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Design, synthesis and structure–activity relationship of novel tricyclic benzimidazolone derivatives as potent 18 kDa translocator protein (TSPO) ligands

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ABSTRACT

The 18 kDa translocator protein (TSPO) was identified as a discrete receptor for diazepam (**1**). Since TSPO in the central nervous system (CNS) is believed to regulate neurosteroids biosynthesis, selective TSPO ligands are expected to be useful in the treatment of psychiatric disorders. We synthesized three novel tricyclic benzimidazolone derivatives, and selected the dihydroimidazoquinolinone derivative **27** as a lead TSPO ligand. Study of the structure–activity relationship (SAR) of dihydroimidazoquinolinone derivatives revealed compounds with potent affinity for TSPO (subnanomolar K_i values), but poor metabolic stability. The optimization of these compounds led to compound **48** with potent affinity for TSPO and good *in vitro* PK profile.

1. Introduction

The 18 kDa translocator protein (TSPO) was identified in 1977 as a discrete receptor for diazepam (**1**).[1] TSPO was previously called the peripheral benzodiazepine receptor (PBR), but was renamed by Papadopoulous and colleagues in 2006 as TSPO.[2] TSPO, which forms a heterotrimeric complex with the 32 kDa voltage-dependent anion channel (VDAC) and the 30 kDa adenine nucleotide transporter (ANT)[3] is mainly distributed on the outer mitochondrial membrane in peripheral tissues and the central nervous system (CNS).[4] Although the physiological role of TSPO in CNS is not clear, a study on the function of TSPO suggested that TSPO regulates cholesterol transport from the outer to the inner mitochondrial membrane,[5] which is known as the rate-determining step of neurosteroids biosynthesis.[6] As evidence has shown that neurosteroids play a role in psychiatric disorders,[7] it is believed that TSPO ligands might be useful in the treatment of these disorders.

As shown in Figure 1, in addition to the classical TSPO ligands Ro5-4864 (**2**)[8] and PK11195 (**3**),[9] several other TSPO ligands have been reported, including the 8-oxopurine derivative AC-5216 (**4**)[10] and indoleacetamide derivative SSR180575 (**5**).[11] We have recently reported a series of benzoxazolone derivatives, including compound **6** as potent and selective TSPO ligands.[12] Structure–activity relationship (SAR) studies of different substitutents at the benzoxazolone ring revealed that the 7-Ph derivative **7** exhibits moderate affinity for TSPO (K_i = 9.0 nM). As part of our search for compounds with various heteroaryls, we obtained interesting results in the benzimidazolone derivatives.[13] Compound **8** exhibited moderate affinity for TSPO (86% inhibition at 100 nM), and introduction of a methyl group at the N^3 position (**9**) increased this affinity (100% inhibition at 100 nM). Considering the results

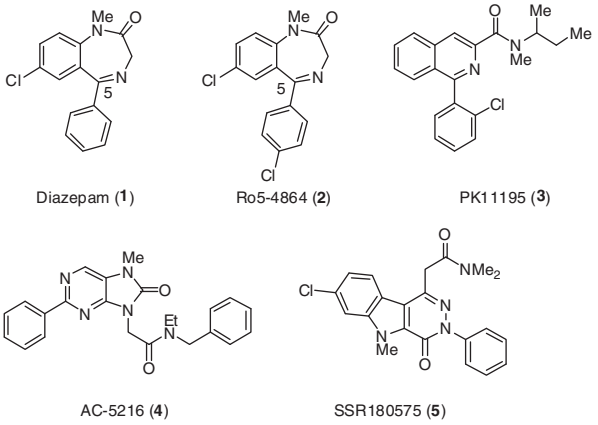

Figure 1. Chemical structures of selected TSPO ligands.

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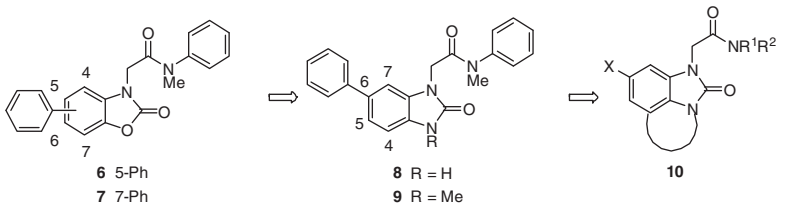
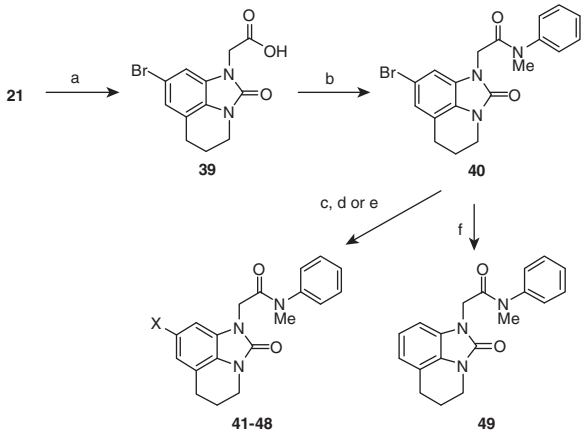


Figure 2. Design of novel TSPO ligands.

obtained with compounds **7** and **9**, introduction of other substitutes at the C-4 position and the N^3 position of the benzimidazolone ring was expected to be tolerated in relation to TSPO activity. As the benzoxazolone ring in compound **6** plays an important role as a planar aromatic region for the interaction with TSPO,[14] conversion of the benzoxazolone skeleton to a tricyclic benzimidazolone ring which expands the planar region was considered as favorable for enhanced interaction with TSPO. Based on these exploratory findings, we have designed tricyclic benzimidazolone derivatives as novel TSPO ligands (**10**). Here we present the SAR of a series of dihydroimidazoquinolinone, dihydroimidazobenzazepinone and dihydroimidazobenzoxazinone derivatives as novel TSPO ligands (see Fig. 2).

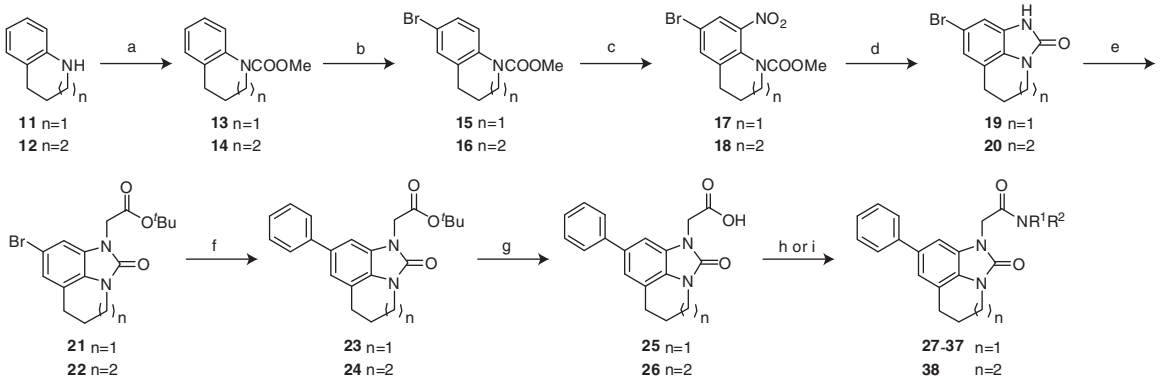
2. Chemistry

The compounds in this study were synthesized as illustrated in Schemes 1–3. Scheme 1 shows the preparation of 8-phenyl dihydroimidazoquinolinone and 9-phenyl dihydroimidazobezazepinone derivatives. Methoxycarbonylation of 1,2,3,4-tetrahydroquinoline (**11**) with methyl chloroformate afforded the *N*-carbamate-protected 1,2,3,4-tetrahydroquinoline **13**. Bromination of **13** with *N*-bromosuccinimide (NBS) in DMF proceeded to give the expected 6-bromo derivative **15**. Subsequent regioselective nitration of **15** was effected by action of NO_2BF_4 in CH_3CN to give compound **17**. Reductive cyclization of **17** was achieved by treatment with iron powder in AcOH at 80 °C to provide compound **19**. Introduction of an acetate moiety was achieved by *N*-alkylation of **19** with *t*-butyl bromoacetate to afford acetate **21**, which was subjected to Suzuki–Miyaura coupling with phenylboronic acid to give compound **23**. Deprotection of compound **23** with HCl provided the key intermediary acid derivative **25**. Condensation of **25** with appropriate amines was carried out by combination of 1-ethyl-3-(3-dimethylaminopropyl)-carbodiimide hydrochloride (EDCI) and 1-hydroxybenzotriazole (HOBt) or by the standard acid chloride method using oxalyl chloride to afford the corresponding amide derivatives **27**–**37**. The 9-Ph dihydroimidazobenzazepinone derivative **38** was obtained from 2,3,4,5-tetrahydro-1*H*-1-benzazepine (**12**) in the same way as that for synthesis of **27**. The chemical structures of compounds **27**–**38** are shown in Tables 1 and 2.

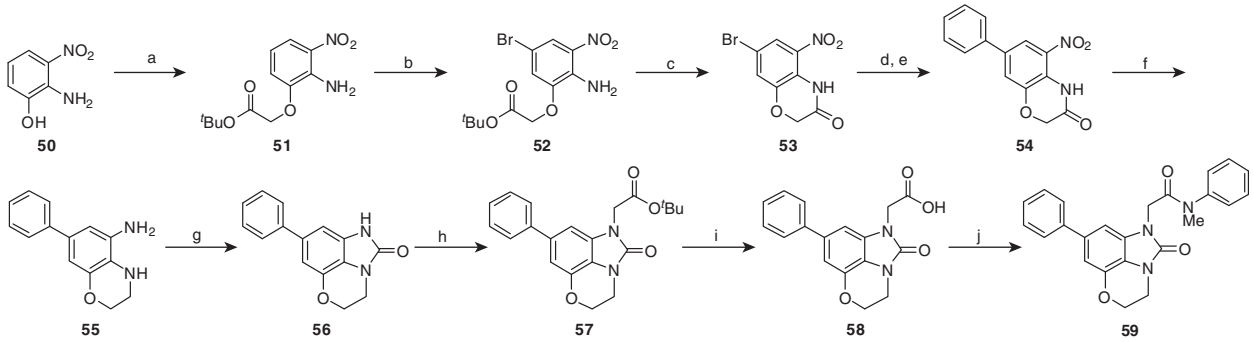


Scheme 2. Synthesis of the dihydroimidazoquinolinone derivatives **41**–**49**. Reagents and conditions: (a) HCl/1,4-dioxane, AcOH, 50 °C; (b) *N*-methylaniline, EDCI, HOBt, DMF, rt; (c) $ArB(OH)_2$, $Pd(PPh_3)_4$, K_2CO_3, 1,4-dioxane, H_2O, reflux; (d) $ArSnBu_3$, $Pd(PPh_3)_4$, toluene, reflux; (e) 3-aminopyridine, $Pd_2(dba)_3$, Xantphos, Cs_2CO_3, toluene, reflux; (f) H_2, 10% Pd-C, MeOH, THF, rt.

