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Synthesis, Structure–Activity Relationship, and Pharmacological Studies of Novel Melanin-Concentrating Hormone Receptor 1 Antagonists 3-Aminomethylquinolines: Reducing Human Ether-a-go-go-Related Gene (hERG) Associated Liabilities

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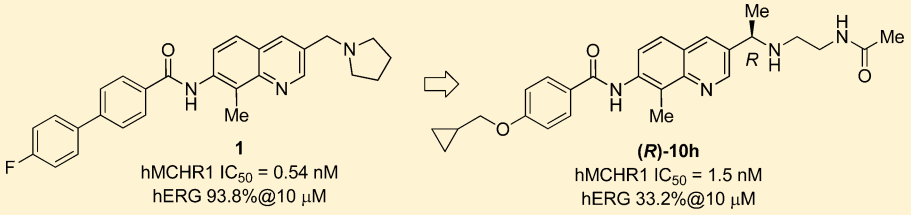
Supporting Information



ABSTRACT: Recently, we discovered 3-aminomethylquinoline derivative **1**, a selective, highly potent, centrally acting, and orally bioavailable human MCH receptor 1 (hMCHR1) antagonist, that inhibited food intake in F344 rats with diet-induced obesity (DIO). Subsequent investigation of **1** was discontinued because **1** showed potent hERG K^+ channel inhibition in a patch-clamp study. To decrease hERG K^+ channel inhibition, experiments with ligand-based drug designs based on **1** and a docking study were conducted. Replacement of the terminal *p*-fluorophenyl group with a cyclopropylmethoxy group, methyl group introduction on the benzylic carbon at the 3-position of the quinoline core, and employment of a [2-(acetylamino)ethyl]amino group as the amine portion eliminated hERG K^+ channel inhibitory activity in a patch-clamp study, leading to the discovery of *N*-{3-[(1*R*)-1-{[2-(acetylamino)ethyl]amino}ethyl]-8-methylquinolin-7-yl}-4-(cyclopropylmethoxy)benzamide (***R*)-10h**. The compound (***R*)-10h** showed potent inhibitory activity against hMCHR1 and dose-dependently suppressed food intake in a 2-day study on DIO-F344 rats. Furthermore, practical chiral synthesis of (***R*)-10h** was performed to determine the molecule's absolute configuration.

INTRODUCTION

The World Health Organization has declared obesity a worldwide epidemic; more than 1 billion adults are overweight, and at least 300 million are obese.[1] The healthcare burden for treating obesity is significantly high because obesity causes secondary chronic diseases such as type 2 diabetes, hypertension, stroke, cardiovascular disease, and certain forms of cancer. Although many molecular targets have been studied, antiobesity drugs remain an unmet medical need.

Melanin-concentrating hormone (MCH) is a disulfide-linked cyclic nonadecapeptide expressed in the lateral hypothalamus, and it is the natural ligand for seven-transmembrane G-protein-coupled receptors MCHR1 and MCHR2.[2−4]

MCHR1 is primarily expressed in the central nervous system in mammals, but MCHR2 is not expressed in rats and mice; thus, little is known about its physiological function.

MCH acutely stimulates feeding behavior in rats when injected intracerebroventricularly,[5] and MCH knockout mice show reduced body weight due to hypophagia and an increase in metabolic rate.[6] Transgenic mice overexpressing the prepro-MCH gene exhibit hyperphagia, mild obesity, and insulin resistance.[7] Moreover, MCHR1-deficient (*MCH1r*−/−) mice are lean (similar to prepro-MCH-deficient mice) but are hyperphagic when maintained on regular chow. *MCH1r*−/− mice are less susceptible to diet-induced obesity because of their hyperactivity and increased energy expenditure.[8a,b] MCHR1 antagonism suppresses food intake and fat accumulation,[9] indicating that MCHR1 is an essential receptor for regulating body weight and composition. These findings suggest that MCHR1 antagonists may provide a new class of antiobesity agents that act by inducing an anorexigenic effect and enhanced energy expenditure.[10a−c]

Small MCHR1 antagonists have been found to be promising agents for treating obesity. Although many pharmaceutical and biotechnology companies have been heavily examining small

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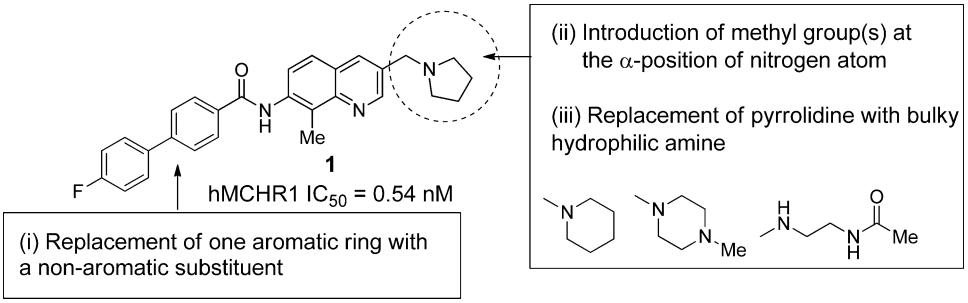
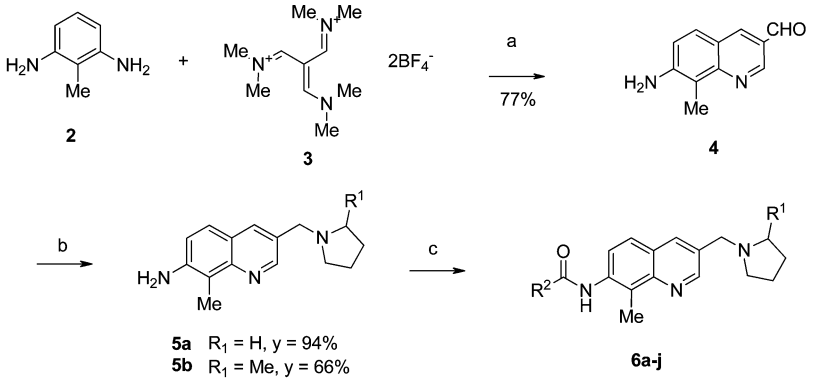
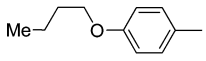
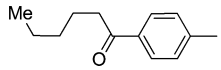
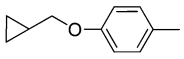
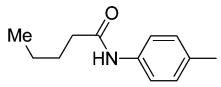
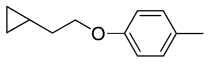
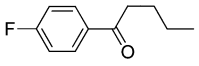
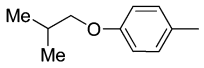
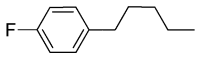
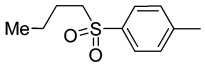
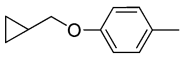


Figure 1. Structure of MCHR1 antagonist **1** and synthetic strategy.

Scheme 1[a]



Cpd	R^1	R^2	yield (%)	Cpd	R^1	R^2	yield (%)
6a	H		20	**6f**	H		36
6b	H		50	**6g**	H		16
6c	H		58	**6h**	H		10
6d	H		51	**6i**	H		12
6e	H		36	**6j**	Me		72

[a]Reagents and conditions: (a) (i) ^{i}PrOH, reflux, (ii) 1 M HCl, 70 °C, (iii) K_2CO_3, AcOEt−H_2O, 90 °C; (b) pyrrolidine, $NaBH(OAc)_3$, dichloroethane, rt; (c) (i) R^2COOH, oxalyl chloride, DMF (cat.), THF, rt, (ii) triethylamine, THF, rt.

molecule MCHR1 antagonists for more than 10 years,[11a−c] few compounds have been evaluated in clinical trials, and none of these compounds are available commercially.[12a−c] A major hurdle in the use of MCHR1 antagonists in clinical settings includes human ether-a-go-go-related gene (hERG) associated cardiovascular risks.[13] Despite attractive in vivo profiles, further development of many MCHR1 antagonists has been discontinued because of significant hERG K^+ channel binding, which can induce QTc interval prolongation frequently associated with potential lethal arrhythmias known as torsades de pointes. The hERG blockade of MCHR1 antagonists can be explained by two common structural elements between a significant number of reported MCHR1 antagonists and well-known hERG-binding agents: a positively charged group and at least one distal aromatic/lipophilic region.[13]

In previous reports, we described efforts undertaken in the field of medicinal chemistry to improve upon a lead compound[14a,b] and to study its relationship with MCHR1 antagonistic activity, pharmacokinetic properties, and selectivity over serotonin 5-HT_{2c} receptor, which culminated in the discovery of 3-aminomethylquinoline-based compound **1**.[15] Although compound **1** displayed excellent in vitro and in vivo activities and selectivity over other enzymes and receptors, including the $5HT_{2c}$ receptor,[15] **1** was identified as a potent hERG K^+ channel blocker in an in vitro patch-clamp study. Herein, we report the structure−activity relationship (SAR) of 3-aminomethylquinoline-based MCHR1 antagonists and de-

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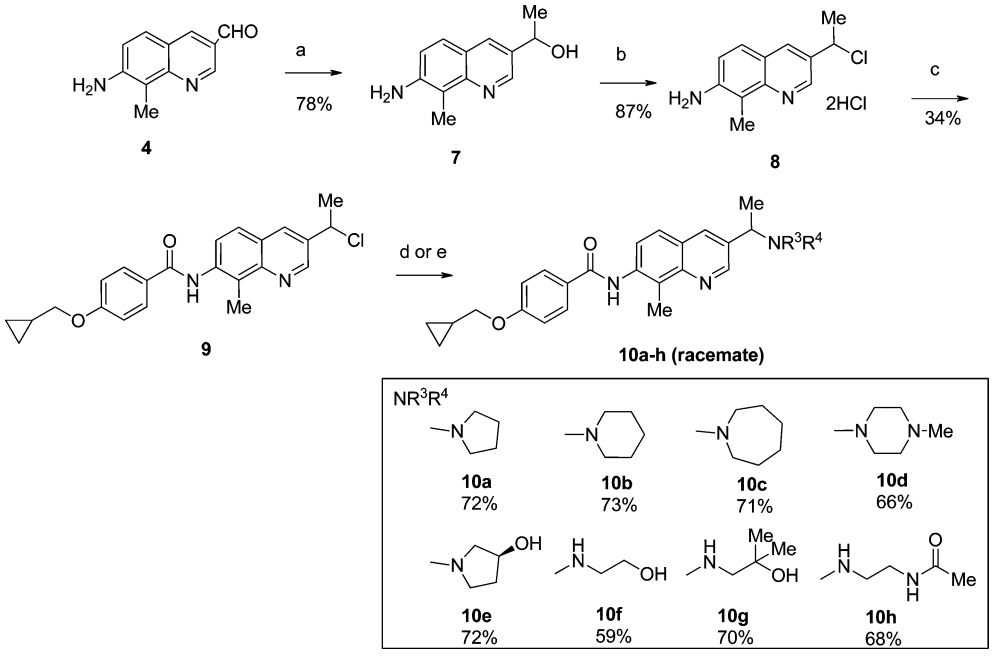
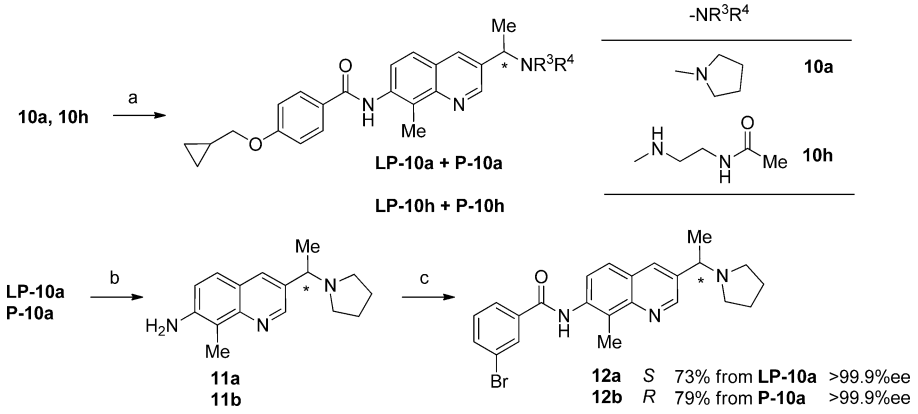
Scheme 2[a]



[a]Reagents and conditions: (a) MeMgBr, THF, 0 °C to rt; (b) $SOCl_2$, rt; (c) 4-cyclopropylmethoxybenzoyl chloride, triethylamine, THF, 0 °C to rt; (d) amine, K_2CO_3, KI, DMF, 70 °C; (e) amine, DIPEA, NMP, 60 °C.

Scheme 3[a]



[a]Reagents and conditions: (a) chiral column separation by HPLC; (b) conc HCl, 100 °C; (c) 3-bromobenzoyl chloride, triethylamine, THF, 0 °C to rt.

scribe pharmacological studies performed to address the issue of hERG K^+ channel blocking.

