3142 J. Med. Chem. 2010, 53, 3142–3153
DOI: 10.1021/jm901775y

Journal of Medicinal Chemistry Article

Optimization of Phenyl-Substituted Benzimidazole Carboxamide Poly(ADP-Ribose) Polymerase Inhibitors: Identification of (*S*)-2-(2-Fluoro-4-(pyrrolidin-2-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (A-966492), a Highly Potent and Efficacious Inhibitor

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Received November 30, 2009

We have developed a series of phenylpyrrolidine- and phenylpiperidine-substituted benzimidazole carboxamide poly(ADP-ribose) polymerase (PARP) inhibitors with excellent PARP enzyme potency as well as single-digit nanomolar cellular potency. These efforts led to the identification of (*S*)-2-(2-fluoro-4-(pyrrolidin-2-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (**22b**, A-966492). Compound **22b** displayed excellent potency against the PARP-1 enzyme with a K_i of 1 nM and an EC_{50} of 1 nM in a whole cell assay. In addition, **22b** is orally bioavailable across multiple species, crosses the blood−brain barrier, and appears to distribute into tumor tissue. It also demonstrated good in vivo efficacy in a B16F10 subcutaneous murine melanoma model in combination with temozolomide and in an MX-1 breast cancer xenograft model both as a single agent and in combination with carboplatin.

Introduction

The poly(ADP-ribose) polymerases (PARPs)[a] are an 18-member family of nuclear enzymes that share a common catalytic PARP homology domain and are involved in the detection and repair of DNA damage. Only PARP-1 and PARP-2 contain a DNA-binding domain, which facilitates localization to the site of DNA damage.[1a] PARP-1 and PARP-2 catalyze the transfer of ADP-ribose units from intracellular nicotinamide adenine dinucleotide (NAD^+) to nuclear acceptor proteins, leading to the formation of ADP-ribose polymers. This is a key process for the repair of DNA damage caused by DNA-damaging chemotherapeutic agents and radiation via base-excision repair (BER)-mediated single strand break repair.[1] Thus, PARP-1 (and to a lesser extent, PARP-2) contributes to the resistance that often develops after cancer therapy.[2] Recent preclinical as well as clinical data have now been reported for several inhibitors of PARP-1,[3−12] including clinical compounds 2-[(*R*)-2-methylpyrrolidin-2-yl]-1*H*-benzimidazole-4-carboxamide (ABT-888, veliparib, **1b**),[4a,12b] 4-(4-(4-(cyclopropanecarbonyl)piperazine-1-carbonyl)-3-fluorobenzyl)phthalazin-1(2*H*)-one (AZD2281, olaparib, **2a**),[9] 8-fluoro-5-(4-((methylamino)methyl)phenyl)-2,3,4,6-tetrahydro-1*H*-azepino[5,4,3-cd]indol-1-one (AG014699, **2b**),[10] and 2-{4-[(3S)-piperidin-3-yl]phenyl}-2*H*-indazole-7-carboxamide (MK-4827, **2c**),[8] demonstrating the ability of these inhibitors to not only enhance the efficacy of multiple chemotherapeutics but also demonstrate single agent efficacy in cancers with deficiencies in DNA-repair genes such as BRCA1 and BRCA2. BRCA1 and BRCA2 mutations are associated with homologous recombination (HR)-mediated double strand break repair defects, and inhibition of single strand break repair via PARP inhibition results in a synthetic lethality.[13] In addition to utility in oncology indications, hyperactivation of PARP-1 as a response to more extensive DNA damage has been associated with several other diseases, including stroke, myocardial ischemia, arthritis, colitis, and allergic encephalomyelitis.[1a] We have previously described optimization efforts on a series of potent benzimidazole-containing PARP inhibitors, including **1a**,[12a] culminating in the identification of a clinical candidate **1b**.[12b] This compound demonstrated significant oral efficacy in a number of preclinical rodent tumor models, potentiating the efficacy of cytotoxic agents such as temozolomide (TMZ), cisplatin, carboplatin, and cyclophosphamide, and has currently progressed into human phase II clinical trials. In this report, we describe a series of phenylpyrrolidine- and phenylpiperidine-substituted benzimidazole carboxamide PARP-1/2 inhibitors. These efforts resulted in the identification of benzimidazole analogue **22b**, a potent inhibitor of both PARP-1 and PARP-2 enzymes (K_i = 1 and 1.5 nM) with excellent potency in C41 whole cells (EC_{50} = 1 nM). In addition, **22b** has excellent pharmaceutical properties and has demonstrated in vivo efficacy in preclinical mouse tumor models in combination with TMZ and carboplatin,

