol) in 1,2-dimethoxyethane (15 mL) were added *N*,*N*-diisopropylethylamine (1.85 mL, 10.8 mmol) and chloromethyl methyl ether (470 μL, 6.45 mmol). After stirring for 15 h at 50 °C, the mixture was poured into 0.2 M HCl and extracted three times with EtOAc. The organic layer was washed with 0.5 M HCl and saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated to give the crude MOM ether.

This crude MOM ether was dissolved in THF (10 mL). Then, 1.0 M tetrabutylammonium fluoride solution in THF (6.40 mL) was added to the solution at room temperature. After stirring for 1 h, the mixture was poured into water and extracted three times with EtOAc. The organic layer was washed with brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (1:4) to give **24a** (1.03 g, 83%). Using a similar procedure, compound **24b** (yield: 94%) was prepared from **23b**.

***tert*-Butyl (2*R*,3*S*)-2-(3-ethoxycarbonyl-2-propenyl)-3-methoxymethoxy-piperidine-1-carboxylate (25).** To a solution of **24a** (425 mg, 1.47 mmol) in CH_2Cl_2 (5.0 mL) were added 15% Dess−Martin periodinane solution in CH_2Cl_2 (6.25 g, 2.21 mmol) and water (100 μL) at 0 °C. After stirring for 30 min, the mixture was poured into 1 M NaOH solution, and extracted three times with Et_2O. The organic layer was washed with brine, dried over anhydrous Na_2SO_4, and evaporated to give the crude aldehyde.

To a solution of triethyl phosphonoacetate (300 μL, 1.51 mmol) in toluene (4.0 mL) was added sodium hydride (60% oil suspension) (73 mg, 1.81 mmol) at 0 °C. After 15 min, the crude aldehyde in toluene (2.0 mL) was added to the mixture. After stirring for 1.5 h at room temperature, the mixture was poured into saturated a NH_4-Cl solution and extracted three times with EtOAc. The organic layer was washed with brine, dried, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (9:1) to give **25** (399 mg, 76% (2 steps)).

***tert*-Butyl (2*R*,3*S*)-2-(4-hydroxybutyl)-3-methoxymethoxypiperidine-1-carboxylate (24c).** Compound **25** (206 mg, 0.576 mmol) and 20% $Pd(OH)_2$ on carbon (40 mg) in THF (3.0 mL) were stirred at room temperature for 1 h under a hydrogen atmosphere. After filtration, the filtrate was evaporated to give an oily residue. To a solution of this residue in toluene (3.5 mL) was added a 1.0 M diisobutylaluminum hydride solution in toluene (1.8 mL) at 0 °C. After stirring for 2 h, a saturated Rochelle salt solution was added. After stirring for an additional 30 min, the mixture was poured into water and extracted three times with Et_2O. The organic layer was washed with brine, dried, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (2:1) to give **24c** (144 mg, 84% (2 steps)).

***tert*-Butyl (2*R*,3*S*)-3-methoxymethoxy-2-[2-(*p*-toluenesulfonyloxy)ethyl]-piperidine-1-carboxylate (26a).** To a solution of **24a** (172 mg, 0.594 mg) in pyridine (1.5 mL) was added *p*-toluenesulfonyl chloride (453 mg, 2.38 mmol) at room temperature. After stirring for 1.5 h, the mixture was poured into 0.2 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with EtOAc to give **26a** (183 mg, 69%). Using a similar procedure, compounds **26b** (yield: 80%) and **26c** (78%) were prepared from **24b** and **24c**, respectively.

3-[2-[(2*R*,3*S*)-1-*tert*-Butyloxycarbonyl-3-methoxymethoxypiperidin-2-yl]ethyl]-3*H*-quinazolin-4-one (27a). To a solution of 4-hydroxyquinazoline (61 mg, 0.414 mmol) in DMF (2.0 mL) was added K_2CO_3 (113 mg, 0.818 mmol) at 0 °C. After 15 min, compound **26a** (183 mg, 0.414 mmol) dissolved in DMF (1.0 mL) was added into the solution. After stirring for 4 h at 80 °C, the mixture was poured into water and extracted three times with EtOAc. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (2:1) to give **27a** (51 mg, 30%). Using a similar procedure, compounds **27b** (yield: 72%) and **27c** (84%) were prepared from **26b** and **26c**, respectively.