DRUG DESIGN

Several reports[16a,b,17,18] indicated that some aromatic amino acid residues constructing the hERG K^+ channel, such as Tyr652 and Phe656, play a key role in creating a π-related or hydrophobic interaction[19,20] with various types of drugs that block hERG K^+ channel with high affinity. An effective method of reducing the hERG K^+ channel binding affinity may be to reduce lipophilicity and/or introduce hydrophilic substituents. Indeed, there are a number of reports that describe decreasing hERG liabilities by lowering the lipophilicity of the compound.[21] On the basis of these findings, we adopted the following strategies (Figure 1): (i) replacement of one aromatic ring in the biphenyl group with a nonaromatic substituent aimed to reduce the lipophilic and/or $\pi-\pi$ interaction, (ii) introduction of methyl group(s) at the α-position of the basic nitrogen atom to prevent the cation$-\pi$ and/or CH$-\pi$ interaction by steric hindrance, and (iii) replacement of pyrrolidine with bulky and/or a hydrophilic amine to reduce the cation$-\pi$ or CH$-\pi$ interaction. For (ii), we also tested ethyl, isopropyl, cyclopropyl, phenyl, and benzyl groups instead of a methyl group. However, these replacements resulted in a loss of MCH antagonistic activity (data not shown). Furthermore, to support these strategies, we carried out a docking study of a key compound with the hERG K^+ channel.[22]

CHEMISTRY

First, we replaced one of the two benzene rings at the terminal biphenyl moiety of **1**. Scheme 1 depicts the synthesis of the key intermediate 7-amino-3-formyl-8-methylquinoline **4** and proposed quinoline derivatives **6a**–**j**. Condensation and cyclization of commercially available 2,6-diaminotoluene **2** with "vinami-

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dinium" bis-tetrafluoroborate salt **3** followed by a hydrolysis reaction yielded the 3-formylquinoline derivative **4**.[23] Subsequent reductive amination of **4** with pyrrolidine or 2-methylpyrrolidine provided 3-pyrrolidinylmethylquinoline derivatives **5a** and **5b**, respectively. A condensation reaction of **5a** or **5b** with benzoic acids gave the desired amide compounds **6a**–**j**.

Scheme 2 describes the synthesis of 1-(8-methylquinolin-3-yl)ethanamine derivatives **10a**–**h** (racemate). Compound **4** was reacted with methylmagnesium bromide to give alcohol **7**, which was chlorinated to provide the key intermediate 3-(1-chloroethyl)-8-methylquinolin-7-amine (**8**). Compound **8** was coupled with 4-(cyclopropylmethoxy)benzoyl chloride prepared from the corresponding acid to provide amide **9**. Finally, **9** was reacted with amines to yield proposed racemic compounds **10a**–**h**.

Scheme 3 shows the chiral separation of **10a** and **10h** and the determination of the absolute configuration. Optical resolution of **10a** by chiral HPLC provided the less polar compound **LP-10a** and the polar compound **P-10a**, respectively. **LP-10a** or **P-10a** was hydrolyzed to afford 7-aminoquinoline derivative **11a** or **11b**, which was subsequently converted to 3-bromobenzoate **12a** or **12b**, respectively, without racemization. Since the structure of **12a**, which was derived from **LP-10a**, was determined using X-ray crystallographic analysis to be an *S*-configuration (Figure 2), the structure of **P-10a** was

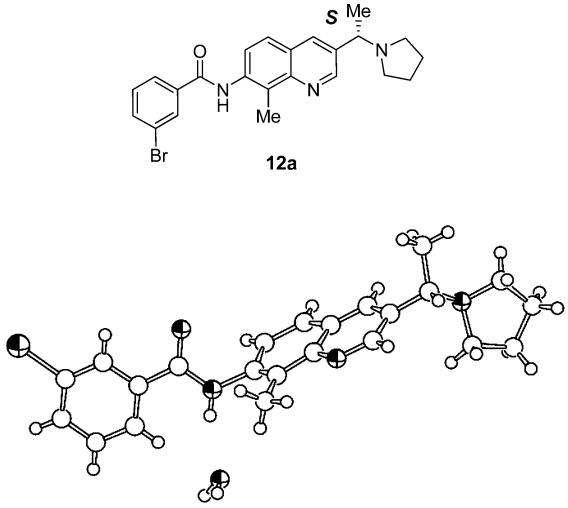

Figure 2. Structure of **12a**.

determined to be an *R*-configuration. Optical resolution of **10h** using chiral HPLC provided a less polar compound **LP-10h** and a more polar compound **P-10h**, and the absolute configuration of **LP-10h** was determined to be *R* using the chiral synthesis described in Scheme 4.

Compound **4** was coupled with 4-cyclopropylmethoxybenzoic acid to give amide **13**, and a subsequent Grignard reaction afforded racemic alcohol (***rac***)**-14**. Oxidation of (***rac***)**-14** with manganese(IV) oxide afforded ketone **15**. To achieve the asymmetric synthesis of (***R***)**-10h**, we designed the synthetic route via Noyori asymmetric hydrogenation[24a–e] of ketone **15** followed by inversion to optically active azide **16** under Mitsunobu conditions reported by Thompson et al.[25] Ohkuma et al. reported a practical method for asymmetric hydrogenation of heteroaromatic ketones, in which [$RuCl_2$\{(*R*)-xylbinap\}\{(*R*)-daipen\}] provides *S*-chiral alcohols.[24e] Ketone **15** was hydrogenated in the presence of [$RuCl_2$\{(*R*)-xylbinap\}\{(*R*)-daipen\}] (substrate-to-catalyst molar ratio (*S*/*C*) of 1000) under a 0.7 MPa hydrogen atmosphere in DMF and 2-propanol solution to produce the prerequisite chiral alcohol (***S***)**-14** with 98% ee. In the next step, we examined Thompson's conditions (diphenylphosphorylazide (DPPA), DBU, in toluene or in THF, at −30 °C or at 0 °C to room temperature). Although these reaction conditions provided the corresponding azide **16** in moderate to good yield (43–76%), the enantiomeric excess was insufficient (<90% ee, data not shown). We focused on the optimization of the reaction conditions, and finally the conversion of the chiral alcohol (***S***)**-14** (98% ee) to the corresponding chiral azide **16** was successfully achieved (90%, 98% ee) using DPPA under an excess of DIPEA as a solvent.

Catalytic hydrogenation of azide **16** quantitatively yielded chiral primary amine **17** with 98% ee. We confirmed the absolute configuration of **17** by synthesis of (***R***)**-10a**: the reaction of **17** with 1,4-dibromobutane provided solely (***R***)**-10a** in 61%. These results suggest that conversion of **15** to (***S***)**-14** and inversion of (***S***)**-14** to **16** were successfully achieved without racemization.

Nosylation of the amino group of **17** gave **18** in excellent yield. The Mitsunobu reaction with *N*-Boc-protected aminoethanol followed by acid-catalyzed removal of the Boc protecting group was achieved in a one-pot reaction. Primary amine **19** was purified by acid–basic extraction without column chromatography. Reaction with acetyl chloride followed by deprotection of the nosyl group afforded the desired compound (***R***)**-10h** in high yield. The overall yield of (***R***)**-10h** from ketone **15** was 82% without racemization. Chiral HPLC analysis showed that **LP-10h** is identical to (***R***)**-10h**; the absolute stereochemistry of the eutomer of compound **10h** was determined to be an *R*-configuration.

Scheme 5 shows the synthesis of 3-(1-methyl-1-pyrrolidin-1-ylethyl)quinoline derivative **22**, which bears two methyl groups on the benzylic carbon. The formyl group of compound **13** was converted to methyl ester using *N*-iodosuccinimide (NIS) in the presence of K_2CO_3 to give **20**.[26] A reaction of the methyl ester of **20** with excess methyllithium yielded tertiary alcohol **21**. Finally, compound **21** was chlorinated and successively coupled with pyrrolidine to afford the desired dimethyl compound **22**.

RESULTS AND DISCUSSION

Compounds synthesized in this study were tested for their binding affinities to the human MCH receptor 1 (hMCHR1) and rat MCH receptor 1 (rMCHR1) by using a stably transfected Chinese hamster ovary (CHO) cell line. Binding assays of the test compounds were performed in the presence of [^{125}I]MCH (4–19) as a ligand. Secondary functional cell-based assays for the inhibition of MCH-stimulated arachidonic acid release from CHO cells were also performed, and the test compounds were found to be antagonists. In our previous study for a MCHR1 antagonist,[14b] we confirmed that the acachidonic acid release inhibition represented the MCHR1 antagonistic activity. The MCHR1 antagonist showed the binding affinity for MCHR1, reversed the MCH-mediated inhibition of intracellular cAMP accumulation, inhibited the MCH-induced intracellular Ca^{2+} increase, and inhibited the MCH-induced arachidonic acid release. Furthermore, we performed Ca^{2+} influx assays for the key two compounds ((***R***)**-10a** and (***R***)**-10h**) and confirmed their antagonistic activities against hMCHR1 (Supporting Information Figure S1).

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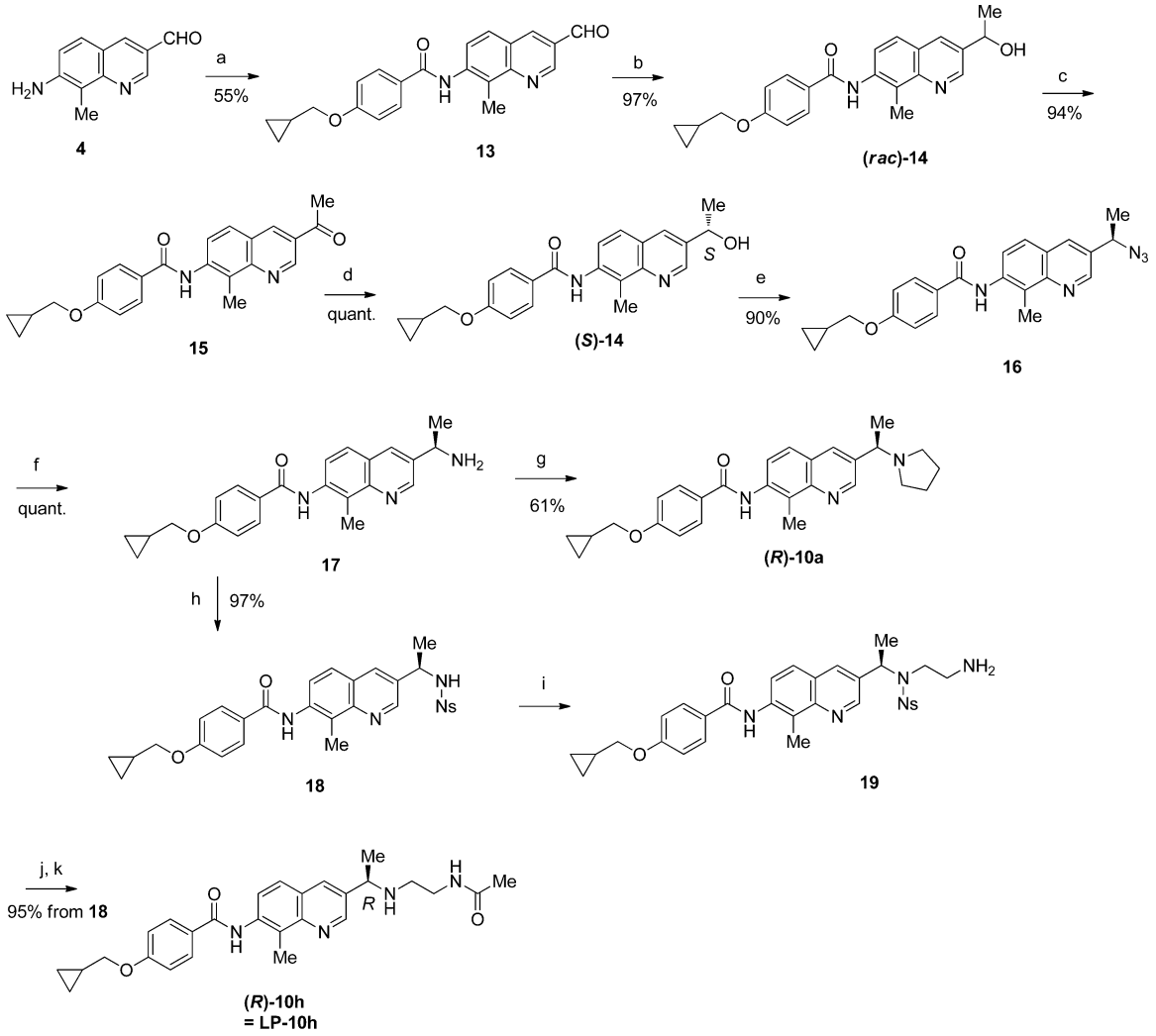
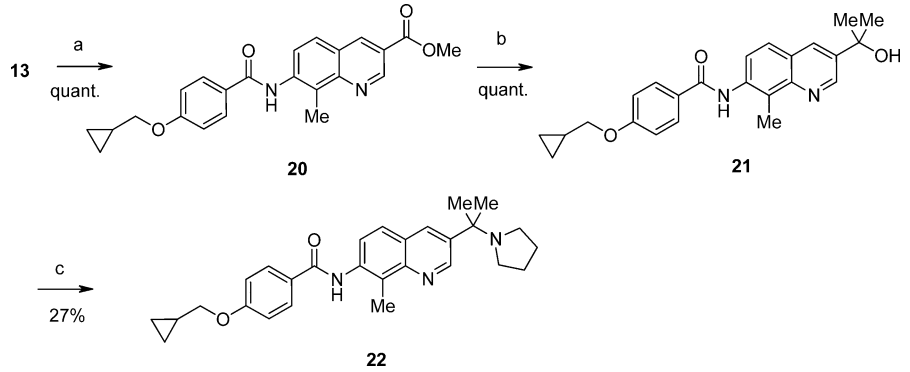
Scheme 4[a]



[a]Reagents and conditions: (a) 4-cyclopropylmethoxybenzoyl chloride, pyridine, 0 °C to rt; (b) MeMgBr, THF, 0 °C to rt, 4 h; (c) MnO_2, THF, reflux, 24 h; (d) KO^{t}Bu, DMF−2-propanol, $RuCl_2$\{(*R*)-xylbinap\}\{(*R*)-daipen\} (*S*/*C* = 1000), then H_2 (0.7 MPa), rt, 24 h, quant, 98% ee; (e) DPPA, DIPEA, rt, 96 h, 98% ee; (f) H_2 (balloon), Pd/C, EtOH, rt, 2 h, 98% ee; (g) 1,4-dibromobutane, Na_2CO_3, DMF, 60 °C, 99% ee; (h) NsCl, triethylamine, THF, 0 °C to rt, 4 h; (i) (i) $HO(CH_2)_2NHBoc$, DIAD, PPh_3, THF, 0 °C to rt, 2.5 h, (ii) HCl−AcOEt, rt, 15 h, one-pot reaction; (j) AcCl, triethylamine, THF, 0 °C, 10 min; (k) $HSCH_2CO_2H$, LiOH·H_2O, DMF, rt, 4 h, 99% ee.