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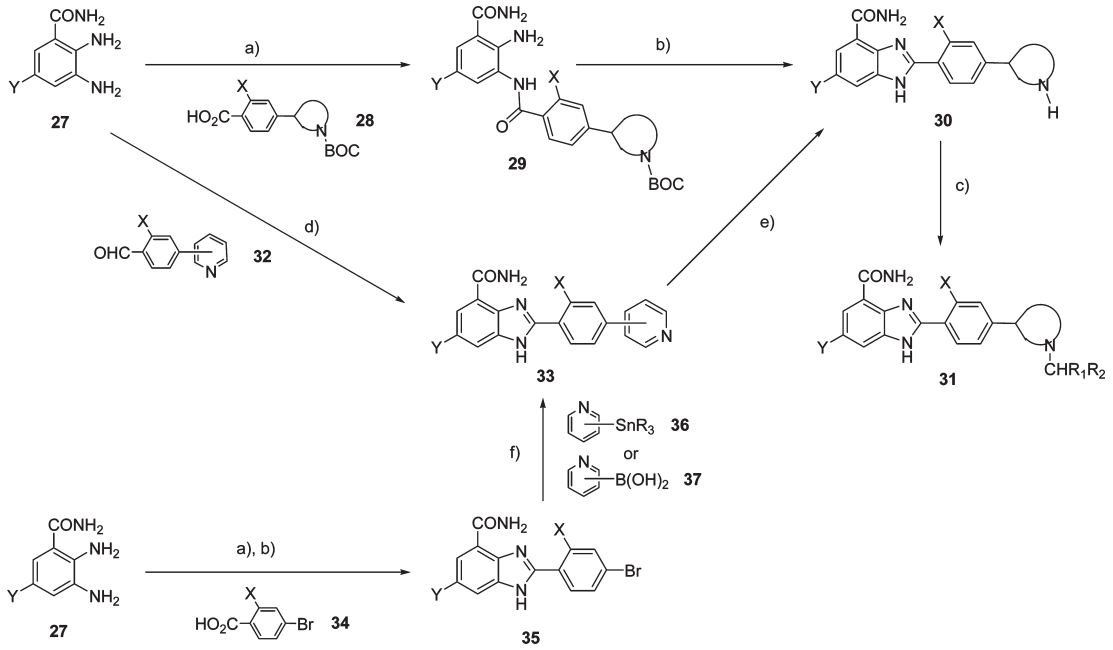
[a] Abbeviations: PARP, poly(ADP-ribose) polymerase; TMZ, temozolomide; HR, homologous recombination; BER, base-excision repair; Boc, *tert*-butoxycarbonyl, CBZ, carbobenzyloxy; TGI, tumor growth inhibition.

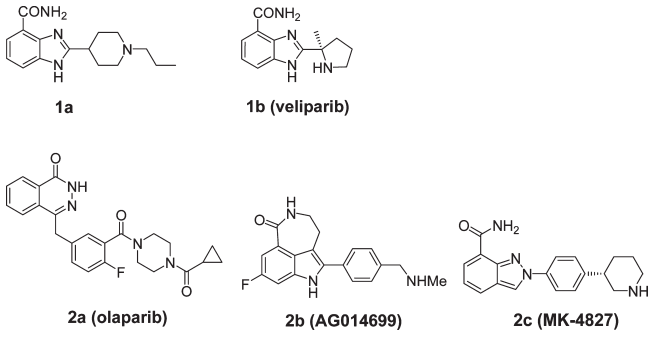
 Published on Web 03/25/2010

Scheme 1[a]