3-[2-[(2*R*,3*S*)-3-Hydroxypiperidin-2-yl]ethyl]-3*H*-quinazolin-4-one (28). To a solution of **27a** (42.1 mg, 0.100 mmol) was added 10% HCl−MeOH (1.5 mL) at room temperature. After stirring for 2 h, the mixture was evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (4:1) to give **28** (25.2 mg, 90%) as a dihydrochloride. Using a similar procedure, compounds **29** (yield: 88%) and **30** (93%) were prepared from **27b** and **27c**, respectively.

2-[(2*R*,3*S*)-1-*tert*-Butyloxycarbonyl-3-methoxymethoxypiperidin-2-yl]acetic acid (31). To a solution of **24a** (163 mg, 0.562 mmol) in CH_2Cl_2 (2.5 mL) were added 15% Dess−Martin periodinane solution in CH_2Cl_2 (2.4 mL, 0.843 mmol) and water (50 μL) at 0 °C. After stirring for 40 min, the mixture was poured into a 1 M NaOH solution and extracted three times with EtOAc. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and evaporated to give the crude aldehyde.

To a solution of the crude aldehyde in *tert*-butyl alcohol (3.5 mL) and water (1.1 mL) were added 2-methyl-2-butene (260 μL, 2.46 mmol), $NaH_2PO_4 \cdot 2H_2O$ (88.3 mg, 0.562 mmol), and $NaClO_2$ (80%) (203 mg, 2.25 mmol) at room temperature. After stirring for 40 min, the mixture was poured into water and extracted three times with EtOAc. The organic layer was dried over anhydrous Na_2SO_4 and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (2:1) to give **31** (142 mg, 83% (2 steps)).

2-[2-[(2*R*,3*S*)-1-*tert*-Butyloxycarbonyl-3-methoxymethoxypiperidin-2-yl]-acetylamino]-3*H*-quinazolin-4-one (32). To a solution of **31** (59 mg, 0.195 mmol) in CH_2Cl_2 (4.8 mL) and DMF (1.2 mL) were added 2-amino-4(*3H*)-quinazolinone[15] (35.0 mg, 0.215 mmol), *O*-(7-azabenzotriazol-1-yloxy)-*N*,*N*,*N*′,*N*′-tetramethyluronium hexafluorophosphate (222 mg, 0.584 mmol), and *N*,*N*-diisopropylethylamine (100 μL, 0.583 mmol) at room temperature. After stirring for 12 h, the mixture was poured into 0.3 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (49:1) to give **32** (33.1 mg, 38%).

2-[2-[(2*R*,3*S*)-3-Hydroxypiperidin-2-yl]acetylamino]-3*H*-quinazolin-4-one (33). To a solution of **32** (25.2 mg, 0.056 mmol) was added 10% HCl−MeOH (2.0 mL) at room temperature. After stirring for 1.5 h, the mixture was evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (3:1) to give **33** (10.3 mg, 45%) as a dihydrochloride.

(3*S*,4*R*)-4-Dibenzylamino-6-(*tert*-butyldimethylsilyloxy)hexan-3-ol (34). DMSO (2.10 mL, 29.6 mmol) and **19a** (3.94 g, 9.86 mmol) dissolved in CH_2Cl_2 (10 mL) were added to a solution of oxalyl chloride (1.30 mL, 14.8 mmol) in CH_2Cl_2 (40 mL) at −78 °C. After stirring for 1 h, triethylamine (6.90 mL, 49.3 mmol) was added to this mixture and stirred at room temperature. The mixture was poured into 0.5 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated to give the crude aldehyde of (*R*)-2-(dibenzylamino)-4-(*tert*-butyldimethylsilyloxy)butanal. The crude aldehyde was dissolved in THF (30 mL) and cooled into −78 °C. Ethylmagnesium bromide dissolved in THF (1.0 M, 14.8 mL) was added to this solution. After stirring for 1 h, the mixture was allowed to warm up to room temperature and poured into 0.5 M HCl. This mixture was extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (19:1) to give **34** (2.76 g, 66% (2 steps)).