Scheme 5[a]



[a]Reagents and conditions: (a) NIS, K_2CO_3, MeOH, rt; (b) MeLi, THF, 0 °C to rt; (c) (1) $SOCl_2$, 0 °C, (2) pyrrolidine, 60 °C.

The test compounds were evaluated for their inhibitory activities against the hERG K^+ channel by using a high throughput nonradioactive rubidium (Rb^+) efflux assay (Rb assay).[27] The Rb assay was performed using CHO cells expressing the hERG K^+ channel in which astemizole was used as a positive control. The results are expressed as relative

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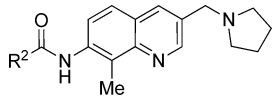
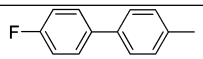
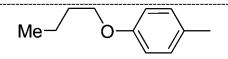
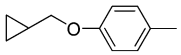
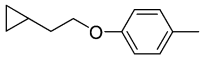
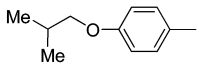
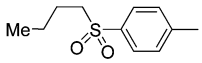
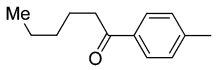
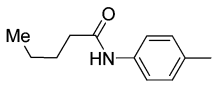
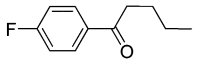
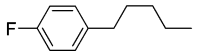
Table 1. In Vitro Pharmacological and Physicochemical Data for MCHR1 Antagonists



Cpd	R^2	IC_{50} (nM)[a] hMCHR1[b]	rMCHR1[c]	AA release[d]	Rb value @ 30 μM[e]	logD @ pH 7.4[f]
1		0.54	1.0	2.8	0.17	3.19
6a		2.0	1.2	2.6	0.41	3.36
6b		1.8	1.6	4.3	0.90	2.77
6c		1.3	2.0	2.0	0.72	3.40
6d		4.4	6.7	NT[g]	0.70	3.35
6e		1600	NT[g]	NT[g]	0.91	2.10
6f		170	NT[g]	NT[g]	0.48	3.50
6g		32	90	NT[g]	0.99	2.31
6h		4.3	12	NT[g]	0.85	2.31
6i		3.2	12	3.9	0.38	3.06

[a]IC_{50} values are calculated with one experiment performed in duplicate, with a standard deviation of 3-fold. [b]Binding affinity for hMCHR1. [c]Binding affinity for rMCHR1. [d]Antagonistic activity against hMCHR1, inhibition of MCH-stimulated arachidonic acid release from CHO cells expressing hMCHR1. [e]Relative activity of inhibition of Rb^+ efflux from CHO cells expressing the hERG channel (positive control is 0, vehicle is 1.0). [f]Measured at pH 7.4. [g]Not tested.

activity (Rb value) to astemizole (positive control is 0) and vehicle (control is 1). The criterion for the Rb value was set to be >0.8, which was determined in an in-house study. Further evaluation of cardiovascular safety was conducted using an in vitro patch-clamp study.[22]

The results of replacement of one of the benzene rings in the 4′-fluorobiphenyl moiety of **1** are shown in Table 1. An *n*-butoxyphenyl (**6a**) or a 4-fluorophenylbutyl (**6i**) analogue maintained in vitro activities (hMCHR1 IC_{50} of 2.0 and 3.2 nM, arachidonic acid release IC_{50} of 2.6 and 3.9 nM, respectively) with slight improvement in the Rb value (0.38−0.41). Replacement of the *n*-butyl group of **6a** with a cyclopropylmethyl (**6b**) or a cyclopropylethyl (**6c**) group led to an improvement of the Rb value with a high in vitro binding affinity. Compound **6b** showed 2-fold weaker antagonistic activity than **6c** (4.3 nM vs 2.0 nM); however, **6b** showed a better Rb value than **6c** did (0.90 vs 0.72). The cyclopropyl derivative **6b** showed more potent in vitro binding affinity (1.8 nM) with an improved Rb value than the corresponding isopropyl analogue **6d** (IC_{50} of 4.4 nM, Rb value of 0.70). These results imply that the π-property confers reduced lipophilicity on the cyclopropane ring, aiding binding to the MCHR1 receptor and contributing to a decrease in hERG inhibition.

Introduction of a more polar substituent such as sulfone (**6e**), ketone (**6h**), or amide (**6g**) improved the Rb value (0.85−0.99), whereas that of the ketone derivative **6f** did not (Rb value of 0.48). A good correlation between the Rb value and log *D* (pH 7.4) value was observed in these compounds: log *D* values of **6e**, **6g**, and **6h** are in the range 2.10−2.31, whereas that of **6f** is 3.50. The ketone derivative **6h** (Rb value of 0.85) showed a high binding affinity for hMCHR1 (IC_{50} of 4.3 nM); however, **6e** and **6g** showed significantly decreased affinities (7- to 370-fold decrease).

We previously discussed the SAR of the biphenyl moiety of the compound **1** analogue in a docking study performed using a homology model of hMCHR1.[14a] In this model, the biphenyl group was bound to the lipophilic binding pocket of MCHR1; however, a hydrophilic space was also observed in the same binding site. On the basis of this observation, we believe that

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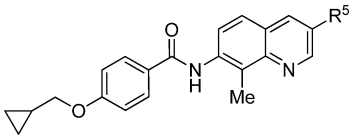
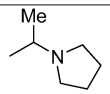
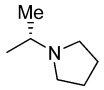
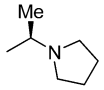
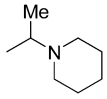
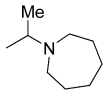
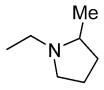
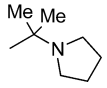
Table 2. In Vitro Pharmacological and Physicochemical Data for MCHR1 Antagonists



		IC_{50} (nM)[a]				
Cpd	R^5	hMCHR1[b]	rMCHR1[c]	AA release[d]	Rb value @ 30 μM[e]	logD @ pH 7.4[f]
10a		1.7	4.2	6.4	1.02	3.00
(*S*)-10a		3.4	8.0	4.0	0.85	2.99
(*R*)-10a		1.1	1.9	2.9	1.11	2.99
10b		2.4	8.3	3.2	0.80	3.49
10c		2.0	5.6	3.4	0.73	3.91
6j		2.5	6.7	NT[g]	0.62	3.16
22		7.8	8.3	NT[g]	0.62	3.08

[a]IC_{50} values are calculated with one experiment performed in duplicate, with a standard deviation of 3-fold. [b]Binding affinity for hMCHR1. [c]Binding affinity for rMCHR1. [d]Antagonistic activity against hMCHR1, inhibition of MCH-stimulated arachidonic acid release from CHO cells expressing hMCHR1. [e]Relative activity of inhibition of Rb^+ efflux from CHO cells expressing the hERG channel (positive control is 0, vehicle is 1.0). [f]Measured at pH 7.4. [g]Not tested.

the binding mode of the alkyl-linked analogue **6i** is probably different from that of **6h**; potent in vitro activity of **6i** can be achieved through a lipophilic interaction with MCHR1, and the ketone group of **6h** may form a hydrogen bond with a hydrophilic amino acid residue.

Among the compounds in Table 1, we chose the cyclopropylmethoxy derivative **6b** for further optimization. Table 2 displays the effects of the methyl group(s) at the α-position of the nitrogen atom of pyrrolidine. The **6b** analogues, **10a** (and its *S*- and *R*-isomers (***S***)-**10a** and (***R***)-**10a**), **10b**, **10c**, **6j**, and **22** showed nanomolar in vitro activities with moderate to excellent Rb values (0.62−1.11). Among these, the optically active pyrrolidine derivative (***R***)-**10a** showed the highest Rb value (1.11) with potent in vitro activities (hMCHR1 IC_{50} of 1.1 nM, arachidonic acid release of 2.9 nM). Considering the influence of the methyl group on the benzylic carbon, Rb values of **10a** and **10b** (1.02 and 0.80) were equal to or higher than those of **6b** and **6c** (0.90 and 0.72), although the log *D* values of **10a** and **10b** (3.00 and 3.49) were higher than those of **6b** and **6c** (2.77 and 3.40). These results imply that the methyl group introduced to the benzylic carbon of **6b** and **6c** attenuates cation−π and/or CH−π interaction(s) with the hERG K^+ channel through steric hindrance. In contrast, 2-methylpyrrolidine derivative **6j** (racemate) decreased the Rb value (0.62). Furthermore, introduction of an additional methyl group on the benzylic carbon of **10a** to yield the dimethyl compound **22** resulted in a loss of affinity (hMCHR1 IC_{50} of 7.8 nM) as well as a decrease in the Rb value (0.62). The methyl group at the 2-position of the pyrrolidine ring or the second methyl group introduced into the benzylic carbon increased lipophilicity and may show new CH−π interaction with the hERG K^+ channel. The patch-clamp study of (***R***)-**10a**, however, revealed that the hERG K^+ inhibitory activity was only minimally improved from that of compound **1** (88.1% vs 93.8%, Table 3).

We also performed a docking study of (***R***)-**10a** to the hERG K^+ channel by characterizing the binding site and binding mode.[22] Briefly, the number and relative position of selected residues involved in (***R***)-**10a** binding to the hERG K^+ channel were determined using site-directed mutagenesis combined with a tandem approach: a tandem dimer of the hERG K^+

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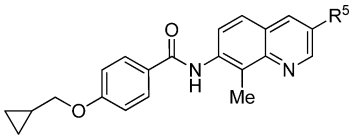
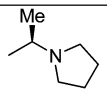
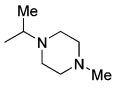
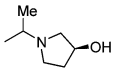
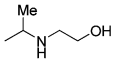
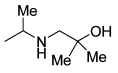
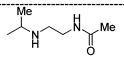
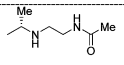
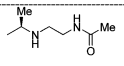
Table 3. In Vitro Pharmacological and Physicochemical Data for MCHR1 Antagonists



		IC$_{50}$ (nM)[a]				
Cpd	R^5	hMCHR1[b]	rMCHR1[c]	AA release[d]	Patch clamp @10 μM[e]	logD @ pH 7.4[f]
(*R*)-10a		1.1	1.9	2.9	88.1	2.99
10d		12	20	NT[g]	NT[g]	2.74
10e		5.5	19	NT[g]	58.4	2.45
10f		3.8	13	NT[g]	84.8	2.06
10g		1.5	2.7	6.4	63.3	2.67
10h		1.9	3.4	NT[g]	29.3	2.14
(*S*)-10h		5.3	12	6.9	33.3	2.13
(*R*)-10h		1.5	2.4	4.7	33.2	2.10
1	-	0.54	1.0	2.8	93.8	2.77

[a]IC$_{50}$ values are calculated with one experiment performed in duplicate, with a standard deviation of 3-fold. [b]Binding affinity for hMCHR1. [c]Binding affinity for rMCHR1. [d]Antagonistic activity against hMCHR1, inhibition of MCH-stimulated arachidonic acid release from CHO cells expressing hMCHR1. [e]% of inhibition. [f]Measured at pH 7.4. [g]Not tested.

channel was constructed, where mutation was introduced into one subunit to determine the residues that bind to (*R*)-**10a**. The results revealed that at least two Tyr652 molecules in the neighboring subunits are simultaneously involved in (*R*)-**10a** binding (Supporting Information Figure S2). On the basis of the topological information of the binding mode, a docking study of (*R*)-**10a** was performed against an hERG homology model. Multiple drug binding modes were generated for (*R*)-**10a**, and binding modes satisfying the above results were selected. Final docking models were generated after energy minimizations were performed using MOE (Chemical Computing Group, Canada).[28]

The conclusions from these studies are summarized in Figure 3. All ring substructures of (*R*)-**10a** were involved in the interaction with the hERG channel. Two Tyr652 side chains in adjacent subunits interacted with the terminal benzene ring and pyridine in the quinoline ring of (*R*)-**10a** via $\pi-\pi$ interactions (red circle), and a terminal pyrrolidine ring was recognized by Phe656, which constitutes a shallow pocket on the surface of the pore (yellow circle). The cyclopropane moiety was accommodated by a small pocket located in the back of the

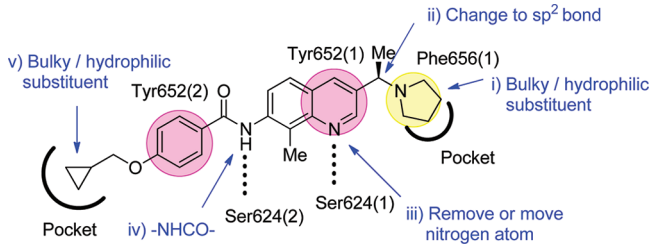


Figure 3. Proposed hERG K$^+$ channel binding mode for compound (*R*)-**10a** (black) and hypothetical modification to avoid hERG binding (blue). The number in parentheses after the residue name represents the subunit number.

selectivity filter, and two nitrogen atoms in the amide and in the quinoline ring formed hydrogen bonds with Ser624 molecules.

Recommended approaches to attenuate the hERG inhibition of (*R*)-**10a** are described in Figure 3 (blue): (i) introduction of a bulky and/or hydrophilic moiety near the terminal amine group; (ii) introduction of an sp^2 bond on the benzylic carbon; (iii) removal of the nitrogen atom from the quinoline core; (iv) reversal of the amide bond from CONH to NHCO; (v)

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Table 4. In Vivo Pharmacokinetic Profile of (*R*)-10h[a]

		iv (1 mg/kg)		po (3 mg/kg)		
Cpd	F[b] (%)	CL_{total}[c] (L h^{-1} kg^{-1}))	V_{ss}[d] (L kg^{-1})	C_{max}[e] (ng mL^{-1})	T_{max}[f] (h)	AUC_{0-24h}[g] (ng h mL^{-1})
(*R*)-10 h	43	0.69	1.21	274	4.0	1940

[a] n = 3; SD rats (male, 8 W). [b] Rat bioavailability. [c] Total clearance. [d] Volume of distribution at the steady state. [e] Maximal plasma concentration. [f] Time of maximal concentration. [g] Area under the blood concentration time curve (0−24 h).

replacement of the terminal cyclopropylmethyl group with a bulky and/or hydrophilic substituent.