[a] Reagents and conditions: (a) CDI, pyridine, DMF or $(CO)_2Cl_2$, DMF, CH_2Cl_2, TEA. (b) (1) HOAc, heat; (2) HCl, MeOH or TFA, CH_2Cl_2. (c) $NaBH_3CN$ or $NaBH(OAc)_3$, $R_1R_2C(O)$, MeOH, AcOH. (d) 10% Pd/C, MeOH. (e) H_2, PtO_2 or Pt/C, AcOH. (f) Compound **36**, $Pd_2(dba)_3$, Pd(o-tol)$_3$, DMF, Et_3N, heat or **37**, $PdCl_2(dppf)_2Cl_2$, Na_2CO_3, dioxane, heat.

as well as single agent activity in a BRCA1-deficient MX-1 tumor model.



Chemistry

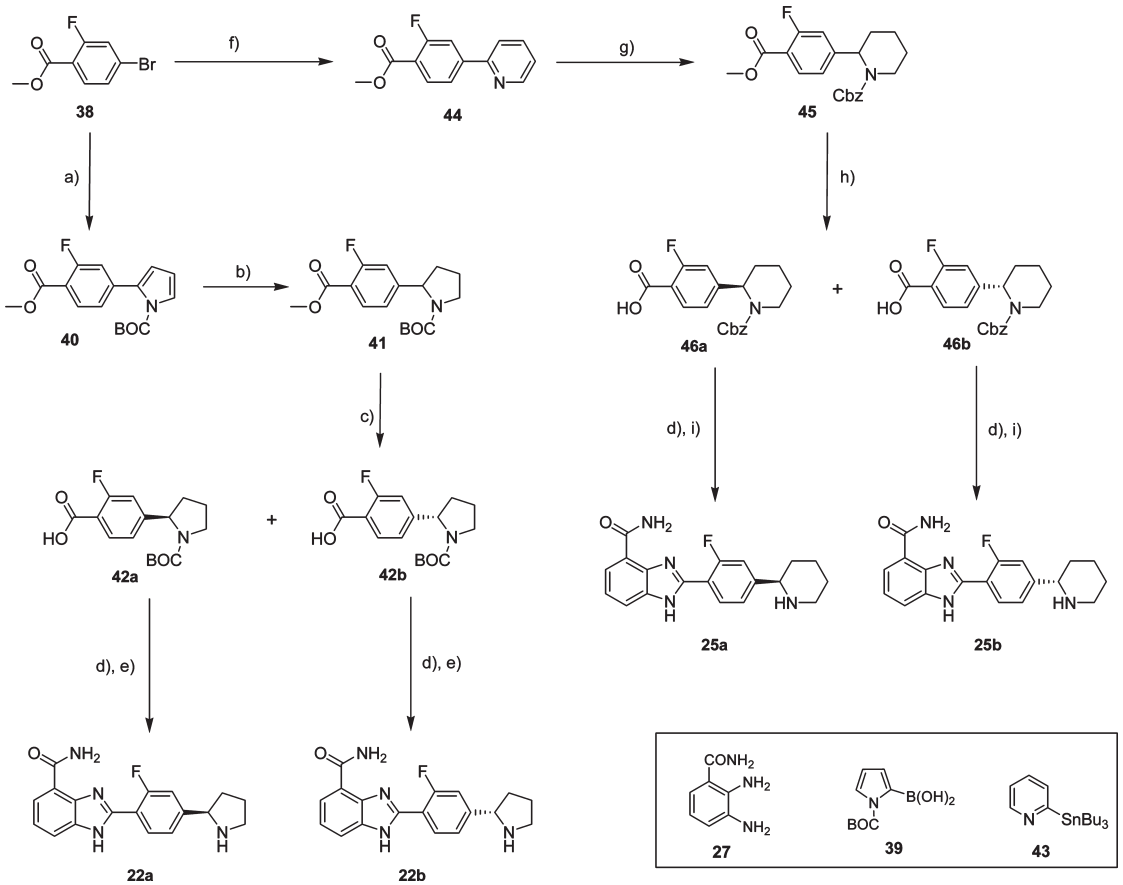
The benzimidazole ring system was constructed as described previously.[12,14] As shown in Scheme 1, a diaminobenzamide **27** was coupled with a pyrrolidine- or piperidine-containing benzoic acid **28** using either carbonyldiimidazole (CDI) or by coupling of the respective acid chloride to give amide **29**. Refluxing in AcOH provided the benzimidazole, which was deprotected under acidic conditions to give **30**. Reductive amination with an appropriate aldehyde or ketone provided the pyrrolidine or piperidine tertiary amines **31**. Alternately, **27** was reacted with pyridine-containing benzaldehyde **32** in the presence of Pd/C to give benzimidazole **33**. Reduction of the pyridyl ring using PtO_2 or Pt/C provided the piperidine **30**. Pyridyl compounds **33** were also synthesized by coupling of bromobenzoic acids **34** with **27**, followed by benzimidazole formation in refluxing AcOH. Stille coupling with pyridyl stannanes **36** or Suzuki coupling of the analogous pyridyl boronic acids **37** provided pyridines **33**. Individual enantiomers of **22a**,**b** and **25a**,**b** were synthesized as shown in Scheme 2. Aryl bromide **38** was coupled with pyrrole boronic acid **39** under Suzuki conditions to give pyrrole **40**. Reduction using Pt/C provided pyrrolidine **41**. Chiral chromatography using a Whelk O column provided, after saponification of the esters, (*R*)-enantiomer **42a** and (*S*)-enantiomer **42b**. Coupling of the acids with diamine **27**, closure to the benzimidazole in refluxing AcOH, and removal of the *tert*-butoxycarbonyl (Boc)-protecting group using TFA gave (*R*)-enantiomer **22a** and (*S*)-enantiomer **22b**. A similar route was employed for the synthesis of piperidine analogues **25a**,**b**. Bromide **38** was coupled with pyridyl stannane **43** under Stille conditions to provide pyridine 44. Reduction of the pyridine ring and carbobenzyloxy (CBZ) protection gave **45**. Chiral chromatography using a Chiralcel OJ column provided, after saponification, (*R*)-enantiomer **46a** and (*S*)-enantiomer **46b**. Benzimidazole ring formation as described above for the pyrrolidine analogues and removal of the CBZ-protecting group under hydrogenolysis conditions gave (*R*)-enantiomer **25a** and (*S*)-enantiomers **25b**. Absolute configurations were determined by alternate asymmetric synthetic routes to be described in a separate publication.[15]