Scheme 2 depicts the synthesis of a series of dihydroimidazoquinolinone derivatives with various substituents at the C-8 position. Deprotection of **21** with HCl followed by condensation of **39** with *N*-methylaniline in the presence of EDCI and HOBt in DMF gave compound **40**. The target compounds **41**–**47** were prepared via Suzuki–Miyaura coupling or Stille coupling of **40** with boronic acid or an organotin reagent. Compound **48** was obtained by coupling reaction with 3-aminopyridine in the presence of $Pd_2(dba)_3$ and 4,5-bis(diphenylphosphino)-9,9-dimethylxanthene (Xantphos) using Cs_2CO_3 as a base in toluene.[15] The bromine atom in compound

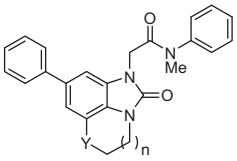


Scheme 1. Synthesis of **27**–**38**. Reagents and conditions: (a) methyl chloroformate, K_2CO_3, DMF, 50 °C; (b) NBS, DMF, rt; (c) NO_2BF_4, CH_3CN, 0 °C; (d) Fe, AcOH, 80 °C; (e) *t*-butyl bromoacetate, K_2CO_3, DMF, 50 °C; (f) $PhB(OH)_2$, $Pd(PPh_3)_4$, K_2CO_3, 1,4-dioxane, H_2O, reflux; (g) HCl/1,4-dioxane, AcOH, 55 °C; (h) amine, EDCI, HOBt, DMF, rt; (i) $(COCl)_2$, DMF, CH_2Cl_2, then amine, Et_3N, THF, rt.



Scheme 3. Synthesis of the dihydroimidazobenzoxazinone derivative **59**. Reagents and conditions: (a) *t*-butyl bromoacetate, K_2CO_3, DMF, rt; (b) NBS, DMF, rt; (c) *p*-TsOH·H_2O, toluene, 80 °C; (d) $PhB(OH)_2$, $Pd(PPh_3)_4$, K_2CO_3, 1,4-dioxane, H_2O, reflux; (e) *p*-TsOH·H_2O, toluene, 80 °C; (f) $LiAlH_4$, THF, rt; (g) CDI, THF, reflux; (h) *t*-butyl bromoacetate, K_2CO_3, DMF, 70 °C; (i) HCl/1,4-dioxane, AcOH, 65 °C; (j) *N*-methylaniline, EDCI, HOBt, DMF, rt.

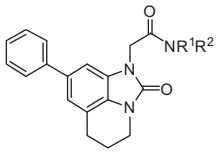
Table 1
TSPO and CBR binding affinity, and metabolic stability of dihydroimidazoquinolinone, dihydroimidazobenzazepinone and dihydroimidazobenzoxazinone derivatives



Compd	*n*	Y	TSPO K_i[a] (nM)	CBR inhibition[b] (%)	Metabolic stability[c] (remaining %)
PK11195 (**3**)			1.7		
6			1.6	5	1
27	1	CH_2	0.94	14	12
38	2	CH_2	0.32	30	1
59	1	O	0.21	68	1

[a] K_i Values represent the means of 1–3 separate experiments run in duplicate using four concentrations of each compound.
[b] Percent inhibition of [^{3}H]-flumazenil specific binding at 10 μM of the compound.
[c] Metabolic stability data refer to percent of compound remaining after incubation with rat liver S-9 fraction (ca. 2.0 mgprotein/ml) and NADPH (ca. 3.0 mM) for 30 min at 37 °C. The initial concentration of each compound was 1.0 μM.

Table 2
TSPO and CBR binding affinity, and metabolic stability of dihydroimidazoquinolinone derivatives with various amide moieties



Compd	R^1	R^2	TSPO K_i[a] (nM)	CBR inhibition[b] (%)	Metabolic stability[c] (remaining %)
27	Me	Ph	0.94	14	12
28	*n*-Pr	*n*-Pr	1.7	11	1
29	Me	Bn	0.23	72	1
30	Me	*m*-MeO-Ph	0.28	6	0
31	Me	*p*-MeO-Ph	0.73	3	0[d]
32	Me	*m*-Cl-Ph	0.44	3	0
33	Me	*p*-Cl-Ph	0.13	4	0
34	Me	2-Py	2.5	34	0
35	Me	3-Py	2.1	36	0
36	Me	4-Py	15	24	0
37	H	Ph	33	27	62

[a] K_i Values represent the means of 1–3 separate experiments run in duplicate using four concentrations of each compound.
[b] Percent inhibition of [^{3}H]-flumazenil specific binding at 10 μM of the compound.
[c] Metabolic stability data refer to percent of compound remaining after incubation with rat liver S-9 fraction (ca. 2.0 mgprotein/ml) and NADPH (ca. 3.0 mM) for 30 min at 37 °C. The initial concentration of each compound was 1.0 μM.
[d] Metabolic stability determined at 10 μM of the compound.

40 was removed by standard hydrogenation over Pd-C to give compound **49**. The chemical structures of compounds **41**–**48** are shown in Table 3.

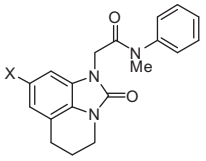
Synthesis of the dihydroimidazobenzoxazinone derivative **59** was achieved as illustrated in Scheme 3. *O*-Alkylation of 2-amino-3-nitrophenol (**50**) with *t*-butyl bromoacetate using K_2CO_3 as a base in DMF followed by bromination of **51** with NBS in DMF afforded compound **52**. Ring closure in **52** was realized with *p*-toluenesulfonic acid in toluene at 80 °C to give compound **53**. Opening of the oxazinone ring was occurred under basic condition by Suzuki–Miyaura coupling of **53** with phenylboronic acid to give [(4-amino-5-nitrobiphenyl-3-yl)oxy]acetic acid, which was again subjected to acidic condition with *p*-toluenesulfonic acid in toluene at 80 °C to afford the desired compound **54**. Reduction of the nitro group and the amide moiety in **54** was simultaneously achieved with $LiAlH_4$ to give compound **55**. Cyclization of **55** by 1,1′-carbonyldiimidazole (CDI) generated the dihydroimidazoquinolinone scaffold **56**. The target compound **59** was obtained from **56** by a method similar to that for the synthesis of **40**.

3. Results and discussion

Affinity of the prepared compounds for TSPO and the central benzodiazepine receptor (CBR) was evaluated by measuring each compound's ability to displace [^{3}H]-PK11195 and [^{3}H]-flumazenil binding to membranes prepared from rat kidney and rat cerebral cortex, respectively. Most of the synthesized compounds showed high selectivity for TSPO over CBR with only few compounds exhibiting more than 50% inhibition of CBR at 10 μM.

As shown in Table 1, all three tricyclic benzimidazolone derivatives were very potent TSPO ligands, showing subnanomolar K_i values. The dihydroimidazoquinolinone derivative **27** exhibited stronger TSPO activity than that of the benzoxazolone derivative **6**. As expected, this finding indicates that the expansion of the planar region strengthens interaction with the TSPO recognition site. Further expansion of the ring to a seven-membered ring, as in the dihydroimidazobenzazepinone derivative **38**, led to a threefold increase in TSPO affinity compared to that of **27**, albeit with poor metabolic stability. In an attempt to improve metabolic stability, we prepared the dihydroimidazobenzoxazinone derivative **59** by replacing methylene chain at the C-6 position of **27** by an oxygen atom. Although compound **59** showed the highest affinity for TSPO among all three tricyclic derivatives (K_i = 0.21 nM), it had low metabolic stability. Taking all these results into account, compound **27**,

Table 3
TSPO and CBR binding affinity, and metabolic stability of dihydroimidazoquinolinone derivatives with various substituents at the C-8 position



Compd	X	TSPO K_i[a] (nM)	CBR inhibition[b] (%)	Metabolic stability[c] (remaining %)
49	H	2.5	0	0[d]
40	Br	0.057	5	0[d]
27	Ph	0.94	14	12
41	*m*-MeO-Ph	0.11	9	1
42	*p*-MeO-Ph	0.30	5	2[d]
43	*m*-CF_3-Ph	0.23	3	0
44	*p*-CF_3-Ph	0.16	0	0
45	2-Py	2.7	48	0
46	3-Py	3.3	47	0[d]
47	4-Py	0.55	41	3
48	3-PyNH	2.0	70	41

[a] K_i Values represent the means of 1–3 separate experiments run in duplicate using four concentrations of each compound.
[b] Percent inhibition of [^{3}H]-flumazenil specific binding at 10 μM of the compound.
[c] Metabolic stability data refer to percent of compound remaining after incubation with rat liver S-9 fraction (ca. 2.0 mg protein/ml) and NADPH (ca. 3.0 mM) for 30 min at 37 °C. The initial concentration of each compound was 1.0 μM.
[d] Metabolic stability determined at 10 μM of the compound.

which showed high selectivity for TSPO over CBR and moderate metabolic stability, was selected as a lead compound for further evaluation.

In our investigation of the amide part of the dihydroimidazoquinolinone derivatives, we kept the phenyl ring at the C-8 position in the dihydroimidazoquinolinone core and explored various substituents on the acetamide nitrogen. TSPO affinity and metabolic stability of the tested compounds are shown in Table 2. First of all, we converted R^1 and R^2 from an alkyl–phenyl to an alkyl–alkyl and alkyl–benzyl, respectively. Compounds **28** ($R^1 = R^2 = {}^n$Pr) and **29** (R^1 = Me, R^2 = Bn) exhibited potent affinity for TSPO. In particular, compound **29** showed fourfold more potent activity than that of **27**. Although compounds **28** and **29** showed excellent activity, their metabolic stability was low. We therefore decided to examine the effect of a substitution at the phenyl ring (R^2) of **27**. Introduction of a methoxy group (**30**, **31**) or a chlorine atom (**32**, **33**) onto the phenyl ring resulted in better activity. In particular, incorporation of a methoxy group into the *meta*-position and a chlorine atom into the *para*-position provided significant increase in TSPO affinity with compounds **30** and **33** showing K_i values of 0.28 and 0.13 nM, respectively. Next we examined the effect of replacing the phenyl ring with a pyridine analogue. Although compound **36** was found to be 16-fold less active than compound **27**, compounds **34** and **35** exhibited potent affinity for TSPO with K_i values of 2.5 and 2.1 nM, respectively. These results surprised us, since replacing the phenyl group with a pyridine in the benzoxazolone derivatives led to at least 10-fold reduction in TSPO affinity.[12] Although we were able to obtain a series of compounds with excellent TSPO affinity and selectivity, these potent compounds suffered from low metabolic stability. In general, compounds with secondary amides have low affinity for TSPO,[16] however compound **37** showed moderate affinity for TSPO (K_i = 33 nM) with remarkable high metabolic stability (62%). This result encouraged us to explore dihydroimidazoquinolinone derivatives having a secondary amide moiety in the side chain, however, our efforts led to no further increase in TSPO affinity (data not shown).