In the SAR and docking study, replacement of the pyrrolidine with a bulky and/or hydrophilic amine appeared to be an ideal design, and the results are described in Table 3. Replacement of the pyrrolidine with piperazine (**10d**) decreased hMCHR1 binding affinity. Introduction of a hydroxyl group at the 3-position of the pyrrolidine ring provided **10e**, which showed potent hMCHR1 binding affinity (IC_{50} = 5.5 nM) with attenuated hERG K^+ inhibition in a patch-clamp study (58.4%). We also introduced a hydroxyl group at the 3-position of pyrrolidine in compound **1**; however, hERG inhibitory activity remained potent (97%, data not shown). These results suggest that replacement of terminal benzene with a cyclopropylmethoxy group and introduction of a hydroxyl group in the amine moiety act additively to decrease hERG potential. The (2-hydroxy-2-methylpropyl)amine derivative **10g** displayed more potent in vitro MCHR1 affinity than the hydroxyethylamine derivative **10f**. Interestingly, although the log *D* of compound **10g** was higher than that of the "less bulky" compound **10f** (2.67 vs 2.06), **10g** showed decreased hERG K^+ channel inhibitory activity in a patch-clamp study (63.3% vs 84.8%). Moreover, replacement of the pyrrolidine with [2-(acetylamino)ethyl]amine to provide **10h** dramatically decreased hERG potency (29.3%) without a significant loss of MCHR1 affinity. Between the two enantiomers of **10h**, the *R*-isomer (***R*)-10h** showed 3.5-fold more potent hMCHR1 binding affinity (IC_{50} = 1.5 nM) than (***S*)-10h** without significant hERG K^+ channel inhibition in a patch clamp study (33.2%).

The pharmacokinetic profile of (***R*)-10h** described in Table 4 suggests that (***R*)-10h** is orally available and could penetrate the brain. Subsequently, the pharmacological effect of compound (***R*)-10h** was evaluated using DIO-F344 rats fed high fat diet ad libitum. The compound (***R*)-10h** (1, 3, 5, and 10 mg/kg) was orally administered to DIO-F344 rats daily at the beginning of the dark cycle (at 5:00 p.m., 0 and 24 h), and the food intake from initial administration to 1 and 2 days later was measured. The results of a 2-day in vivo study of (***R*)-10h** are shown in Figure 4. Compound (***R*)-10h** significantly and dose-dependently suppressed food intake in DIO-F344 rats from 1 mg/kg.

Recently, we reported SAR and pharmacological studies of a potent and selective MCHR1 antagonist **1**, an analogue of (***R*)-10h**. An MCHR1-deficient mice study revealed that the anorectic effect of **1** was provoked by MCHR1 antagonism.[15] Compound (***R*)-10h** showed negligible activity for other receptors, transporters, and enzymes (data not shown), suggesting that the MCHR1 antagonistic activity of (***R*)-10h** exerted the anorectic effect observed in this study. Taken together, these data suggest that (***R*)-10h** is an orally bioavailable MCHR1 antagonist that exhibits excellent in vivo efficacy in a DIO rat model while maintaining a safety profile with respect to QTc prolongation.

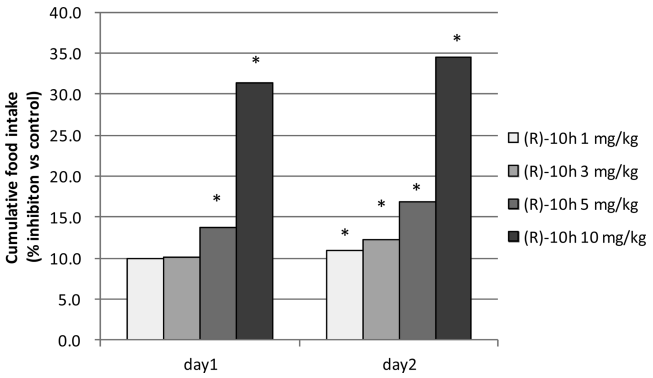

Figure 4. Effect of (***R*)-10 h** on 2-day food intake study in DIO-F344 rats: inhibition of cumulative food intake on days 1 and 2 in DIO-F344 rats. Compound was dosed once daily (at 5:00 p.m., 0 and 24 h). Cumulative food intake inhibition rate was calculated by dividing average food intake of each compound administered group by average food intake of the control group at each measured point. The values shown are a ratio of the control: (∗) $p < 0.025$ vs control group (Williams test, n = 6 for each group).

CONCLUSION

We conducted a SAR study both for MCHR1 antagonistic activity and for hERG K^+ channel inhibitory activity using 3-aminomethylquinoline-based MCHR1 antagonists related to compound **1**. In parallel with the ligand-based SAR study, the docking study of the key compound (***R*)-10a** with the hERG K^+ channel was conducted to support the optimization study. We achieved the following: (i) replacement of the terminal *p*-fluorophenyl moiety with a cyclopropylmethoxy group, introduction of a methyl group onto the benzylic carbon, and employment of a [2-(acetylamino)ethyl]amino group as an amine to synergistically reduce inhibitory activity against the hERG K^+ channel allowed us to identify the optically active compound (***R*)-10h** as a novel, potent MCHR1 antagonist (binding IC_{50} = 1.5 nM, antagonistic activity IC_{50} = 4.7 nM); (ii) a docking study was conducted to support a design for reducing hERG K^+ channel inhibition; (iii) practical asymmetric synthesis of (***R*)-10h** was achieved to determine the absolute configuration. (***R*)-10h** did not show significant hERG K^+ channel inhibitory activity in a patch-clamp study. Furthermore, (***R*)-10h** (1, 3, 5, and 10 mg/kg) dose-dependently and significantly suppressed food intake in a 2-day DIO-F344 rat study. In conclusion, (***R*)-10h** is an orally active MCHR1 antagonist with reduced hERG-associated cardiovascular liabilities.

EXPERIMENTAL SECTION

Melting points (mp) were determined on a Yanagimoto micromelting point apparatus or a Büchi melting point B545 apparatus and are uncorrected. Proton nuclear magnetic resonance (^{1}H NMR) spectra were recorded on a Varian Gemini 200 or Varian Mercury 300 NMR spectrometer. Chemical shifts were reported in δ value (ppm) with tetramethylsilane as an internal standard. Splitting patterns are

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designated as follows: s, singlet; d, doublet; t, triplet; dd, double doublet; q, qualtet; quint, quintet; sext, sextet; m, multiplet; br, broad. Coupling constants (*J*) are reported in hertz (Hz). LC/MS (ESI positive) spectra were recorded on a Waters Micromass ZQ 2000. Elemental analysis (C, H, N) to determine the purity of test compounds was conducted by the Analytical Department of Takeda Pharmaceutical Co., and the results were within 0.4% of theoretical values. Purity of compounds was >95%, as established by elemental analysis. The data from elemental analysis are in Supporting Information. Thin-layer chromatography (TLC) analyses were performed with silica gel 60 F_{254} plate (Merck no. 5715) and alumina 60 F_{254} plate (type E). Chromatographic separations were performed with Merck silica gel 60 (Merck no. 7734), ICN alumina B, Akt. I (activity grade III), and NH-silica gel (Fuji Sylysia).

7-Amino-8-methylquinoline-3-carboxaldehyde (4). (a) 2-Dimethylaminomethylene-1,3-bis(dimethylimmonio)propane Bis(tetrafluoroborate) (3).[23] During 1.5 h, to an ice-cooled mixture of bromoacetic acid (100 g, 0.72 mol) and phosphoryl chloride (200 mL, 2.15 mol) was added DMF (336 mL, 4.36 mol), maintaining below 15 °C. The mixture was stirred at 110 °C for 3 h and then cooled to 0 °C. (*Caution*: While heating, gaseous CO_2 is evolved vigorously and the temperature rose quickly to 150 °C.) To the resulting solution was added a mixture of 48% aqueous solution of HBF_4 (500 g, 3.83 mol) and MeOH (200 mL) dropwise over a period of 1 h. (*Caution*: The reaction is exothermic, and the cooled solution becomes a viscous liquid.) To this mixture was added 2-propanol (1000 mL), and the mixture was stirred at 0 °C for 2 h. The precipitate was filtered, rinsed with ice-cooled 2-propanol, quickly air-dried, and dried at 60 °C under reduced pressure to give the title compound (207 g, 81%) as pale yellow crystals. (*Caution*: The crystals are hygroscopic.) ^{1}H NMR (300 MHz, DMSO-d_6) δ 3.37 (9H, s), 3.53 (9H, s), 8.40 (9H, s).

(b) 7-Amino-8-methylquinoline-3-carboxaldehyde (4). A mixture of 2-methyl-1,3-benzenediamine (**2**, 30.0 g, 246 mmol) and vinamidinium bis-tetrafluoroborate (**3**, 263 g, 737 mmol) in 2-propanol (500 mL) was stirred for 16 h under reflux. The reaction mixture was allowed to cool to room temperature, and to this mixture was added 1 M HCl (500 mL). Then the mixture was stirred at 70 °C for 5 h. The reaction mixture was cooled to room temperature, and the resulting precipitate was collected by filtration. The precipitate was washed with water, MeCN, and isopropyl ether, successively. A mixture of the precipitate (61.6 g) and K_2CO_3 (170 g, 1.23 mol) in AcOEt (500 mL)−H_2O (500 mL) was stirred vigorously at 90 °C for 16 h and then cooled to room temperature. The organic layer was separated, washed with brine, and dried over Na_2SO_4. This was filtered through a pad of silica gel (100 g), and the filtrate was concentrated under reduced pressure to give an orange solid. This solid was triturated with isopropyl ether to give **4** (35.4 g, 77%) as an orange powder: ^{1}H NMR (300 MHz, DMSO-d_6) δ 2.43 (3H, s), 6.17 (2H, br s), 7.15 (1H, t, *J* = 8.7 Hz), 7.71 (1H, d, *J* = 8.7 Hz), 8.51 (1H, d, *J* = 2.4 Hz), 9.04 (1H, d, *J* = 2.4 Hz), 10.0 (1H, s).

8-Methyl-3-(pyrrolidin-1-ylmethyl)quinolin-7-amine (5a). To a suspension of **4** (21.0 g, 113 mmol) and pyrrolidine (28.3 mL, 145 mmol) in dichloroethane (210 mL) was added sodium triacetoxyborohydride (35.8 g, 169 mmol). After the mixture was stirred at room temperature for 4.5 h, to this mixture was added aqueous $NaHCO_3$ and the organic layer was separated. The organic layer was concentrated under reduced pressure and the residue was chromatographed on NH-silica gel (AcOEt) to give **5a** (25.7 g, 94%) as a viscous oil. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.79 (4H, m), 2.54 (4H, m), 2.59 (3H, s), 3.74 (2H, s), 3.98 (2H, s), 6.98 (1H, d, *J* = 8.6 Hz), 7.47 (1H, d, *J* = 8.6 Hz), 7.91 (1H, d, *J* = 2.2 Hz), 8.76 (1H, d, *J* = 2.2 Hz).

8-Methyl-3-[(2-methylpyrrolidin-1-yl)methyl]quinolin-7-amine (5b). The title compound was prepared in 66% yield starting from **4** using the procedure described for **5a**. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.21 (3H, d, *J* = 6.0 Hz), 1.39−1.53 (1H, m), 1.55−1.79 (2H, m), 1.88−2.00 (1H, m), 2.15 (1H, q, *J* = 8.9 Hz), 2.37−2.50 (1H, m), 2.59 (3H, s), 2.85−2.95 (1H, m), 3.31 (1H, d, *J* = 13.2 Hz), 3.98 (2H, s), 4.08−4.16 (1H, m), 6.99 (1H, d, *J* = 8.7 Hz), 7.49 (1H, d, *J* = 8.7 Hz), 7.91 (1H, d, *J* = 2.1 Hz), 8.77 (1H, d, *J* = 2.1 Hz).

4-Butoxy-*N*-[8-methyl-3-(pyrrolidin-1-ylmethyl)quinolin-7-yl]benzamide (6a). The title compound was prepared in 20% yield starting from **5a** using the procedure described for **6d**: mp 161 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.99 (3H, t, *J* = 7.3 Hz), 1.51 (2H, m), 1.80 (6H, m), 2.55 (4H, m), 2.80 (3H, s), 3.80 (2H, s), 4.03 (2H, t, *J* = 6.5 Hz), 6.98 (2H, m), 7.68 (1H, d, *J* = 9.0 Hz), 7.89 (3H, m), 8.04 (1H, d, *J* = 2.2 Hz), 8.22 (1H, d, *J* = 8.8 Hz), 8.87 (1H, d, *J* = 2.2 Hz). Anal. ($C_{26}H_{31}N_3O_2\cdot0.3H_2O$) C, H, N.

4-(Cyclopropylmethoxy)-*N*-[8-methyl-3-(pyrrolidin-1-ylmethyl)quinolin-7-yl]benzamide (6b). The title compound was prepared in 50% yield starting from **5a** using the procedure described for **6d**: mp 168 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.39 (2H, m), 0.69 (2H, m), 1.30 (1H, m), 1.81 (4H, m), 2.56 (4H, m), 2.80 (3H, s), 3.80 (2H, s), 3.88 (2H, d, *J* = 6.8 Hz), 7.00 (2H, m), 7.68 (1H, d, *J* = 8.6 Hz), 7.90 (3H, m), 8.05 (1H, d, *J* = 2.0 Hz), 8.23 (1H, d, *J* = 8.8 Hz), 8.87 (1H, d, *J* = 2.0 Hz). Anal. ($C_{26}H_{29}N_3O_2\cdot0.2H_2O$) C, H, N.