(1*R*,2*S*)-*N*,*N*-Dibenzyl-1-[2-(*tert*-butyldimethylsilyloxy)ethyl]-2-methoxy-methoxybutylamine (35). To a solution of **34** (2.69 g, 6.29 mmol) in CH_2Cl_2 (30 mL) were added *N*,*N*-diisopropyl-

ethylamine (2.55 mL, 15.0 mmol) and chloromethyl methyl ether (1.50 mL, 20.0 mmol) at room temperature. After stirring for 5 h, the mixture was poured into water and extracted three times with EtOAc. The organic layer was washed with 0.5 M HCl, saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (39:1) to give **35** (2.34 g, 79%).

***tert*-Butyl (1*R*,2*S*)-1-[2-(*tert*-butyldimethylsilyloxy)ethyl]-2-methoxymethoxy-butylcarbamate (36).** Compound **35** (2.25 g, 4.78 mmol) and 20% $Pd(OH)_2$ on carbon (1.15 g) in ethanol (24 mL) were stirred at room temperature for 4 h under a hydrogen atmosphere. After filtration, the filtrate was evaporated to give an oily residue. The residue was dissolved in CH_2Cl_2 (25 mL). Triethylamine (900 μL, 6.46 mmol) and di-*tert*-butyl dicarbonate (1.36 g, 6.23 mmol) were added to this solution at 0 °C. After stirring for 1 h, the mixture was poured into 0.1 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (9:1) to give **36** (1.83 g, 98%).

***tert*-Butyl (1*R*,2*S*)-1-(2-hydroxyethyl)-2-methoxymethoxybutylcarbamate (37).** To a solution of **36** (1.53 g, 5.51 mmol) in THF (15 mL) was added 1.0 M tetrabutylammonium fluoride solution in THF (6.60 mL) at room temperature. After stirring for 1 h, the mixture was poured into water and extracted three times with EtOAc. The organic layer was washed with brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (4:1) to give **37** (1.02 g, 67%).

***tert*-Butyl (1*R*,2*S*)-2-methoxymethoxy-1-oxiranylmethylbutylcarbamate (38).** To a solution of **37** (467 mg, 1.69 mmol) in CH_2Cl_2 (5.0 mL) was added Dess−Martin periodinane (855 mg, 2.02 mmol) at 0 °C. After stirring for 1 h, the mixture was poured into a 0.5 M NaOH solution, and extracted three times with EtOAc. The organic layer was washed with brine, dried over anhydrous Na_2SO_4, and evaporated to give the crude aldehyde.

To a solution of sodium hydride (60% oil suspension) (75.0 mg, 1.88 mmol) in DMSO (3.0 mL) was added trimethylsulfoxonium iodide (410 mg, 1.86 mmol) at room temperature. After stirring for 1 h at 70 °C, the crude aldehyde in DMSO (3.0 mL) was added to the mixture at room temperature. After stirring for 1.5 h at 50 °C, the mixture was poured into water and extracted three times with EtOAc. The organic layer was washed with brine, dried, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (4:1) to give **38** (263 mg, 75% (2 steps)).

***tert*-Butyl[(1*R*,2*S*)-2-methoxymethoxy-1-oxiranylmethylbutyl]-methylcarbamate (39).** To a solution of **38** (394 mg, 1.36 mmol) in DMF (7 mL) were added sodium hydride (60% oil suspension) (65.0 mg, 1.63 mmol) and CH_3I (160 μL, 2.57 mmol) at 0 °C. After stirring for 3 h, the mixture was poured into water and extracted three times with EtOAc. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (4:1) to give **39** (374 mg, 91%).