Next, we examined the effect of a substitution at the C-8 position. As shown in Table 3, most compounds showed excellent affinity for TSPO. Compound **40**, an intermediate of the C-8 substituted derivatives, exhibited very potent affinity for TSPO, albeit with poor metabolic stability. Introduction of a methoxy or trifluoromethyl group onto the phenyl ring at the C-8 position of **27** increased TSPO binding affinity. In particular, compounds **41** and **44** were very potent TSPO ligands with K_i values of 0.11 and 0.16 nM, respectively. Based on the results obtained from compounds **41**–**44**, it was predicted that the electron density of the phenyl ring at the C-8 position does not affect the binding affinity for TSPO. Compounds bearing a pyridyl moiety, that is, **45**–**47**, also exhibited high affinity and selectivity for TSPO. Especially, replacement of the phenyl ring at the C-8 position with a 4-pyridyine ring (**47**) increased the affinity for TSPO with a K_i value of 0.55 nM. However, contrary to our expectation, introduction of a *para*-electron withdrawing substituent (**44**) or replacement of the phenyl group with a pyridine (**45**–**47**) did not improve metabolic stability. Finally we prepared compound **48** to examine the effect of inserting a linker between the conjugated two rings. Fortunately, introduction of an aminopyridyl moiety led to a remarkable improvement of metabolic stability (41%). Although not reported here, other compounds with an amino or ether linker between the conjugated two rings exhibited low metabolic stability. As compound **48** showed strong affinity for TSPO and acceptable aqueous solubility (0.004 mg/mL, at pH 7.4 and >1.0 mg/mL, at pH 2.5), further biological evaluation of this compound is in progress.

4. Conclusion

We have synthesized and evaluated three novel tricyclic benzimidazolone derivatives as potent ligands, and selected the dihydroimidazoquinolinone derivative **27** as a lead compound. SAR of dihydroimidazoquinolinone derivatives with various substituents at the amide part and C-8 position revealed very potent TSPO ligands with subnanomolar K_i values, but poor metabolic stability. Optimization of these compounds led to compound **48**, which showed potent affinity for TSPO with good *in vitro* PK profiles. Further biological evaluation of compound **48** is under investigation.

5. Experimental section

5.1. Chemistry

Melting points were determined on Stanford Research Systems OptiMelt MPA100 without correction. NMR spectra were recorded at ambient temperature on a JEOL JNM-AL400 FT NMR spectrometer. Chemical shifts are expressed in δ values (ppm) relative to tetramethylsilane as an internal standard, and signals are expressed as s (singlet), d (doublet), t (triplet), m (multiplet), or br (broad). IR spectra were recorded on a JEOL JIR-SPX60 spectrometer as ATR. High-resolution mass spectra (HRMS) were recorded on a Thermo Fisher Scientific LTQ orbitrap Discovery MS equipment. Elemental analysis was performed on a CE Instrument EA1110 and a Yokokawa analytical system IC7000. In general, reagents and solvents were used as obtained from commercial suppliers without further purification. Reaction progress was determined by thin layer chromatography (TLC) analysis on a Merck silica gel 60 F254 precoated glass plate. Visualization was done with UV light (254 nm) or iodine. Flash column chromatography was conducted using Merck silica gel 60 (70–230 mesh). All reactions were carried out under a nitrogen atmosphere unless otherwise mentioned.

5.1.1. Methyl 3,4-dihydroquinoline-1(2*H*)-carboxylate (13)

To a suspension of tetrahydroquinoline (18.0 mL, 143 mmol) and K_2CO_3 (79.3 g, 574 mmol) in DMF (100 mL) was added methyl-chloroformate (33.2 mL, 430 mmol) with cooling in an ice bath. The reaction mixture was stirred at 50 °C for 6 h and cooled to room temperature. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H_2O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo to give **13** (27.2 g, 99%) as a yellow oil. This product was used in the following reaction without further purification: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.66 (1H, d, J = 7.6 Hz), 7.16 (1H, dd, J = 7.7, 7.7 Hz), 7.09 (1H, d, J = 7.3 Hz), 7.01 (1H, dd, J = 7.3, 7.3 Hz), 3.79 (3H, s), 3.76 (2H, t, J = 6.1 Hz), 2.77 (2H, t, J = 6.6 Hz), 1.99–1.90 (2H, m); IR (ATR) 1701, 1697, 1493, 1439, 1327 cm^{-1}; HRMS (ESI) m/z calcd for $C_{11}H_{14}NO_2$ $[M+H]^+$ 192.1019; found 192.1015.

5.1.2. Methyl 2,3,4,5-tetrahydro-1*H*-1-benzazepine-1-carboxylate (14)

Compound **14** was prepared from 2,3,4,5-tetrahydro-1*H*-benzo[*b*]azepine (800 mg, 5.43 mmol) in a manner similar to that described for compound **14** as a white solid (706 mg, 63%): mp 113–115 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.25–7.10 (4H, m), 4.58–4.32 (1H, m), 3.79 and 3.65 (3H, each s), 2.87–2.58 (3H, m), 2.04–1.75 (3H, m), 1.55–1.27 (1H, m); IR (ATR) 1689, 1495, 1439, 1385, 1307 cm^{-1}; HRMS (ESI) m/z calcd for $C_{12}H_{16}NO_2$ $[M+H]^+$ 206.1176; found 206.1173.

5.1.3. Methyl 6-bromo-3,4-dihydroquinoline-1(2*H*)-carboxylate (15)

To a solution of **13** (26.3 g, 138 mmol) in DMF (140 mL) was added *N*-bromosuccinimide (26.9 g, 151 mmol) with cooling in an ice bath, and the mixture was stirred at room temperature for 2.5 h. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H_2O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo to give **15** (37.1 g, 100%) as a brown oil. This product was used in the following reaction without further purification: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.59 (1H, d, J = 8.5 Hz), 7.28–7.21 (2H, m), 3.79 (3H, s), 3.74 (2H, t, J = 6.1 Hz), 2.74 (2H, t, J = 6.6 Hz), 1.96–1.89 (2H, m); IR (ATR) 1701, 1483, 1441, 1321, 727 cm^{-1}; HRMS (ESI) m/z calcd for $C_{11}H_{13}BrNO_2$ $[M+H]^+$ 270.0124; found 270.0124.

5.1.4. Methyl 7-bromo-2,3,4,5-tetrahydro-1*H*-1-benzazepine-1-carboxylate (16)

Compound **16** was prepared from **14** (521 mg, 2.54 mmol) in a manner similar to that described for compound **15** as a colorless oil (590 mg, 82%): ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.37–7.27 (2H, m), 7.21–6.96 (1H, m), 4.61–4.23 (1H, m), 3.79 and 3.65 (3H, each s), 2.83–2.53 (3H, m), 2.04–1.19 (4H, m); IR (ATR) 1701, 1487, 1441, 1383, 1300 cm^{-1}; HRMS (ESI) m/z calcd for $C_{12}H_{15}BrNO_2$ $[M+H]^+$ 284.0281; found 284.0280.

5.1.5. Methyl 6-bromo-8-nitro-3,4-dihydroquinoline-1(2*H*)-carboxylate (17)

To a solution of nitronium tetrafluoroborate (5.17 g, 38.9 mmol) in CH_3CN (150 mL) was added dropwise a solution of **15** (7.51 g, 27.8 mmol) in CH_3CN (150 mL) with cooling in an ice bath, and the mixture was stirred with cooling in an ice bath for 1 h. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with aqueous saturated $NaHCO_3$ and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (5:1, v/v) as eluent to give **175** (5.77 g, 66%) as a yellow solid: mp 99–101 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.88 (1H, s), 7.49 (1H, s), 3.81 (3H, br s), 3.64–3.62 (2H, br m), 2.85–2.71 (2H, br m), 2.07–1.92 (2H, br m); IR (ATR) 1701, 1483, 1439, 1321, 1190 cm^{-1}; HRMS (ESI) m/z calcd for $C_{11}H_{12}BrN_2O_4$ $[M+H]^+$ 314.9975; found 314.9974.

5.1.6. Methyl 7-bromo-9-nitro-2,3,4,5-tetrahydro-1*H*-1-benzazepine-1-carboxylate (18)

Compound **18** was prepared from **16** (6.09 g, 21.4 mmol) in a manner similar to that described for compound **17** as a yellow solid (4.89 g, 72%): mp 73–75 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.91–7.86 (1H, m), 7.63–7.60 (1H, m), 4.53–4.27 (1H, m), 3.77 and 3.57 (3H, each s), 3.00–2.71 (3H, m), 2.12–1.78 (3H, m), 1.52–1.32 (1H, m); IR (ATR) 1713, 1522, 1303, 1169, 1032 cm^{-1}; HRMS (ESI) m/z calcd for $C_{12}H_{14}BrN_2O_4$ $[M+H]^+$ 329.0131; found 329.0129.

5.1.7. 8-Bromo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-2(1*H*)-one (19)

To a solution of reduced iron (24.1 g, 431 mmol) in AcOH (250 mL) was added dropwise a solution of **17** (19.4 g, 61.6 mmol) in AcOH (200 mL) at 80 °C. The reaction mixture was stirred at 80 °C for 2 h and cooled to room temperature. The reaction mixture was filtered through Celite, and the filtrate was concentrated. The residue was diluted with EtOAc and H_2O. The organic layer was separated, washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo. The resulting solid was triturated with hexane to give **19** (14.6 g, 94%) as a brown solid: mp 235–236 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.81 (1H, s), 6.99 (1H, s), 6.94 (1H, s), 3.68 (2H, t, J = 5.7 Hz), 2.76 (2H, t, J = 6.0 Hz), 2.04–1.94 (2H, m); IR (ATR) 3143, 1707, 1657, 1641, 1491 cm^{-1}; HRMS (ESI) m/z calcd for $C_{10}H_{10}BrN_2O$ $[M+H]^+$ 252.9971; found 252.9967.