4-(2-Cyclopropylethoxy)-*N*-[8-methyl-3-(pyrrolidin-1-ylmethyl)quinolin-7-yl]benzamide (6c). The title compound was prepared in 58% yield starting from **5a** using the procedure described for **6d**: mp 147−148 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.12−0.18 (2H, m), 0.48−0.55 (2H, m), 0.81−0.95 (1H, m), 1.73 (2H, q, *J* = 6.8 Hz), 1.78−1.85 (4H, m), 2.54−2.60 (4H, m), 2.81 (3H, s), 3.81 (2H, s), 4.12 (2H, t, *J* = 6.7 Hz), 6.99−7.05 (2H, m), 7.70 (1H, d, *J* = 8.9 Hz), 7.89−7.96 (3H, m), 8.06 (1H, d, *J* = 2.1 Hz), 8.25 (1H, d, *J* = 8.9 Hz), 8.89 (1H, d, *J* = 2.1 Hz). Anal. ($C_{27}H_{31}N_3O_2\cdot H_2O$) C, H, N.

4-(2-Methylpropoxy)-*N*-[8-methyl-3-(pyrrolidin-1-ylmethyl)-quinolin-7-yl]benzamide (6d). To a mixture of 4-isobutoxybenzoic acid (0.12 g, 0.62 mmol) and oxalyl chloride (0.16 mL, 1.85 mmol) in THF (3 mL) were added 3 drops of DMF. The mixture was stirred at room temperature for 2 h and then concentrated under reduced pressure. The resulting residue was dissolved in THF (3 mL), and the solution was added to a mixture of **5a** (0.12 g, 0.52 mmol) and triethylamine (0.14 mL, 1.03 mmol) in THF (3 mL). After being stirred at room temperature for 15 h, the reaction mixture was concentrated under reduced pressure and the residue was partitioned between AcOEt and water. The organic layer was washed with brine, dried over $MgSO_4$, filtered, and concentrated under reduced pressure. The resulting residue was purified by column chromatography (NH-silica gel, hexane/AcOEt/$CHCl_3$ = 2/1/1, AcOEt) and the resulting solid was recrystallized from isopropyl ether and AcOEt to give **6d** (0.11 g, 0.26 mmol, 51% yield) as a pale yellow solid: mp 138−139 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.06 (6H, d), 1.77−1.85 (4H, m), 2.06−2.21 (1H, m), 2.52−2.61 (4H, m), 2.81 (3H, s), 3.79−3.83 (4H, m), 6.97−7.04 (2H, m), 7.70 (1H, d, *J* = 8.9 Hz), 7.88−7.95 (3H, m), 8.06 (1H, d, *J* = 2.1 Hz), 8.25 (1H, d, *J* = 8.9 Hz), 8.89 (1H, d, *J* = 2.3 Hz). Anal. ($C_{26}H_{31}N_3O_2$) C, H, N.

4-Butylsulfonyl-*N*-[8-methyl-3-(pyrrolidin-1-ylmethyl)-quinolin-7-yl]benzamide (6e). The title compound was prepared in 36% yield starting from **5a** using the procedure described for **6d**: mp 211 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.90 (3 H, t, *J* = 7.3 Hz), 1.38 (2H, m), 1.67 (2H, m), 1.81 (4H, m), 2.56 (4H, m), 2.81 (3H, s), 3.12 (2H, m), 3.81 (2H, s), 7.72 (1H, d, *J* = 8.4 Hz), 8.07 (7H, m), 8.90 (1H, d, *J* = 2.2 Hz). Anal. ($C_{26}H_{31}N_3O_3S\cdot0.2H_2O$) C, H, N.

4-Hexanoyl-*N*-[8-methyl-3-(pyrrolidin-1-ylmethyl)quinolin-7-yl]benzamide (6f). The title compound was prepared in 36% yield starting from **5a** using the procedure described for **6d**: mp 175 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.93 (3H, m), 1.39 (4H, m), 1.82 (6H, m), 2.57 (4H, m), 2.82 (3H, s), 3.02 (2H, t, *J* = 7.5 Hz), 3.82 (2H, s), 7.72 (1H, d, *J* = 9.2 Hz), 8.02 (1H, d, *J* = 2.2 Hz), 8.09 (5H, m), 8.19 (1H, d, *J* = 8.8 Hz), 8.91 (1H, d, *J* = 2.2 Hz). Anal. ($C_{28}H_{33}N_3O_2\cdot0.1H_2O$) C, H, N.

***N*-[8-Methyl-3-(pyrrolidin-1-ylmethyl)quinolin-7-yl]-4-(pentanoylamino)benzamide (6g).** The title compound was prepared in 16% yield starting from **5a** using the procedure described for **6d**: mp 193−194 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.97 (3H, t, *J* = 7.2 Hz), 1.44 (2H, m) 1.73 (2H, m), 1.81 (4H, m), 2.41 (2H, m), 2.57 (4H, m), 2.81 (3H, s), 3.81 (2H, s), 7.30 (1H, s), 7.69 (3H, m),

7.92 (3H, m), 8.05 (1H, d, J = 2.2 Hz), 8.21 (1H, d, J = 8.8 Hz), 8.88 (1H, d, J = 2.4 Hz). Anal. ($C_{27}H_{32}N_4O_2 \cdot 0.4H_2O$) C, H, N.

5-(4-Fluorophenyl)-*N*-[8-methyl-3-(pyrrolidin-1-ylmethyl)-quinolin-7-yl]-5-oxopentanamide (6h). The title compound was prepared in 10% yield starting from **5a** using the procedure described for **6d**: mp 172−173 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.79 (4H, m), 2.23 (2H, m), 2.57 (6H, m), 2.73 (3H, s), 3.15 (2H, t, J = 6.7 Hz), 3.78 (2H, s), 7.12 (2H, t, J = 8.8 Hz), 7.48 (1H, s), 7.64 (1H, d, J = 8.8 Hz), 8.03 (4H, m), 8.85 (1H, d, J = 2.0 Hz). Anal. ($C_{26}H_{28}$ F N_3O_2) C, H, N.

5-(4-Fluorophenyl)-*N*-[8-methyl-3-(pyrrolidin-1-ylmethyl)-quinolin-7-yl]pentanamide (6i). The title compound was prepared in 12% yield starting from **5a** using the procedure described for **6d**: mp 138−139 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.76 (2H, m), 1.88 (4H, m), 2.04 (3H, m), 2.50 (2H, t, J = 6.7 Hz), 2.67 (6H, m), 2.71 (3H, s), 3.91 (2H, s), 6.97 (2H, t, J = 8.7 Hz), 7.15 (1H, m), 7.24 (1H, s), 7.67 (1H, d, J = 8.8 Hz), 8.10 (1H, d, J = 9.1 Hz), 8.16 (1H, s), 8.88 (1H, d, J = 1.9 Hz). Anal. ($C_{26}H_{30}FN_3O$) C, H, N.

4-(Cyclopropylmethoxy)-*N*-{8-methyl-3-[(2-methylpyrrolidin-1-yl)methyl]quinolin-7-yl}benzamide (6j). The title compound was prepared in 72% yield starting from **5b** using the procedure described for **6d**: mp 159 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.36−0.43 (2H, m), 0.65−0.73 (2H, m), 1.22 (3H, d, J = 6.0 Hz), 1.25−1.38 (1H, m), 1.41−1.55 (1H, m), 1.61−1.81 (2H, m), 1.90−2.04 (1H, m), 2.17 (1H, q, J = 8.9 Hz), 2.42−2.54 (1H, m), 2.81 (3H, s), 2.88−2.96 (1H, m), 3.36 (1H, d, J = 13.4 Hz), 3.89 (2H, d, J = 7.0 Hz), 4.19 (1H, d, J = 13.4 Hz), 6.98−7.04 (2H, m), 7.70 (1H, d, J = 8.9 Hz), 7.88−7.95 (3H, m), 8.05 (1H, d, J = 2.1 Hz), 8.24 (1H, d, J = 8.9 Hz), 8.89 (1H, d, J = 2.1 Hz). Anal. ($C_{27}H_{31}N_3O_2 \cdot 0.2H_2O$) C, H, N.

1-(7-Amino-8-methylquinolin-3-yl)ethanol (7). To an ice-cooled solution of methylmagnesium bromide (3 M in Et_2O, 71.6 mL, 214.8 mmol) in THF (200 mL) was added dropwise a solution of **4** (10.0 g, 53.7 mmol) in THF (200 mL). After being stirred at room temperature for 3 h, the reaction mixture was poured into 10% NH_4Cl (1000 mL) and the mixture was extracted with AcOEt (500 mL). The extract was washed with brine and dried over $MgSO_4$. After removal of the solvent under reduced pressure, the residue was chromatographed on NH-silica gel (AcOEt) to give **7** as an orange amorphous (8.45 g, 78%). ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.57 (3H, d, J = 6.4 Hz), 2.57 (3H, s), 4.01 (2H, br s), 5.04 (1H, q, J = 6.4 Hz), 6.98 (2H, d, J = 8.7 Hz), 7.45 (2H, d, J = 8.7 Hz), 7.91 (2H, d, J = 2.4 Hz), 8.78 (2H, d, J = 2.4 Hz).

3-(1-Chloroethyl)-8-methylquinolin-7-amine Dihydrochloride (8). A mixture of **7** (8.45 g, 41.8 mmol) and thionyl chloride (100 mL) was stirred at room temperature for 16 h. After removal of an excess amount of thionyl chloride under reduced pressure, the resulting residue was triturated with THF to give **8** (10.7 g, 87%) as a brown powder. ^{1}H NMR (300 MHz, $CDCl_3$) δ 2.05 (3H, d, J = 6.9 Hz), 3.19 (3H, s), 5.43 (1H, q, J = 6.9 Hz), 8.04 (1H, d, J = 8.7 Hz), 8.58 (1H, d, J = 8.7 Hz), 8.90 (1H, d, J = 2.0 Hz), 9.50 (1H, d, J = 2.0 Hz).

***N*-[3-(1-Chloroethyl)-8-methylquinolin-7-yl]-4-(cyclopropylmethoxy)benzamide (9).** To a mixture of 4-cyclopropylmethoxybenzoic acid (1.44 g, 7.49 mmol) and DMF (0.02 mL, 0.34 mmol) in THF (40 mL) was added a solution of oxalyl chloride (0.70 mL, 8.17 mmol) in THF (10 mL) under ice-cooling. After being stirred at room temperature for 1 h, the reaction mixture was concentrated under reduced pressure and the resulting residue was dissolved in THF (20 mL). This mixture was added to a solution of **8** (2.0 g, 6.81 mmol) and triethylamine (4.7 mL, 34.3 mmol) in THF (40 mL), and the mixture was stirred at room temperature for 16 h. The reaction mixture was filtered through a glass filter, and the filtrate was concentrated under reduced pressure. The resulting residue was partitioned between AcOEt and water. The AcOEt layer was washed with brine and dried over $MgSO_4$. After removal of the solvent under reduced pressure, the residue was purified by column chromatography (NH-silica gel, hexane/AcOEt = 4/1 to 1/1) to give **9** (0.91 g, 34%) as a yellow solid. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.37−0.42 (2H, m), 0.66−0.73 (2H, m), 1.26−1.34 (1H, m), 1.98 (3H, d, J = 6.9 Hz), 2.81 (3H, s), 3.89 (2H, d, J = 6.9 Hz), 5.31 (1H, q, J = 6.8 Hz), 7.01 (2H, d, J = 8.8 Hz), 7.73 (1H, d, J = 8.8 Hz), 7.90−7.93 (3H, m), 8.12 (1H, d, J = 2.4 Hz), 8.34 (1H, d, J = 8.8 Hz), 8.98 (1H, d, J = 2.4 Hz).

4-(Cyclopropylmethoxy)-*N*-[8-methyl-3-(1-pyrrolidin-1-ylethyl)quinolin-7-yl]benzamide (10a). A mixture of **9** (21.9 g, 50.8 mmol), pyrrolidine (12.6 mL, 152 mmol), K_2CO_3 (8.43 g, 61.0 mmol), and KI (10.1 g, 61.0 mmol) in DMF (300 mL) was stirred at 70 °C for 3 h. The reaction mixture was partitioned between AcOEt and water. The organic layer was washed with water and brine and dried over $MgSO_4$. After removal of the solvent under reduced pressure, the residue was chromatographed on NH-silica gel (hexane/AcOEt = 1/1) and silica gel (hexane/AcOEt = 1/1, AcOEt, AcOEt/MeOH = 4/1) to give a pale yellow solid. This was dissolved in AcOEt, and the solution was washed with 10% $NaHCO_3$ and brine and dried over $MgSO_4$. After removal of the solvent under reduced pressure, the resulting residue was triturated with Et_2O to give **10a** (15.8 g, 72%) as a pale yellow powder: mp 189−190 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.39 (2H, m), 0.69 (2H, m), 1.21−1.38 (1H, m), 1.50 (3H, d, J = 6.6 Hz), 1.79 (4H, m), 2.31−2.49 (2H, m), 2.53−2.66 (2H, m), 2.81 (3H, s), 3.37−3.49 (1H, m), 3.89 (2H, d, J = 6.9 Hz), 7.01 (2H, d, J = 8.8 Hz), 7.69 (1H, d, J = 8.8 Hz), 7.90−7.93 (3H, m), 8.05 (1H, d, J = 2.2 Hz), 8.24 (1H, d, J = 8.8 Hz), 8.92 (1H, d, J = 2.2 Hz). Anal. ($C_{27}H_{31}N_3O_2 \cdot 0.1$ H_2O) C, H, N.