(4*R*,5*S*)-3-[5-Methoxymethoxy-4-(*N*-*tert*-butyloxycarbonyl-*N*-methylamino)-2-oxoheptyl]-3*H*-quinazolin-4-one (40). To a solution of 4-hydroxyquinazoline (201 mg, 1.38 mmol) in DMF (3 mL) was added sodium hydride (60% oil suspension) (55.1 mg, 1.38 mmol) at 0 °C. After 30 min, compound **39** (348 mg, 1.15 mmol) dissolved in DMF (1 mL) was added to the solution. After stirring for 20 h at 80 °C, the mixture was poured into water and extracted three times with EtOAc. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (1:1) to give a mixutre of (4*R*,5*S*)-3-[2-hydroxy-5-(methoxymethoxy)-4-(*N*-*tert*-butyloxycarbonyl-*N*-methylamino)heptyl]-3*H*-quinazolin-4-one and 4-hydroxy-quinazoline.

The mixture was dissolved in CH_2Cl_2 (2 mL). Then, 15% Dess−Martin periodinane solution in CH_2Cl_2 (3.90 g, 1.38 mmol) was added to this solution at room temperature. After stirring for 2 h, the mixture was poured into 1 M NaOH solution and extracted three times with EtOAc. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (39:1) to give **40** (272 mg, 41% (2 steps)).

(4*R*,5*S*)-3-(5-Hydroxy-4-methylamino-2-oxoheptyl)-3*H*-quinazolin-4-one (41). To a solution of **50** (39.0 mg, 0.097 mmol) in CH_2Cl_2 (1.5 mL) was added trifluoroacetic acid (1.5 mL) at 0 °C. After stirring for 1 h, the solution was evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (9:1) to give **41** (22.2 mg, 44%) as a ditrifluoroacetate.

3-(Triisopropylsilyloxy)piperidine (43). To a solution of 3-hydroxypiperidine (**42**, 1.01 g, 9.99 mmol) in CH_2Cl_2 (20 mL) were added triethylamine (1.53 mL, 10.9 mmol) and benzyloxycarbonyl chloride (1.56 mL, 10.9 mmol) at 0 °C. After stirring for 30 min, the mixture was poured into 0.2 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (2:1) to give *N*-benzyloxycarbonyl-3-hydroxypiperidine (2.25 g, 96%).

N-Benzyloxycarbonyl-3-hydroxypiperidine (561 mg, 2.39 mmol) was dissolved in DMF (6.0 mL). Imidazole (287 mg, 4.21 mmol) and triisopropylsilyl chloride (900 μL, 4.21 mmol) were added to this solution at room temperature. After stirring for 4 h, the mixture was poured into 0.2 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated.

The residue was dissolved in MeOH (6 mL), and 20% $Pd(OH)_2$ on carbon (119 mg) was added to the solution. After stirring for 4 h under a H_2 atmosphere, the reaction mixture was filtered through a Celite pad, and the filter cake was washed with MeOH. The filtrate was evaporated, and the residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (4:1) to give **43** (433 mg, 71% (2 steps)).

1-Oxiranylmethyl-3-(triisopropylsilyloxy)piperidine (44). To a solution of **43** (396 mg, 1.54 mmol) in DMF (4.0 mL) were added epibromohydrin (160 μL, 1.87 mmol) and K_2CO_3 (255 mg, 1.85 mmol) at room temperature. After stirring for 5 h, the mixture was poured into water and extracted three times with EtOAc. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (9:1) to give **44** (344 mg, 71%).

3-{2-Hydroxy-3-[3-(triisopropylsilyloxy)piperidin-1-yl]propyl}-3*H*-quinazolin-4-one (45). To a solution of 4-hydroxyquinazoline (157 mg, 1.07 mmol) in DMF (4 mL) was added sodium hydride (60% oil suspension) (43.0 mg, 1.07 mmol) at room temperature. After 15 min, compound **44** (280 mg, 0.892 mmol) dissolved in DMF (0.5 mL) was added to the solution. After stirring for 18 h at 80 °C, the mixture was poured into water and extracted three times with EtOAc. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (1:1) to give **45** (277 mg, 68%).