5.1.8. 9-Bromo-4,5,6,7-tetrahydroimidazo[4,5,1-*jk*][1]benzazepin-2(1*H*)-one (20)

Compound **20** was prepared from **18** (2.53 g, 7.69 mmol) in a manner similar to that described for compound **19** as a white solid (1.73 g, 84%): mp 147–149 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.01 (1H, br s), 6.99 (1H, d, J = 1.5 Hz), 6.94 (1H, d, J = 2.0 Hz), 3.75 (2H, t, J = 5.2 Hz), 2.93 (2H, t, J = 5.6 Hz), 2.53–2.48 (4H, m); IR (ATR) 2864, 1686, 1610, 1471, 1149 cm^{-1}; HRMS (ESI) m/z calcd for $C_{11}H_{12}BrN_2O$ $[M+H]^+$ 267.0128; found 267.0126.

5.1.9. *tert*-Butyl (8-bromo-2-oxo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetate (21)

To a suspension of **19** (28.9 g, 114 mmol) and K_2CO_3 (23.7 g, 171 mmol) in DMF (400 mL) was added *tert*-butyl bromoacetate (18.5 mL, 126 mmol) with cooling in an ice bath. The reaction mixture was stirred at 50 °C for 3 h and cooled to room temperature. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H_2O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was triturated with hexane to give **21** (41.5 g, 99%) as a beige solid: mp 178–180 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.02 (1H, d, J = 1.7 Hz), 6.87 (1H, d, J = 1.7 Hz), 4.47 (2H, s), 3.85 (2H, t, J = 5.7 Hz), 2.82 (2H, t, J = 6.1 Hz), 2.15–2.07 (2H, m), 1.48 (9H, s); IR (ATR) 1741, 1697, 1498, 1421, 1232 cm^{-1}; HRMS (ESI) m/z calcd for $C_{16}H_{19}BrN_2O_3Na$ $[M+Na]^+$ 389.0471; found 389.0473; Anal. Calcd for $C_{16}H_{19}BrN_2O_3 \cdot 0.10H_2O$: C, 52.07; H, 5.24; N, 7.59; Br, 21.65. Found: C, 52.43; H, 5.29; N, 7.45; Br, 21.25.

5.1.10. *tert*-Butyl (9-bromo-2-oxo-4,5,6,7-tetrahydroimidazo[4,5,1-*jk*][1]benzazepin-1(2*H*)-yl)acetate (22)

Compound **22** was prepared from **20** (1.71 g, 6.40 mmol) in a manner similar to that described for compound **21** as a white solid

(2.42 g, 99%): mp 172–174 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.02 (1H, s), 6.84 (1H, s), 4.48 (2H, s), 3.95 (2H, t, J = 5.4 Hz), 2.98 (2H, t, J = 4.8 Hz), 2.06–1.93 (4H, m), 1.48 (9H, s); IR (ATR) 1741, 1693, 1425, 1230, 1149 cm^{-1}; HRMS (ESI) m/z calcd for $C_{17}H_{22}BrN_2O_3$ $[M+H]^+$ 381.0808; found 381.0804.

5.1.11. *tert*-Butyl (2-oxo-8-phenyl-5,6-dihydro-4H-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetate (23)

To a suspension of **21** (1.50 g, 4.09 mmol) and phenylboronic acid (0.647 g, 5.31 mmol) in 2 M K_2CO_3 solution (6.14 mL, 12.3 mmol) and 1,4-dioxane (30 mL) was added $Pd(PPh_3)_4$ (236 mg, 0.204 mmol) in room temperature. The reaction mixture was stirred at reflux for 2.5 h and cooled to room temperature. Water was then added, and the mixture was extracted with EtOAc. The organic layer was washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using hexane/EtOAc (1:1, v/v) as eluent to give **23** (0.929 g, 62%) as a beige solid: mp 202–203 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.52 (2H, d, J = 8.0 Hz), 7.41 (2H, m), 7.31 (1H, t, J = 7.1 Hz), 7.09 (1H, s), 6.91 (1H, s), 4.54 (2H, s), 3.90 (2H, t, J = 5.7 Hz), 2.90 (2H, t, J = 6.0 Hz), 2.20–2.12 (2H, m), 1.47 (9H, s); IR (ATR) 1741, 1713, 1701, 1228, 1155 cm^{-1}; HRMS (ESI) m/z calcd for $C_{22}H_{25}N_2O_3$ $[M+H]^+$ 365.1860; found 365.1855; Anal. Calcd for $C_{22}H_{24}N_2O_3{\cdot}0.25H_2O$: C, 71.62; H, 6.69; N, 7.59. Found: C, 71.50; H, 6.64; N, 7.71.

5.1.12. *tert*-Butyl (2-oxo-9-phenyl-4,5,6,7-tetrahydroimidazo[4,5,1-*jk*][1]benzazepin-1(2*H*)-yl)acetate (24)

Compound **24** was prepared from **22** (1.84 g, 4.83 mmol) in a manner similar to that described for compound **23** as a white solid (1.74 g, 95%): mp 141–143 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.54 (2H, d, J = 7.3 Hz), 7.45–7.38 (2H, m), 7.32 (1H, t, J = 7.1 Hz), 7.10 (1H, s), 6.89 (1H, s), 4.56 (2H, s), 4.03–3.94 (2H, m), 3.11–3.04 (2H, m), 2.09–1.97 (4H, m), 1.47 (9H, s); IR (ATR) 1741, 1697, 1234, 1153, 754 cm^{-1}; HRMS (ESI) m/z calcd for $C_{23}H_{27}N_2O_3$ $[M+H]^+$ 379.2016; found 379.2009.

5.1.13. (2-Oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetic acid (25)

To a solution of **23** (19.6 g, 53.8 mmol) in AcOH (100 mL) was added 4 N HCl in 1,4-dioxane (80.7 mL). The reaction mixture was stirred at 55 °C for 3 h and cooled to room temperature. The solvent was removed in vacuo, and the resulting solid was triturated with toluene to give **25** (13.6 g, 84%) as a brown solid: mp 216–218 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.03 (1H, br s), 7.64–7.60 (2H, m), 7.45–7.40 (2H, m), 7.34–7.27 (2H, m), 7.17 (1H, s), 4.64 (2H, s), 3.78 (2H, t, J = 5.6 Hz), 2.87 (2H, t, J = 5.9 Hz), 2.11–2.01 (2H, m); IR (ATR) 2364, 1728, 1668, 1659, 1643 cm^{-1}; HRMS (ESI) m/z calcd for $C_{18}H_{16}N_2O_3Na$ $[M+Na]^+$ 331.1053; found 331.1046; Anal. Calcd for $C_{18}H_{16}N_2O_3{\cdot}0.50H_2O$: C, 68.13; H, 5.40; N, 8.83. Found: C, 67.76; H, 5.19; N, 8.78.

5.1.14. (2-Oxo-9-phenyl-4,5,6,7-tetrahydroimidazo[4,5,1-*jk*][1]benzazepin-1(2*H*)-yl)acetic acid (26)

Compound **26** was prepared from **24** (1.57 g, 4.15 mmol) in a manner similar to that described for compound **25** as a yellow solid (1.30 g, 97%): mp 207–209 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.05 (1H, br s), 7.69–7.64 (2H, m), 7.46–7.40 (2H, m), 7.37 (1H, d, J = 1.2 Hz), 7.31 (1H, t, J = 7.3 Hz), 7.20 (1H, s), 4.68 (2H, s), 3.89–3.81 (2H, m), 3.09–3.00 (2H, m), 2.03–1.88 (4H, m); IR (ATR) 2931, 1732, 1662, 1655, 1227 cm^{-1}; HRMS (ESI) m/z calcd for $C_{19}H_{19}N_2O_3$ $[M+H]^+$ 323.1390; found 323.1385.

5.1.15. *N*-Methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-*N*-phenylacetamide (27)

To a solution of **25** (462 mg, 1.50 mmol) in DMF (3.0 mL) were added *N*-methylaniline (244 μL, 2.25 mmol), 1-hydroxybenzotriazole (203 g, 1.50 mmol) and 1-ethyl-3-(3-dimethylaminopropyl)-carbodiimide hydrochloride (431 mg, 2.25 mmol) at room temperature. The reaction mixture was stirred at room temperature for 7 h. Water was then added, and the mixture was extracted with a mixture of toluene and EtOAc (1:1). The organic layer was washed with H_2O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using $CHCl_3$/EtOAc (4:1, v/v) as eluent to give **27** (460 mg, 77%) as a white solid: mp 126–127 °C (MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.56–7.51 (2H, m), 7.51–7.45 (2H, m), 7.45–7.38 (3H, m), 7.37–7.27 (3H, m), 7.06 (1H, s), 6.89 (1H, s), 4.41 (2H, s), 3.84 (2H, t, J = 5.7 Hz), 3.30 (3H, s), 2.88 (2H, t, J = 6.0 Hz), 2.18–2.08 (2H, m); IR (ATR) 1713, 1666, 1493, 1423, 762 cm^{-1}; HRMS (ESI) m/z calcd for $C_{25}H_{24}N_3O_2$ $[M+H]^+$ 398.1863; found 398.1855; Anal. Calcd for $C_{25}H_{23}N_3O_2{\cdot}0.25H_2O$: C, 74.70; H, 5.89; N, 10.45. Found: C, 74.65; H, 5.83; N, 10.51.

5.1.16. 2-(2-Oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-*N*,*N*-dipropylacetamide (28)

Compound **28** was prepared from **25** (308 mg, 1.00 mmol) and dipropylamine (165 μL, 1.20 mmol) in a manner similar to that described for compound **27** as a white solid (244 mg, 62%): mp 129–131 °C (*i*PrOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.53 (2H, dd, J = 8.2, 1.3 Hz), 7.42–7.37 (2H, m), 7.32–7.27 (1H, m), 7.08 (1H, d, J = 1.2 Hz), 7.02 (1H, d, J = 1.2 Hz), 4.69 (2H, s), 3.90 (2H, t, J = 5.9 Hz), 3.39–3.24 (4H, m), 2.90 (2H, t, J = 6.0 Hz), 2.21–2.10 (2H, m), 1.72–1.50 (4H, m), 0.99 (3H, t, J = 7.4 Hz), 0.86 (3H, t, J = 7.4 Hz); IR (ATR) 1705, 1649, 1230, 764, 702 cm^{-1}; HRMS (ESI) m/z calcd for $C_{24}H_{30}N_3O_2$ $[M+H]^+$ 392.2333; found 392.2324; Anal. Calcd for $C_{24}H_{29}N_3O_2$: C, 73.63; H, 7.47; N, 10.73. Found: C, 73.27; H, 7.44; N, 10.71.