Optical Resolution of 10a. A 200 mL solution of **10a** (2.0 g) in ethanol was loaded onto Chiralpak AD (50 mm i.d. × 500 mm) through an injection line. Preparative HPLC was run at a flow of 80 mL/min ethanol at 35 °C (UV 254 nm). Fractions eluting at 17 min (tR1, **LP-10a**) and at 32 min (tR2, **P-10a**) were collected and concentrated. To the resulting residue was added hexane, and the mixture was concentrated under reduced pressure to afford **LP-10a** (1.0 g, >99.9% ee) and **P-10a** (1.0 g, >99.9% ee), respectively.

4-(Cyclopropylmethoxy)-*N*-{8-methyl-3-[(1*S*)-1-pyrrolidin-1-ylethyl]quinolin-7-yl}benzamide (LP-10a = (*S*)-10a). White solid, mp 189−190 °C. $[\alpha]^{25}_D$ −37.25 (c 0.996, MeOH). Anal. ($C_{27}H_{31}N_3O_2$) C, H, N.

4-(Cyclopropylmethoxy)-*N*-{8-methyl-3-[(1*R*)-1-pyrrolidin-1-ylethyl]quinolin-7-yl}benzamide (P-10a = (*R*)-10a). White solid, mp 189−190 °C. $[\alpha]^{25}_D$ +39.32 (c 1.00, MeOH). Anal. ($C_{27}H_{31}N_3O_2$) C, H, N.

Synthesis of 4-(Cyclopropylmethoxy)-*N*-{8-methyl-3-[(1*R*)-1-pyrrolidin-1-ylethyl]quinolin-7-yl}benzamide ((*R*)-10a) from 17. A mixture of **17** (6.00 g, 16.0 mmol, 99% ee), 1,4-dibromobutane (4.14 g, 19.2 mmol), and Na_2CO_3 (5.10 g, 48.0 mmol) in DMF (150 mL) was stirred at 60 °C for 14 h. After the reaction was completed, water (200 mL) was added to the reaction mixture and the mixture was extracted with AcOEt (500 mL). The extract was washed with brine, dried over $MgSO_4$, filtered, and concentrated under reduced pressure. The resulting residue was chromatographed on silica gel (AcOEt/MeOH = 5/1) to give (***R***)-**10a** as colorless crystals (4.20 g, 61%, 98% ee by chiral HPLC analysis).

4-(Cyclopropylmethoxy)-*N*-[8-methyl-3-(1-piperidin-1-ylethyl)quinolin-7-yl]benzamide (10b). The title compound was prepared in 73% yield starting from **9** using the procedure described for **10a**: mp 184−186 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.34−0.47 (2H, m), 0.62−0.77 (2H, m), 1.19−1.45 (3H, m), 1.48 (3H, d, J = 6.8 Hz), 1.52−1.65 (4H, m), 2.29−2.55 (4H, m), 2.82 (3H, s), 3.68 (1H, q, J = 6.8 Hz), 3.89 (2H, d, J = 7.0 Hz), 7.01 (2H, d, J = 8.7 Hz), 7.71 (1H, d, J = 8.9 Hz), 7.86−8.00 (4H, m), 8.26 (1H, d, J = 8.9 Hz), 8.93 (1H, d, J = 2.1 Hz). Anal. ($C_{28}H_{33}N_3O_2 \cdot 0.1$ H_2O) C, H, N.

***N*-[3-(1-Azepan-1-ylethyl)-8-methylquinolin-7-yl]-4-(cyclopropylmethoxy)benzamide (10c).** The title compound was prepared in 71% yield starting from **9** using the procedure described for **10a**: mp 169−171 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.34−0.46 (2H, m), 0.62−0.76 (2H, m), 1.21−1.39 (1H, m), 1.47 (3H, d, J = 6.6 Hz), 1.59 (8H, br s), 2.66 (4H, br s), 2.82 (3H, s), 3.89 (2H, d, J = 7.0 Hz), 4.03 (1H, q, J = 6.6 Hz), 7.01 (2H, d, J = 8.9 Hz), 7.70 (1H, d, J = 8.9 Hz), 7.85−8.05 (4H, m), 8.24 (1H, d, J = 8.9 Hz), 9.05 (1H, d, J = 2.3 Hz). Anal. ($C_{29}H_{35}N_3O_2 \cdot 0.2$ H_2O) C, H, N.

4-(Cyclopropylmethoxy)-*N*-{8-methyl-3-[1-(4-methylpiperazin-1-yl)ethyl]quinolin-7-yl}benzamide (10d). The title compound was prepared in 66% yield starting from **9** using the procedure

described for **10a**: mp 168−169 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.31−0.45 (2H, m), 0.58−0.77 (2H, m), 1.19−1.39 (1H, m), 1.47 (3H, d, J = 6.8 Hz), 2.27 (3H, s), 2.45 (8H, m), 2.82 (3H, s), 3.63 (1H, q, J = 6.6 Hz), 3.90 (2H, d, J = 7.0 Hz), 7.01 (2H, d, J = 8.9 Hz), 7.70 (1H, d, J = 8.9 Hz), 7.85−7.96 (3H, m), 7.99 (1H, d, J = 2.1 Hz), 8.27 (1H, d, J = 8.9 Hz), 8.94 (1H, d, J = 2.1 Hz). Anal. ($C_{28}H_{34}N_4O_2\cdot 0.2\ H_2O$) C, H, N.

4-(Cyclopropylmethoxy)-*N*-(3-{1-[(3*S*)-3-hydroxypyrrolidin-1-yl]ethyl}-8-methylquinolin-7-yl)benzamide (10e). The title compound was prepared in 72% yield starting from **9** using the procedure described for **10a**: mp 168 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ 0.28−0.45 (2 H, m), 0.53−0.69 (2 H, m), 1.13−1.36 (1 H, m), 1.42 (3H, d, J = 6.4 Hz), 1.49−1.65 (1H, m), 1.88−2.09 (1H, m), 2.17−2.46 (1H, m), 2.63 (3H, s), 2.68−2.95 (2H, m), 3.42−3.64 (1H, m), 3.92 (2H, d, J = 7.0 Hz), 4.10−4.30 (1H, m), 4.59−4.76 (1H, m), 7.07 (2H, d, J = 8.7 Hz), 7.58 (1H, d, J = 8.7 Hz), 7.80 (1H, d, J = 8.9 Hz), 8.02 (2H, d, J = 8.9 Hz), 8.19 (1H, d, J = 2.0 Hz), 8.90 (1H, s), 10.05 (1H, s). Anal. ($C_{27}H_{31}N_3O_3\cdot 0.1H_2O$) C, H, N.

4-(Cyclopropylmethoxy)-*N*-(3-{1-[(2-hydroxyethyl)amino]-ethyl}-8-methylquinolin-7-yl)benzamide (10f). The title compound was prepared in 59% yield starting from **9** using the procedure described for **10a**: mp 158 °C. ^{1}H NMR (300 MHz, DMSO-d_6) δ 0.31−0.42 (2H, m), 0.53−0.67 (2H, m), 1.18−1.35 (1H, m), 1.40 (3H, d, J = 6.6 Hz), 2.35−2.47 (1, m), 2.52−2.59 (1H, m), 2.63 (3H, s), 3.45 (2H, q, J = 5.5 Hz), 3.92 (2H, d, J = 7.2 Hz), 3.95−4.09 (1H, m), 4.50 (1H, t, J = 5.4 Hz), 7.07 (2H, d, J = 8.9 Hz), 7.58 (1H, d, J = 8.7 Hz), 7.78 (1H, d, J = 8.9 Hz), 8.02 (2H, d, J = 8.9 Hz), 8.20 (1H, d, J = 2.1 Hz), 8.93 (1H, d, J = 2.3 Hz), 10.05 (1 H, s). Anal. ($C_{25}H_{29}N_3O_3\cdot 0.5H_2O$) C, H, N.

4-(Cyclopropylmethoxy)-*N*-(3-{1-[(2-hydroxy-2-methylpropyl)amino]ethyl}-8-methylquinolin-7-yl)benzamide (10g). A mixture of **9** (0.44 g, 1.02 mmol), 1-amino-2-methylpropan-2-ol (0.91 g, 10.2 mmol), and DIPEA (0.45 mL, 2.58 mmol) in NMP (10 mL) was stirred at 60 °C for 5 h. The reaction mixture was partitioned between AcOEt and water. The organic layer was washed with brine, dried over Na_2SO_4, filtered, and concentrated in vacuo. The residue was chromatographed on NH-silica gel (AcOEt/MeOH = 95/5) followed by recrystallization from AcOEt and diisopropyl ether to obtain **10g** (0.22 g, 0.71 mmol, 70%) as a colorless crystalline solid, mp 124−125 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.36−0.43 (2H, m), 0.65−0.74 (2H, m), 1.15 (3H, s), 1.18 (3H, s), 1.25−1.39 (1H, m), 1.51 (3H, d, J = 6.6 Hz), 2.38 (1H, d, J = 11.7 Hz), 2.54 (1H, d, J = 11.7 Hz), 2.76 (1H, br), 2.82 (3H, s), 3.90 (2H, d, J = 7.0 Hz), 4.00 (1H, q, J = 6.6 Hz), 7.01 (2H, d, J = 8.7 Hz), 7.71 (1H, d, J = 8.9 Hz), 7.87−7.96 (3H, m), 8.00 (1H, d, J = 2.1 Hz), 8.27 (1H, d, J = 8.9 Hz), 8.91 (1H, d, J = 2.1 Hz). Anal. ($C_{27}H_{33}N_3O_3$) C, H, N.

***N*-[3-(1-{[2-(Acetylamino)ethyl]amino}ethyl)-8-methylquinolin-7-yl]-4-(cyclopropylmethoxy)benzamide (10h).** The title compound was prepared in 68% yield starting from **9** using the procedure described for **10g**: mp 169−170 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.36−0.43 (2H, m), 0.65−0.73 (2H, m), 1.26−1.36 (1H, m), 1.48 (3H, d, J = 6.8 Hz), 1.97 (3H, s), 2.51−2.62 (1H, m), 2.71−2.82 (1H, m), 2.81 (3H, s), 3.31 (2H, q, J = 6.0 Hz), 3.90 (2H, d, J = 7.0 Hz), 4.00 (1H, q, J = 6.6 Hz), 5.86 (1H, s), 7.02 (2H, d, J = 8.9 Hz), 7.70 (1H, d, J = 8.9 Hz), 7.91 (1H, s), 7.92 (2H, d, J = 8.9 Hz), 7.99 (1H, d, J = 2.3 Hz), 8.27 (1H, d, J = 8.9 Hz), 8.91 (1H, d, J = 2.3 Hz). Anal. ($C_{27}H_{32}N_4O_3\cdot 0.2H_2O$) C, H, N.

Optical Resolution of 10h. The optical resolution of **10h** was carried out using a method similar to the one described for **10a**: Chiralpak AD (50 mm i.d. × 500 mm), hexane/EtOH/DIPEA = 50/50/0.1, UV 220 nm, 60 mL/min, 30 °C. Less polar **LP-10h** (84 mg, >99.9% ee) and polar **P-10h** (85 mg, >99.9% ee) were collected from **10h** (174 mg).

***N*-{3-[(1*R*)-1-{[2-(Acetylamino)ethyl]amino}ethyl]-8-methylquinolin-7-yl}-4-(cyclopropylmethoxy)benzamide (LP-10h = (*R*)-10h).** White solid:, mp 169−170 °C. $[\alpha]^{25}_D$ +52.60° (c 0.1980, MeOH). Anal. ($C_{27}H_{32}N_4O_3\cdot 0.2H_2O$) C, H, N.

***N*-{3-[(1*S*)-1-{[2-(Acetylamino)ethyl]amino}ethyl]-8-methylquinolin-7-yl}-4-(cyclopropylmethoxy)benzamide (P-10h = (*S*)-10h).** White solid, mp 169−170 °C. $[\alpha]^{25}_D$ −52.60° (c 0.20, MeOH). Anal. ($C_{27}H_{32}N_4O_3$) C, H, N.

3-Bromo-*N*-[8-methyl-3-(1-pyrrolidin-1-ylethyl)quinolin-7-yl]benzamide (12a). A mixture of **LP-10a** (2.50 g, 5.82 mmol) and concentrated HCl (40 mL) was stirred at 100 °C for 16 h. After removal of the solvent under reduced pressure, the residue was dissolved in water and the mixture was basified with 10% $NaHCO_3$ under ice-cooling. The mixture was extracted with AcOEt by the salting out technique to give **11a** as a pale brown oil (1.33 g, 90%). To a mixture of **11a** (0.17 g, 0.65 mmol) and triethylamine (0.09 mL, 0.65 mmol) in THF (10 mL) was added 3-bromobenzoyl chloride (0.14 g, 0.65 mmol) at 0 °C. After being stirred at room temperature for 16 h, the reaction mixture was poured into water and the mixture was extracted with AcOEt. The extract was washed with 10% $NaHCO_3$ and brine and dried over $MgSO_4$. After removal of the solvent under reduced pressure, **12a** (0.23 g, 81%) was obtained as a pale yellow solid. This was recrystallized from hexane−AcOEt to give pale yellow crystals: mp 123−124 °C. $[\alpha]^{25}_D$ −34.65° (c 1.00, MeOH). ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.51 (3H, d, J = 6.6 Hz), 1.69−1.87 (4 H, m), 2.42 (2H, dd, J = 1.8, 6.9 Hz), 2.52−2.70 (2H, m), 2.82 (3H, s), 3.45 (1H, q, J = 6.6 Hz), 7.41 (1H, t, J = 7.9 Hz), 7.62−7.77 (2H, m), 7.81−7.95 (2H, m), 8.01−8.24 (3H, m), 8.94 (1H, d, J = 2.3 Hz). Anal. ($C_{23}H_{24}BrN_3O\cdot 1.0\ H_2O$) C, H, N.