3-[3-(3-Hydroxypiperidin-1-yl)-2-oxopropyl]-3*H*-quinazolin-4-one (46). To a solution of oxalyl chloride (60 μL, 0.69 mmol) in CH_2Cl_2 (1.5 mL) was added DMSO (90 μL, 1.3 mmol) at −78 °C. After 20 min, compound **45** (67.2 mg, 0.15 mmol) in CH_2Cl_2 (0.5 mL) was added to the solution. After 40 min, triethylamine (300 μL, 2.15 mmol) was added to the solution. The solution was stirred, warmed to room temperature, poured into water, and extracted three times with EtOAc. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (99:1) to give 3-{3-[3-(triisopropylsilyloxy)piperidin-1-yl]-2-oxopropyl}-3*H*-quinazolin-4-one (48 mg, 72%).

This ketone (11.1 mg, 0.024 mmol) was dissolved in 5% HCl−MeOH (1.0 mL). After stirring for 2 h at 50 °C, the solution was evaporated. The residue was chromatographed over silica gel eluted

with $CHCl_3$−MeOH (49:1) to give **46** (5.8 mg, 80%) as a dihydrochloride.

1-*tert*-Butyloxycarbonyl-3-(2,2-dimethyl[1,3]dioxolan-4-yl-methoxy)piperidine (47). To a solution of 3-hydroxypiperidine (**42**, 1.01 g, 9.95 mmol) in CH_2Cl_2 (20 mL) were added triethylamine (1.66 mL, 11.9 mmol) and di-*tert*-butyl dicarbonate (2.63 g, 12.1 mmol) at 0 °C. After stirring for 1 h, the mixture was poured into 0.1 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (4:1) to give 1-*tert*-butyloxycarbonyl-3-hydroxypiperidine (1.90 g, 95%).

To a solution of 1-*tert*-butyloxycarbonyl-3-hydroxypiperidine (998 mg, 4.96 mmol) in DMF (16 mL) were added sodium hydride (60% oil suspension) (760 mg, 19.0 mmol) and 2,2-dimethyl-1,3-dioxolan-4-ylmethyl tosylate (3.13 g, 10.9 mmol). After stirring for 16 h at 90 °C, the mixture was poured into 0.2 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (9:1) to give **47** (891 mg, 57%).

1-*tert*-Butyloxycarbonyl-3-oxiranylmethoxypiperidine (48). Compound **47** (812 mg, 2.58 mmol) was dissolved in 10% HCl−MeOH (15 mL). After refluxing for 3 h, the solution was evaporated. The residue was dissolved in CH_2Cl_2 (12 mL) and MeOH (3.0 mL). Triethylamine (1.82 mL, 13.1 mmol), and di-*tert*-butyl dicarbonate (683 mg, 3.13 mmol) was added to this solution at 0 °C. After stirring for 2 h, the mixture was poured into 0.5 M HCl and extracted three times with EtOAc. The organic layer was washed with saturated $NaHCO_3$ solution and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (99:1) to give 1-*tert*-butyloxycarbonyl-3-(2,3-dihydroxypropoxy)piperidine (565 mg, 80%).

To a solution of 1-*tert*-butyloxycarbonyl-3-(2,3-dihydroxypropoxy)piperidine (534 mg, 1.94 mmol) in DMF (15 mL) were added sodium hydride (60% oil suspension) (194 mg, 4.85 mmol) and 1-(*p*-toluenesulfonyl)imidazole (603 mg, 2.71 mmol) at 0 °C. After stirring for 5 h at room temperature, the mixture was poured into 0.5 M HCl and extracted three times with EtOAc. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (4:1) to give **48** (159 mg, 32%).

3-[3-(1-*tert*-Butyloxycarbonylpiperidin-3-yloxy)-2-oxopropyl]-3*H*-quinazolin-4-one (49). To a solution of 4-hydroxyquinazoline (89.3 mg, 0.611 mmol) in DMF (4.0 mL) was added sodium hydride (60% oil suspension) (24.6 mg, 0.615 mmol) at room temperature. After 25 min, compound **48** (132 mg, 0.513 mmol) dissolved in DMF (2.0 mL) was added to the solution. After stirring for 10 h at 80 °C, the mixture was poured into water and extracted three times with EtOAc. The organic layer was washed with water and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (2:1) to give a mixutre of 3-[3-(1-*tert*-butyloxycarbonylpiperidin-3-yloxy)-2-hydroxypropyl]-3*H*-quinazolin-4-one and 4-hydroxyquinazoline.