5.1.17. *N*-Benzyl-*N*-methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetamide (29)

Compound **29** was prepared from **25** (462 mg, 1.50 mmol) and benzylmethylamine (290 μL, 2.25 mmol) in a manner similar to that described for compound **27** as a white solid (473 mg, 77%): mp 173–175 °C (MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.55 (2H, d, J = 8.0 Hz), 7.46–7.38 (2H, m), 7.34–7.21 (5H, m), 7.13–7.06 (2H, m), 7.03 (1H, s), 4.77–4.58 (4H, m), 3.91 (1H, t, J = 5.7 Hz), 3.73 (1H, t, J = 5.7 Hz), 3.05 and 3.01 (3H, each s), 2.91 (1H, t, J = 6.0 Hz), 2.84 (1H, t, J = 6.0 Hz), 2.21–2.13 (1H, m), 2.11–2.03 (1H, m); IR (ATR) 1693, 1653, 1493, 756, 704 cm^{-1}; HRMS (ESI) m/z calcd for $C_{26}H_{26}N_3O_2$ $[M+H]^+$ 412.2020; found 412.2007; Anal. Calcd for $C_{26}H_{25}N_3O_2$: C, 75.89; H, 6.12; N, 10.21. Found: C, 75.66; H, 6.02; N, 10.26.

5.1.18. *N*-(3-Methoxyphenyl)-*N*-methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetamide (30)

To a suspension of **25** (308 mg, 1.00 mmol) in CH_2Cl_2 (3.0 mL) were added oxalyl choride (96.0 μM, 1.10 mmol) and DMF (5.0 μL) with cooling in an ice bath, and then the mixture was stirred at room temperature for 1 h. The solvent was removed in vacuo, and then the residue was azeotropied with toluene. A solution of acid chloride thus obtained in THF (3.0 mL) was added to a solution of 3-methoxy-*N*-methylaniline (165 mg, 1.20 mmol) and triethylamine (209 μM, 1.50 mmol) in THF (3.0 mL) at room temperature, and then the mixture was stirred at room temperature for 1 h. The reaction was quenched by adding aqueous saturated $NaHCO_3$, and then the mixture was extracted with EtOAc. The organic layer was washed with H_2O and brine and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using EtOAc as eluent to give **30** (220 mg, 52%) as a yellow solid: mp 166–168 °C (MeOH); ^{1}H NMR (400 MHz, $CDCl_3$)

δ 7.56–7.50 (2H, m), 7.46–7.28 (4H, m), 7.06 (1H, s), 6.96–6.88 (3H, m), 6.84–6.80 (1H, m), 4.46 (2H, s), 3.89–3.79 (5H, m), 3.29 (3H, s), 2.88 (2H, t, *J* = 6.0 Hz), 2.18–2.08 (2H, m); IR (ATR) 1705, 1674, 1489, 1421, 754 cm^{-1}; HRMS (ESI) *m*/*z* calcd for $C_{26}H_{26}N_3O_3$ $[M+H]^+$ 428.1969; found 428.1962; Anal. Calcd for $C_{26}H_{25}N_3O_3$: C, 73.05; H, 5.89; N, 9.83. Found: C, 72.72; H, 5.94; N, 9.75.

5.1.19. *N*-(4-Methoxyphenyl)-*N*-methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetamide (31)

Compound **31** was prepared from **25** (308 mg, 1.00 mmol) and *N*-methyl-*p*-anisidine (165 mg, 1.20 mmol) in a manner similar to that described for compound **30** as a brown solid (348 mg, 81%): mp 172–174 °C (MeOH); ^1H NMR (400 MHz, $CDCl_3$) δ 7.53 (2H, d, *J* = 7.3 Hz), 7.44–7.38 (2H, m), 7.31 (1H, t, *J* = 7.4 Hz), 7.28–7.24 (2H, m), 7.05 (1H, s), 6.97 (2H, d, *J* = 8.8 Hz), 6.88 (1H, s), 4.39 (2H, s), 3.89–3.80 (5H, m), 3.27 (3H, s), 2.88 (2H, t, *J* = 5.9 Hz), 2.18–2.09 (2H, m); IR (ATR) 1691, 1662, 1504, 1425, 1238 cm^{-1}; HRMS (ESI) *m*/*z* calcd for $C_{26}H_{26}N_3O_3$ $[M+H]^+$ 428.1969; found 428.1965; Anal. Calcd for $C_{26}H_{25}N_3O_3{\cdot}0.25H_2O$: C, 72.29; H, 5.95; N, 9.73. Found: C, 72.60; H, 5.87; N, 9.79.

5.1.20. *N*-(3-Chlorophenyl)-*N*-methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetamide (32)

Compound **32** was prepared from **25** (308 mg, 1.00 mmol) and 3-chloro-*N*-methylaniline (170 mg, 1.20 mmol) in a manner similar to that described for compound **27** as a white solid (135 mg, 31%): mp 161–162 °C (MeOH); ^1H NMR (400 MHz, $CDCl_3$) δ 7.54 (2H, d, *J* = 7.8 Hz), 7.46–7.23 (7H, m), 7.08 (1H, s), 6.91 (1H, s), 4.45 (2H, s), 3.83 (2H, t, *J* = 5.4 Hz), 3.28 (3H, s), 2.89 (2H, t, *J* = 6.0 Hz), 2.19–2.09 (2H, m); IR (ATR) 1697, 1686, 1674, 760, 694 cm^{-1}; HRMS (ESI) *m*/*z* calcd for $C_{25}H_{23}ClN_3O_2$ $[M+H]^+$ 432.1473; found 432.1467; Anal. Calcd for $C_{25}H_{22}ClN_3O_2$: C, 69.52; H, 5.13; N, 9.73; Cl, 8.21. Found: C, 69.15; H, 5.15; N, 9.80; Cl, 8.12.

5.1.21. *N*-(4-Chlorophenyl)-*N*-methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetamide (33)

Compound **33** was prepared from **25** (308 mg, 1.00 mmol) and 4-chloro-*N*-methylaniline (145 μL, 1.20 mmol) in a manner similar to that described for compound **30** as a pale yellow solid (306 mg, 71%): mp 178–180 °C (MeOH); ^1H NMR (400 MHz, $CDCl_3$) δ 7.53 (2H, d, *J* = 7.6 Hz), 7.46–7.38 (4H, m), 7.31 (1H, t, *J* = 7.1 Hz), 7.28–7.22 (2H, m), 7.07 (1H, s), 6.89 (1H, s), 4.41 (2H, s), 3.83 (2H, t, *J* = 5.6 Hz), 3.27 (3H, s), 2.88 (2H, t, *J* = 6.0 Hz), 2.19–2.07 (2H, m); IR (ATR) 1695, 1670, 1489, 1425, 754 cm^{-1} cm^{-1}; HRMS (ESI) *m*/*z* calcd for $C_{25}H_{23}ClN_3O_2$ $[M+H]^+$ 432.1473; found 432.1466; Anal. Calcd for $C_{25}H_{22}ClN_3O_2$: C, 69.52; H, 5.13; N, 9.73; Cl, 8.21. Found: C, 69.23; H, 5.15; N, 9.75; Cl, 8.22.

5.1.22. *N*-Methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-*N*-(pyridin-2-yl)acetamide (34)

Compound **34** was prepared from **25** (462 mg, 1.50 mmol) and 2-(methylamino)pyridine (185 μL, 1.80 mmol) in a manner similar to that described for compound **27** as a white solid (331 mg, 55%): mp 171–172 °C (MeOH); ^1H NMR (400 MHz, $CDCl_3$) δ 8.50 (1H, d, *J* = 2.9 Hz), 7.79 (1H, t, *J* = 7.0 Hz), 7.54 (2H, d, *J* = 8.0 Hz), 7.44–7.19 (5H, m), 7.08 (1H, s), 7.01 (1H, s), 4.82 (2H, s), 3.87 (2H, t, *J* = 5.7 Hz), 3.44 (3H, s), 2.89 (2H, t, *J* = 6.0 Hz), 2.19–2.10 (2H, m); IR (ATR) 1697, 1660, 1587, 1419, 1313 cm^{-1}; HRMS (ESI) *m*/*z* calcd for $C_{24}H_{23}N_4O_2$ $[M+H]^+$ 399.1816; found 399.1810; Anal. Calcd for $C_{24}H_{22}N_4O_2{\cdot}0.25H_2O$: C, 71.53; H, 5.63; N, 13.90. Found: C, 71.80; H, 5.65; N, 13.94.

5.1.23. *N*-Methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-*N*-(pyridin-3-yl)acetamide (35)

Compound **35** was prepared from **25** (308 mg, 1.00 mmol) and *N*-methy-3-pyridinamine (108 mg, 1.00 mmol) in a manner similar to that described for compound **27** as a white solid (302 mg, 76%): mp 165–167 °C (*i*PrOH); ^1H NMR (400 MHz, $CDCl_3$) δ 8.65 (1H, br s), 8.58 (1H, d, *J* = 2.7 Hz), 7.73 (1H, d, *J* = 8.0 Hz), 7.56–7.50 (2H, m), 7.47–7.37 (3H, m), 7.31 (1H, t, *J* = 7.3 Hz), 7.08 (1H, s), 6.93 (1H, s), 4.41 (2H, s), 3.86–3.78 (2H, m), 3.32 (3H, s), 2.88 (2H, t, *J* = 6.0 Hz), 2.19–2.08 (2H, m); IR (ATR) 1709, 1666, 1493, 1423, 764 cm^{-1}; HRMS (ESI) *m*/*z* calcd for $C_{24}H_{23}N_4O_2$ $[M+H]^+$ 399.1816; found 399.1810; Anal. Calcd for $C_{24}H_{22}N_4O_2{\cdot}0.25H_2O$: C, 71.53; H, 5.63; N, 13.90. Found: C, 71.43; H, 5.64; N, 13.87.

5.1.24. *N*-Methyl-2-(2-oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-*N*-(pyridin-4-yl)acetamide (36)

Compound **36** was prepared from **25** (308 mg, 1.00 mmol) and 4-(methylamino)pyridine (108 mg, 1.00 mmol) in a manner similar to that described for compound **27** as a white solid (225 mg, 56%): mp 141–143 °C (*i*PrOH); ^1H NMR (400 MHz, $CDCl_3$) δ 8.68 (2H, d, *J* = 5.9 Hz), 7.53 (2H, d, *J* = 7.1 Hz), 7.45–7.38 (2H, m), 7.34–7.23 (3H, m), 7.09 (1H, s), 6.92 (1H, s), 4.60 (2H, s), 3.83 (2H, t, *J* = 5.7 Hz), 3.37 (3H, s), 2.89 (2H, t, *J* = 6.0 Hz), 2.19–2.07 (2H, m); IR (ATR) 1693, 1672, 1587, 1491, 1429 cm^{-1}; HRMS (ESI) *m*/*z* calcd for $C_{24}H_{23}N_4O_2$ $[M+H]^+$ 399.1816; found 399.1807; Anal. Calcd for $C_{24}H_{22}N_4O_2{\cdot}0.25H_2O$: C, 71.53; H, 5.63; N, 13.90. Found: C, 71.80; H, 5.56; N, 13.94.