X-ray Crystallography Analysis of 12a. Results are as follows: compound formula $C_{23}H_{24}BrN_3O\cdot H_2O$, M_r = 456.38, triclinic, $P1$, a = 7.4429(9) Å, b = 13.6860(12) Å, c = 20.469(2) Å, α = 90.422(3)°, β = 90.138(4)°, γ = 90.044(4)°, V = 2085.0(4) Å^3, Z = 4, D_{calc} = 1.454 g/cm^3, monochromatized radiation λ(Mo Kα) = 0.710 75 Å, μ = 2.002 mm^{-1}, F(000) = 944, T = 93 K. Data were collected on a Rigaku RAXIS RAPID imaging plate to a θ limit of 27.49° which yielded 16 908 reflections. There are 12 512 unique reflections with 8788 observed at the 2σ level. The structure was solved by direct methods (SIR92)[29] and refined using full-matrix least-squares on F^2 (SHELXL-97).[30] The final model was refined using 648 parameters and all 12 512 data. All non-hydrogen atoms were refined with isotropic thermal displacements. The final agreement statistics are as follows: R = 0.0788 (based on 8788 reflections with $I > 2\sigma(I)$), wR = 0.1856, S = 1.025. The maximum peak height in a final difference Fourier map is 0.975 e Å^3, and this peak is without chemical significance. The absolute configuration was determined based on the Flack parameter,[31] 0.038(13), refined using 4052 Friedel pairs. CCDC 859003 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request/cif.

3-Bromo-*N*-[8-methyl-3-(1-pyrrolidin-1-ylethyl)quinolin-7-yl]benzamide (12b). The title compound was prepared in 79% yield starting from **P-10a** using the procedure described for **12a**: mp 123−124 °C. $[\alpha]^{25}_D$ +34.65° (c 1.00, MeOH); ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.51 (3H, d, J = 6.6 Hz), 1.69−1.87 (4 H, m), 2.42 (2H, dd, J = 1.8, 6.9 Hz), 2.52−2.70 (2H, m), 2.82 (3H, s), 3.45 (1H, q, J = 6.6 Hz), 7.41 (1H, t, J = 7.9 Hz), 7.62−7.77 (2H, m), 7.81−7.95 (2H, m), 8.01−8.24 (3H, m), 8.94 (1H, d, J = 2.3 Hz).

HPLC Analysis of 12a and 12b. Column, Chiralpak AD-H FA031, 4.6 mm i.d. × 250 mmL; mobile phase, hexane/ethanol = 500/500 (v/v); flow rate of 1.0 mL/min; temperature 30 °C; detection, UV 254 nm; concentration, 0.1 mg/mL; injection, 0.010 mL. **12a**: >99.9% ee (tR1). **12b**: 99.9% ee (tR2).

4-(Cyclopropylmethoxy)-*N*-(3-formyl-8-methylquinolin-7-yl)benzamide (13). To an ice-cooled solution of cyclopropylmethoxybenzoic acid (20.0 g, 107 mmol) and DMF (0.40 mL, 5.37 mmol) in THF (350 mL) was added dropwise a solution of oxalyl chloride (10.1 mL, 118 mmol) in THF (50 mL). After being stirred at room temperature for 1 h, the reaction mixture was concentrated under reduced pressure and the resulting residue was dissolved in pyridine (200 mL). This solution was added to a solution of **4** in pyridine (200 mL) under ice-cooling and then stirred at room temperature for 16 h. The reaction mixture was diluted with THF (400 mL), and the mixture was filtered through a Celite pad. The filtrate was concentrated under reduced pressure and the resulting residue was chromatographed on silica gel (CH_2Cl_2, AcOEt) to give a yellow solid. This was triturated with Et_2O to give **13** (20.1 g, 55%) as a yellow powder. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.37−0.41 (2H, m), 0.66−

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0.73 (2H, m), 1.20−1.33 (1H, m), 2.86 (3H, s), 3.90 (2H, d, J = 6.9 Hz), 6.93 (1H, d, J = 9.0 Hz), 7.03 (2H, d, J = 9.3 Hz), 7.93 (2H, d, J = 9.3 Hz), 8.05 (1H, s), 8.57−8.60 (2H, m), 9.35 (1H, d, J = 2.1 Hz), 10.25 (1H, s).

4-(Cyclopropylmethoxy)-*N*-[3-(1-hydroxyethyl)-8-methylquinolin-7-yl]benzamide ((*rac*)-14). The title compound was prepared in 97% yield starting from **13** using the procedure described for **7**. ^{1}H NMR (300 MHz, DMSO-d_6) δ 0.36 (2H, td, J = 5.23, 4.24 Hz), 0.60 (2H, m), 1.27 (1H, m), 1.48 (3H, d, J = 6.41 Hz), 2.64 (3H, s), 3.92 (2H, d, J = 6.97 Hz), 5.00 (1H, dt, J = 9.80, 6.22 Hz), 5.49 (1H, d, J = 4.14 Hz), 7.07 (2H, d, J = 8.85 Hz), 7.59 (1H, d, J = 8.85 Hz), 7.81 (1H, d, J = 8.67 Hz), 8.02 (2H, d, J = 8.85 Hz), 8.23 (1H, d, J = 2.07 Hz), 8.94 (1H, d, J = 2.26 Hz), 10.06 (1H, s).

***N*-(3-Acetyl-8-methylquinolin-7-yl)-4-(cyclopropylmethoxy)-benzamide (15).** A solution of (*rac*)-**14** (10.0 g, 26.6 mmol) and MnO_2 (40.0 g, 460 mmol) in THF (1000 mL) was heated at reflux for 24 h. The reaction mixture was passed through a Celite and concentrated in vacuo. The residue was crystallized from AcOEt to give **15** (9.4 g, 94%) as a white powder. ^{1}H NMR (300 MHz, DMSO-d_6) δ 0.34−0.39 (2H, m), 0.57−0.63 (2H, m), 1.24−1.29 (1H, m), 2.68 (3H, s), 2.74 (3H, s), 3.93 (2H, d, J = 7.0 Hz), 7.08 (2H, d, J = 8.9 Hz), 7.78 (1H, d, J = 8.7 Hz), 8.02 (3H, d, J = 8.9 Hz), 9.00 (1H, d, J = 2.1 Hz), 9.35 (1H, d, J = 2.1 Hz), 10.13 (1H, s).

4-(Cyclopropylmethoxy)-*N*-{3-[(1*S*)-1-hydroxyethyl]-8-methylquinolin-7-yl}benzamide ((*S*)-14). In a stainless autoclave (1000 mL) that was filled with argon were added **15** (10.3 g, 27.5 mmol), potassium *tert*-butoxide (25 g, 220.0 mmol), and 2-propanol (200 mmol) in DMF (440 mL). After the mixture was stirred at room temperature for 30 min under an argon atmosphere, $RuCl_2$\{(*R*)-xylbinap\}\{(*R*)-daipen\} (33.6 mg, S/C = 1000) was added to the reaction mixture. The reduction was performed at room temperature under 0.7 MPa of hydrogen for 24 h. After the reaction was finished, hydrogen was released carefully and the solvent was removed by rotary evaporation. The crystals were washed with 1 M HCl (220 mL), water (300 mL) and dried in vacuo to provide brown crystals (10.4 g, quant). ^{1}H NMR (400 MHz, DMSO-d_6) δ: 0.35−0.38 (2H, m), 0.59−0.61 (2H, m), 1.25 (1H, m), 1.47 (3H, d, J = 6.3 Hz), 2.64 (3H, s), 3.92 (2H, d, J = 7.0 Hz), 4.98 (1H, q, J = 6.3 Hz), 7.06 (2H, d, J = 8.5 Hz), 7.58 (1H, d, J = 8.5 Hz), 7.80 (1H, d, J = 8.5 Hz), 8.01 (2H, d, J = 8.5 Hz), 8.22 (1H, d, J = 2.0 Hz), 8.93 (1H, d, J = 2.0 Hz), 10.04 (1H, s). $[\alpha]_D^{25}$ −35.60° (c 0.20, DMF). 98% ee by HPLC (Chiralcel OD-RH, H_2O/CH_3CN = 60/40, 1.0 mL/min, 35 °C).

***N*-{3-[(1*R*)-1-Azidoethyl]-8-methylquinolin-7-yl}-4-(cyclopropylmethoxy)benzamide (16).** DPPA (34 mL, 100 mmol) was added to a mixture of (*S*)-**14** (9.9 g, 26.3 mmol, 98.0% ee) and *N*, *N*-diisoproylethylamine (300 mL) at −40 °C. The reaction mixture was stirred at room temperature for 4 days. Water (200 mL) was added to the reaction mixture, and the solution was extracted with AcOEt (500 mL). The extracts were washed with brine and dried ($MgSO_4$). The solvent was evaporated in vacuo, and the residue was passed through a silica gel plug, eluting with AcOEt/hexane = 1/3. Solvent was removed under vacuum to yield the chiral azide **16** as yellow crystals (9.5 g, 90% yield). ^{1}H NMR (400 MHz, $CDCl_3$) δ: 0.38−0.42 (2H, m), 0.67−0.70 (2H, m), 1.31 (1H, m), 1.68 (3H, d, J = 6.8 Hz), 2.82 (3H, s), 3.89 (2H, d, J = 7.1 Hz), 4.86 (1H, q, J = 6.8 Hz), 7.02 (2H, d, J = 8.8 Hz), 7.74 (1H, d, J = 8.8 Hz), 7.91−7.93 (3H, m), 8.06 (1H, d, J = 2.2 Hz), 8.34 (1H, d, J = 8.8 Hz), 8.90 (1H, d, J = 2.2 Hz). $[\alpha]_D^{25}$ +71.30° (c 0.20, $CHCl_3$). 98% ee by HPLC (Chiralcel OJ-RH, 0.1 M KPF_6/CH_3CN = 50/50, 0.6 mL/min, 30 °C).

***N*-{3-[(1*R*)-1-Aminoethyl]-8-methylquinolin-7-yl}-4-(cyclopropylmethoxy)benzamide (17).** A mixture of **16** (6.70 g, 16.7 mmol, 98% ee) and palladium−carbon (1.70 g, 5%) in ethanol (1100 mL) was stirred at room temperature under hydrogen atmosphere for 2 h. After the reaction was finished, hydrogen was released carefully and the reaction mixture was passed through Celite, eluting with methanol. Solvent was removed under vacuum to yield **17** as pale yellow crystals (6.27 g, quant). ^{1}H NMR (400 MHz, $CDCl_3$) δ 0.38−0.42 (2H, m), 0.67−0.70 (2H, m), 1.29 (1H, m), 1.51 (3H, d, J = 6.8 Hz), 2.81 (3H, s), 3.89 (2H, d, J = 6.8 Hz), 4.37 (1H, q, J = 6.8 Hz), 7.01 (2H, d, J = 8.8 Hz), 7.71 (1H, d, J = 8.8 Hz), 7.89−7.93 (3H, m), 8.08 (1H, d, J = 2.0 Hz), 8.26 (1H, d, J = 8.8 Hz), 8.93 (1H, d, J = 2.0 Hz). $[\alpha]_D^{25}$ +26.50° (c 0.20, $CHCl_3$). 98% ee by HPLC (SumiChiral OA 8000, *n*-hexane/EtOH/CF_3COOH = 750/25/1, 1.2 mL/min, 35 °C).

4-(Cyclopropylmethoxy)-*N*-{8-methyl-3-[(1*R*)-1-{[(2-nitrophenyl)sulfonyl]amino}ethyl]quinolin-7-yl}benzamide (18). A solution of 2-nitrobenzenesulfonyl chloride (7.15 g, 32.3 mmol) in THF (100 mL) was added to a solution of **17** (>99.9% ee, 10.1 g, 26.9 mmol) and triethylamine (5.6 mL, 40.2 mmol) in THF (300 mL) at 0 °C. The mixture was stirred at room temperature for 12 h. The reaction mixture was concentrated in vacuo, and the residue was partitioned between AcOEt and water. The mixture was washed with brine, dried over Na_2SO_4, filtered, and concentrated under reduced pressure. The residue was purified by recrystallization from AcOEt to obtain **18** (14.6 g, 26.0 mmol, 97%) as white crystals. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.36−0.44 (2H, m), 0.65−0.74 (2H, m), 1.26−1.36 (1H, m), 1.67 (3H, d, J = 7.2 Hz), 2.71 (3H, s), 3.90 (2H, d, J = 6.8 Hz), 4.92 (1H, quint, J = 7.2 Hz), 5.89 (1H, d, J = 8.3 Hz), 7.02 (2H, d, J = 8.7 Hz), 7.17 (1H, td, J = 7.8, 1.1 Hz), 7.40 (1H, td, J = 7.8, 1.5 Hz), 7.53 (1H, d, J = 8.7 Hz), 7.59 (1H, dd, J = 7.8, 1.3 Hz), 7.67 (1H, dd, J = 8.0, 1.1 Hz), 7.87 (1H, d, J = 2.7 Hz), 7.88 (1H, s), 7.91 (2H, d, J = 9.1 Hz), 8.23 (1H, d, J = 9.1 Hz), 8.70 (1H, d, J = 2.3 Hz).