The mixture was dissolved in CH_2Cl_2 (4.0 mL). Then, 15% Dess−Martin periodinane solution in CH_2Cl_2 (5.80 g, 2.05 mmol) was added to this solution at room temperature. After stirring for 3 h, the mixture was poured into 10% sodium thiosulfate solution and extracted three times with EtOAc. The organic layer was washed with 0.5 M NaOH, water, and brine, dried over anhydrous Na_2SO_4, and evaporated. The residue was chromatographed over silica gel eluted with hexanes−EtOAc (1:1) to give **49** (137 mg, 67% (2 steps)).

3-[2-Oxo-3-(piperidin-3-yloxy)propyl]-3*H*-quinazolin-4-one (50). Compound **49** (18.8 mg, 0.047 mmol) was dissolved in 5% HCl−MeOH (2.0 mL). After stirring for 1 h at room temperature, the solution was evaporated. The residue was chromatographed over silica gel eluted with $CHCl_3$−MeOH (4:1) to give **50** (15.6 mg, 89%) as dihydrochloride.

Antimalarial Assay in Vitro. Type A^+ human plasma and erythrocytes were obtained from healthy volunteers at our institute. *Plasmodium falciparum* strains FCR-3 (chloroquine sensitive) and K1 (chloroquine resistant) were cultured in the human erythrocytes in an RPMI medium (RPMI-1640 with 25 mM HEPES buffer, 24 mM $NaHCO_3$, 0.2% glucose, 0.05% L-glutamine, 50 μg/mL of hypoxanthine, and 25 μg/mL of gentamisin) supplemented with 10% human plasma at 37 °C, under 93% N_2, 4% CO_2, and 3% O_2. The antimalarial activity of the test compounds have been achieved by a dose−response curve using the parasite lactate dehydrogenase (pLDH) assay.[19] Asynchronous parasites (2.0% hematocrit and 0.5% parasitaemia) (90 μL) were seeded in a 96-well microplate, and a solution of the test compound dissolved in distilled water or 5% DMSO (10 μL) was added. After incubation at 37 °C for 72 h, the parasite suspension (20 μL) was transferred to another plate containing Malstat reagent (100 μL). The plate was further incubated for 15 min at room temperature, and a 1:1 mixture of nitroblue tetrazolium and phenazine ethosulfate (2 mg and 0.1 mg/mL, respectively) (20 μL) was added to each well. After incubation for 2 h at room temperature in darkness, the blue formazan product was measured at 650 nm. The EC_{50} value was estimated from a dose−response curve.

Cytotoxicity Test. The cytotoxicity test of compounds was measured by the colorimetric MTT assay.[20] Mouse L929 cell suspension in RPMI-1640 with 10% FBS (90 μL) was added in 96-well microplates at 1.8×10^4 cells/well. Then, a solution of the test compound dissolved in distilled water or 10% DMSO (10 μL) was added to each well. After incubation for 48 h at 37 °C under 5% CO_2, 2.5 mg/mL of MTT solution (10 μL) was added to each well. The plate was incubated for a further 4 h. Then, the incubation medium was aspirated, and DMSO (100 μL) was added to solubilize the MTT formazan product. After mixing, the absorbances at 570 and 655 nm were measured. The EC_{50} value was estimated from a dose−response curve.

Antimalarial Assay in Vivo. The in vivo antimalarial activity was determined against rodent malaria-derived *P.berghei* NK65 according to the 4-day suppressive test.[21] Male ICR mice at weight 18−20 g were intravenously inoculated with 10^6 parasitized red blood cells. Test compounds were suspended in 5% gum arabic and orally administered to the mice 2 h after infection (Day 0). The test compounds were successively administered to the mice once a day for 3 consecutive days (Days 1−3). The day after the last treatment (Day 4), blood was obtained from the tail vein of the infected mice. Parasitaemia was determined by thin blood films made from the blood samples. The ED_{50} value was estimated from a dose−response curve.

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Supporting Information Available: Analytical data of new compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

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