5.1.25. 2-(2-Oxo-8-phenyl-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-*N*-phenylacetamide (37)

Compound **37** was prepared from **25** (462 mg, 1.50 mmol) and aniline (164 μL, 1.80 mmol) in a manner similar to that described for compound **27** as a white solid (479 mg, 83%): mp 223–224 °C (MeOH); ^1H NMR (400 MHz, $CDCl_3$) δ 8.49 (1H, br s), 7.58–7.47 (4H, m), 7.45–7.38 (2H, m), 7.36–7.23 (3H, m), 7.17 (2H, d, *J* = 6.1 Hz), 7.09 (1H, t, *J* = 7.4 Hz), 4.67 (2H, s), 3.95 (2H, t, *J* = 5.7 Hz), 2.94 (2H, t, *J* = 6.0 Hz), 2.24–2.16 (2H, m); IR (ATR) 1687, 1558, 1497, 1238, 700 cm^{-1}; HRMS (ESI) *m*/*z* calcd for $C_{24}H_{22}N_3O_2$ $[M+H]^+$ 384.1707; found 384.1700; Anal. Calcd for $C_{24}H_{21}N_3O_2$: C, 75.18; H, 5.52; N, 10.96. Found: C, 75.07; H, 5.61; N, 11.00.

5.1.26. *N*-Methyl-2-(2-oxo-9-phenyl-4,5,6,7-tetrahydroimidazo[4,5,1-*jk*][1]benzazepin-1(2*H*)-yl)-*N*-phenylacetamide (38)

Compound **38** was prepared from **26** (129 mg, 0.400 mmol) and *N*-methylaniline (43.3 μL, 0.400 mmol) in a manner similar to that described for compound **27** as a beige solid (164 mg, 100%): mp 157–159 °C (MeOH); ^1H NMR (400 MHz, $CDCl_3$) δ 7.54 (2H, d, *J* = 7.1 Hz), 7.51–7.46 (2H, m), 7.46–7.29 (6H, m), 7.07 (1H, s), 6.88 (1H, s), 4.42 (2H, s), 3.96–3.87 (2H, m), 3.30 (3H, s), 3.09–3.01 (2H, m), 2.07–1.95 (4H, m); IR (ATR) 1707, 1662, 1425, 758, 700 cm^{-1}; HRMS (ESI) *m*/*z* calcd for $C_{26}H_{26}N_3O_2$ $[M+H]^+$ 412.2020; found 412.2013; Anal. Calcd for $C_{26}H_{25}N_3O_2$: C, 75.89; H, 6.12; N, 10.21. Found: C, 75.78; H, 6.16; N, 10.23.

5.1.27. (8-Bromo-2-oxo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)acetic acid (39)

Compound **39** was prepared from **21** (16.7 g, 45.5 mmol) in a manner similar to that described for compound **25** as a yellow solid (14.1 g, 100%): mp 213–215 °C; ^1H NMR (400 MHz, DMSO-d_6) δ 13.08 (1H, br s), 7.28 (1H, d, *J* = 1.7 Hz), 7.05 (1H, d, *J* = 1.7 Hz), 4.58 (2H, s), 3.74 (2H, t, *J* = 5.7 Hz), 2.79 (2H, t, *J* = 6.0 Hz), 2.05–1.95 (2H, m); IR (ATR) 1718, 1653, 1635, 1624, 1427 cm^{-1}; HRMS (ESI) *m*/*z* calcd for $C_{12}H_{11}BrN_2O_3Na$ $[M+Na]^+$ 332.9845; found 332.9841.

5.1.28. 2-(8-Bromo-2-oxo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-*N*-methyl-*N*-phenylacetamide (40)

Compound **40** was prepared from **39** (11.2 g, 36.0 mmol) and *N*-methylaniline (4.68 mL, 43.2 mmol) in a manner similar to that de-

scribed for compound **27** as a yellow solid (11.3 g, 78%): mp 195–197 °C (MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.53–7.46 (2H, m), 7.42 (1H, t, J = 7.1 Hz), 7.35 (2H, d, J = 7.8 Hz), 6.98 (1H, s), 6.84 (1H, s), 4.32 (2H, s), 3.80 (2H, t, J = 5.9 Hz), 3.31 (3H, s), 2.79 (2H, t, J = 6.0 Hz), 2.13–2.03 (2H, m); IR (ATR) 1705, 1662, 1497, 1421, 1406 cm^{-1}; HRMS (ESI) m/z calcd for $C_{19}H_{19}BrN_3O_2$ $[M+H]^+$ 400.0655; found 400.0652; Anal. Calcd for $C_{19}H_{18}BrN_3O_2{\cdot}0.25H_2O$: C, 56.38; H, 4.61; N, 10.38; Br, 19.74. Found: C, 56.11; H, 4.51; N, 10.25; Br, 20.03.

5.1.29. 2-[8-(3-Methoxyphenyl)-2-oxo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl]-*N*-methyl-*N*-phenylacetamide (41)

Compound **41** was prepared from **40** (200 mg, 0.500 mmol) and 3-methoxyphenylboronic acid (98.8 mg, 0.650 mmol) in a manner similar to that described for compound **23** as a white solid (105 mg, 49%): mp 221–223 °C (MeOH); ^{1}H NMR ($CDCl_3$) δ 7.52–7.46 (2H, m), 7.41 (1H, t, J = 7.2 Hz), 7.37–7.30 (3H, m), 7.12 (1H, d, J = 7.8 Hz), 7.08–7.04 (2H, m), 6.90–6.84 (2H, m), 4.40 (2H, s), 3.87 (3H, s), 3.84 (2H, t, J = 5.9 Hz), 3.30 (3H, s), 2.88 (2H, t, J = 6.0 Hz), 2.18–2.08 (2H, m); IR (ATR) 1705, 1660, 1491, 1429, 1236 cm^{-1}; HRMS (ESI) m/z calcd for $C_{26}H_{26}N_3O_3$ $[M+H]^+$ 428.1969; found 428.1960; Anal. Calcd for $C_{26}H_{25}N_3O_3{\cdot}0.25H_2O$: C, 72.29; H, 5.95; N, 9.73. Found: C, 72.09; H, 6.02; N, 9.39.

5.1.30. 2-[8-(4-Methoxyphenyl)-2-oxo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl]-*N*-methyl-*N*-phenylacetamide (42)

Compound **42** was prepared from **40** (200 mg, 0.500 mmol) and 4-methoxyphenylboronic acid (98.8 mg, 0.650 mmol) in a manner similar to that described for compound **23** as a white solid (165 mg, 77%): mp 138–140 °C (MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.51–7.38 (5H, m), 7.34 (2H, d, J = 7.3 Hz), 7.01 (1H, s), 6.96 (2H, d, J = 8.8 Hz), 6.84 (1H, s), 4.40 (2H, s), 3.85 (3H, s), 3.83 (2H, t, J = 5.9 Hz), 3.30 (3H, s), 2.87 (2H, t, J = 6.0 Hz), 2.16–2.08 (2H, m); IR (ATR) 1695, 1668, 1497, 1242, 825 cm^{-1}; HRMS (ESI) m/z calcd for $C_{26}H_{26}N_3O_3$ $[M+H]^+$ 428.1969; found 428.1960; Anal. Calcd for $C_{26}H_{25}N_3O_3{\cdot}0.75H_2O$: C, 70.81; H, 6.06; N, 9.53. Found: C, 70.59; H, 5.91; N, 9.55.

5.1.31. *N*-Methyl-2-{2-oxo-8-[3-(trifluoromethyl)phenyl]-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl}-*N*-phenylacetamide (43)

Compound **43** was prepared from **40** (200 mg, 0.500 mmol) and 3-(trifluoromethyl)phenylboronic acid (123 mg, 0.650 mmol) in a manner similar to that described for compound **23** as a white solid (207 mg, 89%): mp 259–260 °C (MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.77 (1H, s), 7.71 (1H, d, J = 7.3 Hz), 7.58–7.47 (4H, m), 7.42 (1H, t, J = 7.1 Hz), 7.36 (2H, d, J = 7.3 Hz), 7.06 (1H, s), 6.89 (1H, s), 4.42 (2H, s), 3.85 (2H, t, J = 5.6 Hz), 3.31 (3H, s), 2.89 (2H, t, J = 6.0 Hz), 2.20–2.09 (2H, m); IR (ATR) 1716, 1660, 1421, 1329, 1117 cm^{-1}; HRMS (ESI) m/z calcd for $C_{26}H_{23}F_3N_3O_2$ $[M+H]^+$ 466.1737; found 466.1726; Anal. Calcd for $C_{26}H_{22}F_3N_3O_2$: C, 67.09; H, 4.76; N, 9.03; F, 12.24. Found: C, 66.97; H, 4.78; N, 9.03; F, 12.26.

5.1.32. *N*-Methyl-2-{2-oxo-8-[4-(trifluoromethyl)phenyl]-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl}-*N*-phenylacetamide (44)

Compound **44** was prepared from **40** (200 mg, 0.500 mmol) and 4-(trifluoromethyl)phenylboronic acid (123 mg, 0.650 mmol) in a manner similar to that described for compound **23** as a white solid (196 mg, 84%): mp 207–209 °C (MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.68–7.61 (4H, m), 7.53–7.46 (2H, m), 7.42 (1H, t, J = 7.1 Hz), 7.36 (2H, d, J = 7.3 Hz), 7.07 (1H, s), 6.90 (1H, s), 4.41 (2H, s), 3.85 (2H, t, J = 5.7 Hz), 3.31 (3H, s), 2.89 (2H, t, J = 6.0 Hz), 2.20–2.10 (2H, m); IR (ATR) 1708, 1660, 1323, 1111, 1065 cm^{-1}; HRMS (ESI) m/z calcd for $C_{26}H_{23}F_3N_3O_2$ $[M+H]^+$ 466.1737; found 466.1724; Anal. Calcd for $C_{26}H_{22}F_3N_3O_2{\cdot}0.50H_2O$: C, 65.82; H, 4.89; N, 8.86; F, 12.01. Found: C, 65.69; H, 4.81; N, 8.87; F, 11.95.