***N*-{3-[(1*R*)-1-{(2-Aminoethyl)[(2-nitrophenyl)sulfonyl]-amino}ethyl]-8-methylquinolin-7-yl}-4-(cyclopropylmethoxy)-benzamide (19).** A solution of *tert*-butyl *N*-(2-hydroxyethyl)-carbamate (28.8 g, 179 mmol) in THF (65 mL) was added to a suspension of **18** (50.0 g, 89.2 mmol) and triphenylphosphine (46.8 g, 178 mmol) in THF (850 mL) at 0 °C. A solution of diisopropyl azodicarboxylate (DIAD, 90%, 40.1 g, 178 mmol) in THF (65 mL) was added at 0 °C for 45 min. The mixture was stirred at room temperature for 2.5 h. A solution of 4 M HCl in AcOEt (334 mL) was added to the above reaction mixture at 0 °C for 15 min. The mixture was stirred at room temperature for 15 h. The reaction mixture was concentrated under reduced pressure. AcOEt was added. The mixture was extracted with 2 M HCl (aq)/DMSO = 5/3. The aqueous layers were treated with sodium carbonate to adjust the pH to 9. The mixture was extracted with AcOEt. The organic layer was washed with brine, dried over Na_2SO_4, filtered, and concentrated under reduced pressure to obtain **19** (63.9 g) as pale yellow foam. This foam was used for the next step without further purification. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.36−0.44 (2H, m), 0.65−0.74 (2H, m), 1.27−1.38 (3H, m), 1.70 (3H, d, J = 6.8 Hz), 2.44 (1H, dt, J = 12.9, 7.2 Hz), 2.64 (1H, dt, J = 13.1, 7.2 Hz), 2.78 (3H, s), 3.29 (2H, t, J = 7.2 Hz), 3.90 (2H, d, J = 7.2 Hz), 5.49 (1H, q, J = 7.2 Hz), 7.02 (2H, d, J = 8.7 Hz), 7.62−7.76 (4H, m), 7.92 (2H, d, J = 8.7 Hz), 7.92 (1H, s), 8.02 (1H, d, J = 1.9 Hz), 8.05−8.11 (1H, m), 8.31 (1H, d, J = 9.1 Hz), 8.90 (1H, d, J = 2.3 Hz).

***N*-{3-[(1*R*)-1-{[2-(Acetylamino)ethyl]amino}ethyl]-8-methylquinolin-7-yl}-4-(cyclopropylmethoxy)benzamide ((*R*)-10h).** A solution of acetyl chloride (10.5 g, 134 mmol) in THF (30 mL) was added to a solution of **19** (63.9 g, all amounts from above-mentioned reaction) and triethylamine (18.6 mL, 133 mmol) in THF (450 mL) at 0 °C. The mixture was stirred at 0 °C for 10 min. AcOEt was added. The mixture was washed with water, Na_2CO_3 aqueous solution, and brine. The organic layer was dried over Na_2SO_4, filtered through a silica gel plug, and concentrated under reduced pressure to obtain acetamide intermediate as a yellow foam (61.0 g). The above acetamide intermediate (all amount) was dissolved with DMF (250 mL). Lithium hydroxide monohydrate (29.9 g, 713 mmol) was added. A solution of mercaptoacetic acid (90%, 36.5 g, 357 mmol) in DMF (40 mL) was added dropwise at 0 °C over 10 min. The mixture was stirred at room temperature for 4 h. AcOEt was added. The mixture was washed with 2.5% $NaHCO_3$ aqueous solution and brine. The organic layer was dried over Na_2SO_4, filtered, and concentrated under reduced pressure. The residue was purified by recrystallization from AcOEt to obtain (*R*)-**10h** (39.2 g, 85.1 mmol, 95% from **18**) as white crystals, 99.6% ee: mp 169−170 °C. $[\alpha]_D^{25}$ +52.6° (c 0.1980 in MeOH). ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.36−0.43 (2H, m), 0.65−

0.73 (2H, m), 1.26−1.36 (1H, m), 1.48 (3H, d, J = 6.8 Hz), 1.97 (3H, s), 2.51−2.62 (1H, m), 2.71−2.82 (1H, m), 2.81 (3H, s), 3.31 (2H, q, J = 6.0 Hz), 3.90 (2H, d, J = 7.0 Hz), 4.00 (1H, q, J = 6.6 Hz), 5.86 (1H, s), 7.02 (2H, d, J = 8.9 Hz), 7.70 (1H, d, J = 8.9 Hz), 7.91 (1H, s), 7.92 (2H, d, J = 8.9 Hz), 7.99 (1H, d, J = 2.3 Hz), 8.27 (1H, d, J = 8.9 Hz), 8.91 (1H, d, J = 2.3 Hz). Anal. Calcd for $C_{27}H_{32}N_4O_3 \cdot 0.1H_2O$: C, 70.14; H, 7.02; N, 12.12. Found: C, 69.98; H, 7.17; N, 12.08.

Methyl 7-{[4-(Cyclopropylmethoxy)benzoyl]amino}-8-methylquinoline-3-carboxylate (20). To a solution of **13** (3.75 g, 10.4 mmol) in MeOH (200 mL) was added NIS (6.0 g, 26.9 mmol) and K_2CO_3 (3.7 g, 26.9 mmol). The resulting dark mixture was stirred for 16 h, at which time LC/MS analysis indicated complete consumption of starting material. Water (5 mL) and $Na_2S_2O_3 \cdot 5H_2O$ (5.0 g) were added to destroy remaining NIS or hypoiodite species. The resulting mixture was extracted with Et_2O−hexane (1:1). The combined organic extract was washed with brine (50 mL), and the solvent was removed under reduced pressure to give **20** (3.92 g, quant) as a yellow powder. This compound was used for the next reaction without further purification or identification.

4-(Cyclopropylmethoxy)-*N*-[3-(1-hydroxy-1-methylethyl)-8-methylquinolin-7-yl]benzamide (21). To a solution of methyllithium (1.0 M ethereal solution, 50 mL, 50 mmol) in THF (300 mL) was added **20** (3.92 g, 10.4 mmol) at 0 °C. The reaction mixture was stirred at room temperature for 2 h. The reaction mixture was partitioned between AcOEt and water. The AcOEt layer was washed with brine and dried over $MgSO_4$. The solution was concentrated under reduced pressure to afford **21** (4.06 g, quant) as a pale brown powder. This compound was used for the next reaction without further purification or identification.

4-(Cyclopropylmethoxy)-*N*-[8-methyl-3-(1-methyl-1-pyrrolidin-1-ylethyl)quinolin-7-yl]benzamide (22). A mixture of **21** (4.06 g, 10.4 mmol) and thionyl chloride (30 mL) was stirred at 0 °C for 30 min and at room temperature for 2 h. After removal of the excess amount of thionyl chloride under reduced pressure, to the resulting residue was added pyrrolidine (30 mL). The mixture was stirred at 60 °C for 3 h. The reaction mixture was partitioned between AcOEt and water. The AcOEt layer was washed with brine and concentrated under reduced pressure. The resulting residue was chromatographed on silica gel (AcOEt/MeOH = 5/1) to give **22** (1.20 g, 27%) as a pale brown powder: mp 161−163 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.37 (2H, m), 0.68 (2H, m), 1.32 (1H, m), 1.54 (6H, s), 1.72 (4H, m), 2.01 (3H, s), 2.56 (4H, m), 3.89 (2H, d, J = 6.9 Hz), 7.00 (2H, d, J = 8.4 Hz), 7.25 (1H, s), 7.69 (1H, d, J = 8.7 Hz), 7.90 (2H, m), 8.07 (1H, s), 8.23 (1H, d, J = 8.7 Hz), 9.19 (1H, d, J = 2.1 Hz). Anal. ($C_{28}H_{33}N_3O_2 \cdot 0.3H_2O$) C, H, N.

Measurement of Binding Affinities. The MCHR1 binding assays were based on the method of Takekawa et al.[14b] with minor modification. CHO cells stably expressing human MCHR1 and rat MCHR1 were prepared.[2,14b] The frozen cell homogenate was thawed, suspended in assay buffer (25 mM Tris-HCl, 1 mM EDTA, 0.1% BSA, 0.5 mM phenylmethylsulfonyl fluoride, 1 μg/mL pepstatin A, 10 μg/mL phosphoramidon, and 20 μg/mL leupeptine, pH 7.5), and used for the binding assay. The membrane fraction was diluted to 2 μg/mL (human MCHR1) or 4 μg/mL (rat MCHR1) with the assay buffer, and then 175 μL/well aliquot was dispensed into polypropylene 96-well plate (type 3363, Corning). Then 2 μL of DMSO solution of a test compound was mixed with homogenates and [^{125}I]MCH (4−19) peptide in a total volume of 200 μL/well and incubated at room temperature for 60 min. The binding reaction was terminated by rapid filtration using FilterMate harvester (PerkinElmer) followed by three 300 μL/well washes with 50 mM Tris-HCl buffer (pH 7.5). Nonspecific binding was defined in the presence of 0.3 μM MCH (1−19) peptide. GF/C filter plates were dried, and radioactivity was determined after addition of 25 μL/well Microscint-0 (PerkinElmer) using TopCount liquid scintillation counter (PerkinElmer). MCH (4−19) peptide and MCH (1−19) peptide were purchased from Peptide Institute. MCH (4−19) peptide was labeled with ^{125}I by the Bolton−Hunter method. The 50% inhibitory concentration (IC_{50}) was calculated by nonlinear logistic regression analysis in GraphPad Prism software (GraphPad Software Inc.).

Measurement of Arachidonic Acid Release. CHO cells expressing the human MCHR1 were plated in 24-well plates at a density of 50 000 cells/well and cultured for 1 day. The cells were incubated with [^{3}H]arachidonic acid (0.2 μCi/well) for 16 h and washed twice with 500 μL of Dulbecco's modified Eagle's medium (DMEM) supplemented with 20 mM *N*-(2-hydroxyethyl)piperazine-*N*′-ethanesulfonic acid (HEPES) (pH 7.4) and 0.2% bovine serum albumin. The cells were then preincubated with compounds at various concentrations at 37 °C for 30 min, and the reaction was started by addition of MCH. After incubation for 45 min, the radioactivity in the medium was measured with a liquid scintillation counter.

Rubidium (Rb^+) Efflux Assay. HMZ/CHO.8B4 cells, which stably expressed the hERG K^+ channel, were established by Takeda Pharmaceutical Company. HMZ/CHO.8B4 cells were cultured in F-12 nutrient mixture [Ham] (Invitrogen, Carlsbad, CA) containing 10% fetal bovine serum (FBS; Trace Scientific Ltd., Melbourne, Australia), 500 μg/mL Geneticin (Invitrogen) in a humidified atmosphere in 5% CO_2 at 37 °C. For the Rb^+ efflux assay, HMZ/CHO.8B4 cells (4×10^4 cells/well) were seeded on a collagen-coated 96-well assay plate (Becton Dickinson, Billerica, MA). After 24 h culture, assay plates were washed with phosphate-buffered saline to remove supplemented medium. Cells were then incubated (37 °C in 5% CO_2) with Rb-loading buffer containing the following: 150 mM NaCl, 5.4 mM RbCl, 150 mM NaCl, 2 mM $CaCl_2$, 0.8 mM NaH_2PO_4, 1 mM $MgCl_2$, 5 mM glucose, 25 mM HEPES, 20% FBS, pH 7.4. After 3 h of incubation, each well was spiked to give a test compound and incubated for an additional 30 min (n = 7). Wells were then washed with minimum essential medium (MEM, Invitrogen) containing 10 mM HEPES to remove excess Rb loading buffer. To activate the opening of hERG channels, cells were stimulated with buffer containing MEM containing 50 mM KCl and 10 mM HEPES for an additional 5 min. Finally, the supernatant was collected in a separate assay plate, while cells were lysed with the addition 1% Triton X-100 to each well. To determine fractional Rb^+ efflux, the supernatant and lysate from each experiment were run separately through the ICR8000 flame atomic absorption spectrometer (Aurora Biomed Inc., Vancouver, BC, Canada). Rb^+ efflux was represented as the ratio of the Rb^+ content of the supernatant with respect to the total Rb^+ in each well. Each sample was measured twice. Data were then normalized to account for variation in total Rb^+ efflux between multiple experiments using the following equation: remaining acivity = $([Rb^+]_{sample} - [Rb^+]_{bkgd})/([Rb^+]_{max} - [Rb^+]_{bkgd})$, where $[Rb^+]_{sample}$ is the fractional efflux, $[Rb^+]_{bkgd}$ is the unstimulated efflux, and $[Rb^+]_{max}$ is the maximum efflux.

In Vivo Pharmacological Study (2-Day Assay). All animal experiments were performed in compliance with the Guidelines for the Care and Use of Laboratory Animals of Takeda Pharmaceutical Company.

Male F344/Jcl rats (32-week-old, CLEA Japan, Inc.) loaded with a high-fat diet (Research Diets, Inc., D12451) from 5 weeks of age were used (DIO-F344 rats). Before the start of experiment, the rats were independently raised (light cycle 7:00 a.m. to 7:00 p.m.), were allowed access to a powder high-fat diet (D12451M, Research Diets) and tap water ad libitum, and were administered tap water (0.5 mL) orally for acclimation to oral dosing. Thereby the rats were habituated. The food intake from evening of the day before the start of experiment to morning of the next day was measured, and the rats were grouped based on the food intake and the body weight of the previous day as indices (mean body weight of 438.7 g, n = 6 for each group). On the day of the start of experiment and the next day at 5:00 p.m., after body weight measurement, 0.5% methylcellulose solution (vehicle) was administered orally to the control group, and 0.5% methylcellulose suspension (1, 3, 5, and 10 mg/kg) of the compound was administered orally to the compound administration group at 2 mL/kg based on the body weight of each day (volume range actually used was 0.79−0.94 mL). The food intake from the initial administration to 1 day and 2 days later was measured manually. The food intake inhibition rate of each compound administration group to the control group was

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calculated. Food intake data were analyzed by Williams test, and values of $P < 0.025$ were considered statistically significant.

■ ASSOCIATED CONTENT

Supporting Information

Synthesis of starting materials, elemental analysis results of final compounds, MCHR1 Ca^{2+} influx assays for (*R*)-**10a** and (*R*)-**10h**, binding of (*R*)-**10a** to wild type, monomer mutant (Y652A) and tandem dimer mutant (Y652A) hERG, and effect of (*R*)-**10h** on 2-day food intake study in DIO-F-344 rats. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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■ ABBREVIATIONS USED

MCH, melanin-concentrating hormone; hERG, human ether-a-go-go-related gene; DIO, diet-induced obesity; SAR, structure−activity relationship; *S*/*C*, substrate-to-catalyst molar ratio; DPPA, diphenylphosphorylazide; DIPEA, *N*,*N*-diisopropylethylamine; NIS, *N*-iodosuccinimide; CHO, Chinese hamster ovary; DIAD, diisopropyl azodicarboxylate

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