5.1.33. *N*-Methyl-2-[2-oxo-8-(pyridin-2-yl)-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl]-*N*-phenylacetamide (45)

To a solution of **40** (200 mg, 0.500 mmol) and 2-(tributylstannyl)pyridine (0.192 mL, 0.600 mmol) in toluene (3.0 mL) was added $Pd(PPh_3)_4$ (28.9 mg, 0.025 mmol) at room temperature. The reaction mixture was stirred at reflux for 14.5 h and cooled to room temperature. Water was then added, and the mixture was extracted with EtOAc. The organic layer was washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using EtOAc/MeOH (10:1, v/v) as eluent to give **45** (42.9 mg, 22%) as a white solid: mp 200–202 °C (MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.65 (1H, d, J = 4.9 Hz), 7.74–7.65 (2H, m), 7.53–7.45 (3H, m), 7.44–7.33 (4H, m), 7.20–7.15 (1H, m), 4.44 (2H, s), 3.85 (2H, t, J = 5.7 Hz), 3.30 (3H, s), 2.90 (2H, t, J = 6.0 Hz), 2.18–2.09 (2H, m); IR (ATR) 1704, 1666, 1425, 781, 706 cm^{-1}; HRMS (ESI) m/z calcd for $C_{24}H_{23}N_4O_2$ $[M+H]^+$ 399.1816; found 399.1805; Anal. Calcd for $C_{24}H_{22}N_4O_2{\cdot}0.50H_2O$: C, 70.74; H, 5.69; N, 13.75. Found: C, 70.55; H, 5.49; N, 13.72.

5.1.34. *N*-Methyl-2-[2-oxo-8-(pyridin-3-yl)-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl]-*N*-phenylacetamide (46)

Compound **46** was prepared from **40** (200 mg, 0.500 mmol) and 3-pyridineboronic acid (79.9 mg, 0.650 mmol) in a manner similar to that described for compound **23** as a white solid (199 mg, 100%): mp 130–132 °C (MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.79 (1H, s), 8.55 (1H, d, J = 4.9 Hz), 7.87–7.79 (1H, m), 7.54–7.31 (6H, m), 7.05 (1H, s), 6.88 (1H, s), 4.42 (2H, s), 3.86 (2H, t, J = 5.7 Hz), 3.31 (3H, s), 2.90 (2H, t, J = 6.0 Hz), 2.20–2.10 (2H, m); IR (ATR) 1691, 1659, 1643, 1491, 1431 cm^{-1}; HRMS (ESI) m/z calcd for $C_{24}H_{23}N_4O_2$ $[M+H]^+$ 399.1816; found 399.1810; Anal. Calcd for $C_{24}H_{22}N_4O_2{\cdot}1.50H_2O$: C, 67.75; H, 5.92; N, 13.17. Found: C, 67.41; H, 5.84; N, 13.02.

5.1.35. *N*-Methyl-2-[2-oxo-8-(pyridin-4-yl)-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl]-*N*-phenylacetamide (47)

Compound **47** was prepared from **40** (100 mg, 0.250 mmol) and 4-pyridineboronic acid (36.9 mg, 0.300 mmol) in a manner similar to that described for compound **23** as a white solid (39.7 mg, 40%): mp 238–239 °C (MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.62 (2H, d, J = 6.3 Hz), 7.57 (2H, d, J = 6.3 Hz), 7.54–7.48 (2H, m), 7.43 (1H, t, J = 7.1 Hz), 7.37 (2H, d, J = 7.6 Hz), 7.16 (1H, s), 6.99 (1H, s), 4.42 (2H, s), 3.86 (2H, t, J = 5.7 Hz), 3.32 (3H, s), 2.91 (2H, t, J = 6.0 Hz), 2.21–2.08 (2H, m); IR (ATR) 1709, 1695, 1662, 1497, 1429 cm^{-1}; HRMS (ESI) m/z calcd for $C_{24}H_{23}N_4O_2$ $[M+H]^+$ 399.1816; found 399.1803; Anal. Calcd for $C_{24}H_{22}N_4O_2{\cdot}1.75H_2O$: C, 67.04; H, 5.98; N, 13.03. Found: C, 66.77; H, 5.75; N, 12.83.

5.1.36. *N*-Methyl-2-[2-oxo-8-(pyridin-3-ylamino)-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl]-*N*-phenylacetamide (48)

A mixture of **40** (200 mg, 0.500 mmol), 3-aminopyridine (70.6 mg, 0.750 mmol), $Pd_2(dba)_3$ (22.9 mg, 0.0250 mmol), Xantphos (43.4 mg, 0.0750 mmol) and Cs_2CO_3 (228 mg, 0.700 mmol) in toluene (4.0 mL) was heated at reflux for 11 h and cooled to room temperature. The reaction was quenched by adding aqueous saturated $NaHCO_3$, and then the mixture was extracted with CH_2Cl_2. The organic layer was washed with brine and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using EtOAc/MeOH (20:1, v/v) as eluent to give **48** (110 mg, 53%) as a yellow solid: mp 194–196 °C (MeOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.25 (1H, d, J = 2.7 Hz), 8.06 (1H, dd, J = 4.4,

1.2 Hz), 7.50–7.44 (2H, m), 7.40 (1H, t, J = 6.7 Hz), 7.32 (2H, d, J = 7.3 Hz), 7.20–7.15 (1H, m), 7.12–7.06 (1H, m), 6.65 (1H, s), 6.54 (1H, s), 4.32 (2H, s), 3.80 (2H, t, J = 5.7 Hz), 3.29 (3H, s), 2.77 (2H, t, J = 6.0 Hz), 2.14–2.04 (2H, m); IR (ATR) 1695, 1662, 1506, 1429, 694 cm^{-1}; HRMS (ESI) m/z calcd for $C_{24}H_{24}N_5O_2$ $[M+H]^+$ 414.1925; found 414.1911; Anal. Calcd for $C_{24}H_{23}N_5O_2{\cdot}0.25H_2O$: C, 68.96; H, 5.67; N, 16.76. Found: C, 69.21; H, 5.63; N, 16.47.

5.1.37. *N*-Methyl-2-(2-oxo-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-1(2*H*)-yl)-*N*-phenylacetamide (49)

To a solution of **40** (100 mg, 0.250 mmol) in MeOH (5.0 mL) and THF (5.0 mL) was added 10% Pd/C (50% wet, 10.0 mg), and stirred at room temperature for 1 h under hydrogen atmosphere. The reaction mixture was filtered through Celite, and the filtrate was concentrated. The residue was recrystallized from *i*PrOH to give **49** (27.6 mg, 34%) as a white solid: mp 173–174 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.51–7.44 (2H, m), 7.40 (1H, t, J = 7.7 Hz), 7.32 (2H, d, J = 7.1 Hz), 6.95 (1H, dd, J = 7.7, 7.7 Hz), 6.83 (1H, d, J = 7.7 Hz), 6.71 (1H, d, J = 7.7 Hz), 4.37 (2H, s), 3.81 (2H, t, J = 5.9 Hz), 3.30 (3H, s), 2.83 (2H, t, J = 6.1 Hz), 2.14–2.06 (2H, m); IR (ATR) 1699, 1660, 1497, 1421, 739 cm^{-1}; HRMS (ESI) m/z calcd for $C_{19}H_{20}N_3O_2$ $[M+H]^+$ 322.1550; found 322.1548; Anal. Calcd for $C_{19}H_{19}N_3O_2{\cdot}0.25H_2O$: C, 70.03; H, 6.03; N, 12.89. Found: C, 69.90; H, 5.94; N, 12.82.

5.1.38. *tert*-Butyl (2-amino-3-nitrophenoxy)acetate (51)

To a suspension of 2-amino-3-nitrophenol (2.51 g, 16.3 mmol) and K_2CO_3 (3.15 g, 22.8 mmol) in DMF (15 mL) was added *tert*-butyl bromoacetate (2.55 mL, 17.3 mmol) with cooling in an ice bath. The reaction mixture was stirred at room temperature for 2.5 h. Water was then added, and the mixture was extracted with toluene. The organic layer was washed with H_2O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo, and the residue was purified by silica gel column chromatography using Hexane/EtOAc (10:1, v/v) as eluent to give **51** (3.59 g, 82%) as an orange solid: mp 103–105 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.79 (1H, dd, J = 8.8, 1.2 Hz), 6.82 (1H, d, J = 7.8 Hz), 6.62–6.52 (3H, m), 4.58 (2H, s), 1.50 (9H, s); IR (ATR) 3479, 1736, 1628, 1522, 1151 cm^{-1}; HRMS (ESI) m/z calcd for $C_{12}H_{17}N_2O_5$ $[M+H]^+$ 269.1132; found 269.1127.

5.1.39. *tert*-Butyl (2-amino-5-bromo-3-nitrophenoxy)acetate (52)

To a solution of **51** (2.54 g, 9.47 mmol) in DMF (15 mL) was added *N*-bromosuccinimide (1.77 g, 9.94 mmol) with cooling in an ice bath, and the mixture was stirred at room temperature for 3 h. Water was then added, and the mixture was extracted with Et_2O. The organic layer was washed with H_2O and brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo. The resulting solid was triturated with hexane to give **52** (3.26 g, 99%) as a brown solid: mp 73–74 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.95 (1H, d, J = 2.2 Hz), 6.89 (1H, s), 6.61 (2H, br s), 4.58 (2H, s), 1.51 (9H, s); IR (ATR) 3369, 1743, 1516, 1209, 1151 cm^{-1}; HRMS (ESI) m/z calcd for $C_{12}H_{16}BrN_2O_5$ $[M+H]^+$ 347.0237; found 347.0236.

5.1.40. 7-Bromo-5-nitro-2*H*-1,4-benzoxazin-3(4*H*)-one (53)

To a solution of **52** (2.33 g, 6.71 mmol) in toluene (10 mL) was added *p*-toluenesulfonic acid monohydrate (0.100 g, 0.526 mmol) at room temperature The reaction mixture was stirred at 80 °C for 2 h, and cooled to room temperature. The reaction mixture was concentrated, and the residue was diluted with CH_2Cl_2 and aqueous saturated $NaHCO_3$. The organic layer was separated, washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo. The resulting solid was triturated with hexane to give **53** (1.82 g, 99%) as a yellow solid: mp 179–180 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.50 (1H, s), 7.91 (1H, d, J = 2.2 Hz), 7.68 (1H, d, J = 2.2 Hz), 4.78 (2H, s); IR (ATR) 2343, 1699, 1684, 1338, 1286 cm^{-1}; HRMS (ESI) m/z calcd for $C_8H_4BrN_2O_4$ $[M-H]^-$ 270.9360; found 270.9352.

5.1.41. 5-Nitro-7-phenyl-2*H*-1,4-benzoxazin-3(4*H*)-one (54)

To a suspension of **53** (1.30 g, 4.76 mmol) and phenylboronic acid (0.697 g, 5.71 mmol) in 2 M K_2CO_3 solution (7.14 mL, 14.3 mmol) and 1,4-dioxane (15 mL) was added $Pd(PPh_3)_4$ (275 mg, 0.238 mmol) at room temperature. The reaction mixture was stirred at reflux for 3 h and cooled to room temperature. 1 M HCl solution was then added, and the separated solid was collected by filtration to give [(4-amino-5-nitrobiphenyl-3-yl)oxy]acetic acid (921 mg, 67%) as a brown solid. To a suspension of [(4-amino-5-nitrobiphenyl-3-yl)oxy]acetic acid (900 mg, 3.12 mmol) in toluene (15 mL) was added *p*-toluenesulfonic acid monohydrate (0.119 g, 0.624 mmol) at room temperature The reaction mixture was stirred at reflux for 1.5 h, and cooled to room temperature. The reaction mixture was concentrated, and the residue was diluted with CH_2Cl_2 and aqueous saturated $NaHCO_3$. The organic layer was separated, washed with brine, and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo. The resulting solid was triturated with hexane to give **54** (718 mg, 85%) as a yellow solid: mp 163–165 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.47 (1H, s), 8.01 (1H, d, J = 2.2 Hz), 7.78–7.71 (3H, m), 7.52–7.45 (2H, m), 7.42 (1H, t, J = 7.1 Hz), 4.80 (2H, s); IR (ATR) 3261, 1716, 1699, 1338, 1176 cm^{-1}; HRMS (ESI) m/z calcd for $C_{14}H_9N_2O_4$ $[M-H]^-$ 269.0568; found 269.0561.

5.1.42. 7-Phenyl-3,4-dihydro-2*H*-1,4-benzoxazin-5-amine (55)

To a solution of $LiAlH_4$ (387 mg, 10.2 mmol) in THF (15 mL) was added dropwise a solution of **54** (689 mg, 2.55 mmol) in THF (25 mL) at 50 °C. The reaction mixture was stirred at 50 °C for 3 h and cooled to room temperature. The reaction was then quenched by dropwise addition of H_2O (0.400 mL), 15% NaOH solution (0.400 mL) and H_2O (1.20 mL) with cooling in an ice bath. The reaction mixture was filtered through Celite, and the filtrate was concentrated. The residue was purified by silica gel column chromatography using hexane/EtOAc (1:4, v/v) as eluent to give **55** (279 mg, 48%) as a brown oil: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.55–7.47 (2H, m), 7.41–7.34 (2H, m), 7.30–7.23 (1H, m), 6.65 (1H, d, J = 2.0 Hz), 6.60 (1H, d, J = 2.0 Hz), 4.23 (2H, t, J = 4.4 Hz), 3.46 (2H, t, J = 4.4 Hz); IR (ATR) 3334, 2870, 1487, 1340, 754 cm^{-1}; HRMS (ESI) m/z calcd for $C_{14}H_{15}N_2O$ $[M+H]^+$ 227.1179; found 227.1176.

5.1.43. 8-Phenyl-4,5-dihydroimidazo[1,5,4-*de*][1,4]benzoxazin-2(1*H*)-one (56)

To a solution of **55** (263 mg, 1.16 mmol) in THF (5.0 mL) was added 1,1′-carbonyldiimidazole (226 mg, 1.39 mmol) at room temperature. The mixture was stirred at reflux for 1 h and cooled to room temperature. The reaction was then quenched by adding 2 M HCl solution, and the mixture was extracted with EtOAc. The organic layer was washed with brine and dried over anhydrous sodium sulfate. After filtration, the solvent was removed in vacuo. The residue was purified by silica gel column chromatography using EtOAc as eluent to give **56** (234 mg, 80%) as a brown solid: mp 195–197 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.84 (1H, s), 7.60–7.53 (2H, m), 7.45–7.37 (2H, m), 7.30 (1H, t, J = 7.3 Hz), 6.83 (2H, s), 4.41 (2H, t, J = 4.5 Hz), 3.92 (2H, t, J = 4.5 Hz); IR (ATR) 3000, 1686, 1660, 1490, 1410 cm^{-1}; HRMS (ESI) m/z calcd for $C_{15}H_{13}N_2O_2$ $[M+H]^+$ 253.0972; found 253.0965.

5.1.44. *tert*-Butyl (2-oxo-8-phenyl-4,5-dihydroimidazo[1,5,4-*de*][1,4]benzoxazin-1(2*H*)-yl)acetate (57)

Compound **57** was prepared from **56** (228 mg, 0.904 mmol) in a manner similar to that described for compound **21** as a beige solid

(259 mg, 78%): mp 160–162 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.51 (2H, d, J = 7.6 Hz), 7.44–7.38 (2H, m), 7.32 (1H, t, J = 7.6 Hz), 6.88 (1H, s), 6.73 (1H, s), 4.54 (2H, s), 4.44 (2H, t, J = 4.6 Hz), 4.05 (2H, t, J = 4.6 Hz), 1.48 (9H, s); IR (ATR) 1741, 1707, 1653, 1234, 1157 cm^{-1}; HRMS (ESI) m/z calcd for $C_{21}H_{23}N_2O_4$ $[M+H]^+$ 367.1652; found 367.1648.

5.1.45. (2-Oxo-8-phenyl-4,5-dihydroimidazo[1,5,4-*de*][1,4]benzoxazin-1(2*H*)-yl)acetic acid (58)

Compound **58** was prepared from **57** (239 mg, 0.652 mmol) in a manner similar to that described for compound **25** as a beige solid (202 mg, 100%): mp 226–228 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.08 (1H, br s), 7.61 (2H, t, J = 4.3 Hz), 7.46–7.39 (2H, m), 7.31 (1H, t, J = 7.3 Hz), 7.16 (1H, d, J = 1.0 Hz), 6.91 (1H, d, J = 1.0 Hz), 4.64 (2H, s), 4.44 (2H, t, J = 4.6 Hz), 3.98 (2H, t, J = 4.6 Hz); IR (ATR) 2875, 1718, 1647, 1201, 1034 cm^{-1}; HRMS (ESI) m/z calcd for $C_{17}H_{15}N_2O_4$ $[M+H]^+$ 311.1026; found 311.1022.

5.1.46. *N*-Methyl-2-(2-oxo-8-phenyl-4,5-dihydroimidazo[1,5,4-*de*][1,4]benzoxazin-1(2*H*)-yl)-*N*-phenylacetamide (59)

Compound **59** was prepared from **58** (55.0 mg, 0.177 mmol) and *N*-methylaniline (19.2 μL, 0.177 mmol) in a manner similar to that described for compound **27** as a white solid (50.1 mg, 71%): mp 166–167 °C (*i*PrOH); ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.55–7.46 (4H, m), 7.45–7.38 (3H, m), 7.37–7.29 (3H, m), 6.85 (1H, s), 6.71 (1H, s), 4.46–4.35 (4H, m), 3.99 (2H, t, J = 4.6 Hz), 3.31 (3H, s); IR (ATR) 1718, 1662, 1497, 1284, 1192 cm^{-1}; HRMS (ESI) m/z calcd for $C_{24}H_{22}N_3O_3$ $[M+H]^+$ 400.1656; found 400.1648; Anal. Calcd for $C_{24}H_{21}N_3O_3$: C, 72.16; H, 5.30; N, 10.52. Found: C, 71.80; H, 5.43; N, 10.37.

5.2. Biology

5.2.1. TSPO-binding assay

Male SD rats (Japan Charles River) were decapitated, and the kidney was dissected. The kidney was homogenized in five volumes of ice-cold 50 mM Tris–HCl buffer (pH 7.6) with a microhomogenizer (Physcotron, Niti-on Inc.). The homogenate was centrifuged at 20,000×g and 4 °C for 10 min, and the resulting pellet was resuspended in the same volume of fresh buffer and recentrifuged. These resuspension and centrifugation procedures were repeated once more, and the obtained pellet was resuspended in 50 mM Tris–HCl buffer (pH 7.6) at ae protein concentration of 2.63 mg/mL and stored frozen at −80 °C until use. The crude mitochondrial preparation (0.895 mL) was incubated with [^{3}H]-PK11195 (final concentration 1.0 nM) and various concentrations of test compounds in a total volume of 1.0 mL for 1 h at 4 °C. The reaction was terminated by rapid fitration through a GF/B glass filter presoaked with 0.3% polyethyleneimine. The filters were immediately washed with ice-cold 50 mM Tris–HCl buffer (pH 7.6), and the filter-bound radioactivity was quantified using a liquid scintillation analyzer (Tri Carb 2700TR, Packard). Nonspecific binding was determined in the presence of 10 μM PK11195. All assays were carried out in duplicate except for the total binding and nonspecific binding, which were in quadruplicate. Specific binding was determined by subtracting nonspecific binding from total binding. The IC_{50} values for each test compound were determined according to a nonlinear least-square curve-fitting method using the SAS® system (SAS Institute Inc.). In the assay with rat kidney TSPOs, K_i values were calculated according to the following formula: $K_i = IC_{50}/(1 + [L]/K_D)$, where $[L]$ and K_D are the concentration of [^{3}H]-PK11195 and the dissociation constant of PK11195 calculated by Scatchard analysis, respectively.

5.2.2. CBR-binding assay

Male SD rats (Japan Charles River) were decapitated, and the cerebral cortex was dissected. The cerebral cortex was homogenized in 10 volumes of ice-cold potassium phosphate buffer (200 mM KCl, 20 mM KOH, 20 mM KH_2PO_4, pH 7.4) with a microhomogenizer (Physcotron, Niti-on Inc.). The homogenate was centrifuged at 32,500×g and 4 °C for 15 min, and the resulting pellet was resuspended in the same volume of fresh buffer and recentrifuged. These resuspension and centrifugation procedures were repeated once more, and the obtained pellet was resuspended in potassium phosphate buffer (200 mM KCl, 20 mM KOH, 20 mM KH_2PO_4, pH 7.4) at a protein concentration of 2.63 mg/mL and stored frozen at −80 °C until use. The crude cerebral cortex preparation (0.895 mL) was incubated with [^{3}H]-flumazenil (final concentration 1.0 nM) and 10 μM of each test compound in a total volume of 1.0 mL for 1 h at 25 °C. The reaction was terminated by rapid fitration through a GF/B glass filter presoaked with 0.3% polyethyleneimine. The filters were immediately washed with ice-cold potassium phosphate buffer (200 mM KCl, 20 mM KOH, 20 mM KH_2PO_4, pH 7.4), and the filter-bound radioactivity was quantified using a liquid scintillation analyzer (Tri Carb 2700TR, Packard). Nonspecific binding was determined in the presence of 10 μM [^{3}H]-flumazenil. All assays were done in duplicate. Specific binding was determined by subtracting nonspecific binding from total binding.

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Supplementary data

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