Results and Discussion

We previously described a series of potent benzimidazole-containing PARP-1 inhibitors, culminating in the identification of clinical candidate **1b**.[12] In this report, we sought to extend the substituent at the 2-position of the benzimidazole scaffold further into the adenosine-ribose binding pocket in an effort to not only expand upon the structure−activity relationship (SAR) of the benzimidazole class and modify associated physiochemical properties but also to increase potency by exploiting additional binding interactions in this binding pocket as previously described for other series of PARP inhibitors.[16]

The simple phenyl-substituted analogue **3a** (Table 1) showed modest enzyme activity, however, relatively poor cellular potency. As we have described previously in the benzimidazole class,[12] cellular penetration could be improved

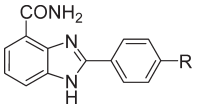
Scheme 2[a]

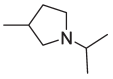
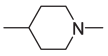
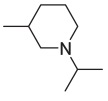
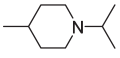


[a] Reagents and conditions: (a) Compound **39**, $Pd(PPh_3)_2Cl_2$, Na_2CO_3. (b) 5% Pt/C, H_2, HOAc. (c) (1) Chiral HPLC, Whelk O column; (2) LiOH. (d) (1) CDI, DMF, pyridine, **27**; (2) HOAc, heat. (e) TFA. (f) Compound **43**, $Pd_2(dba)_3$, (o-tol)$_3$P, DMF. (g) (1) H_2, Pd/C, HCl; (2) CBZ-Cl, K_2CO_3, dioxane, H_2O. (h) (1) Chiralcel OJ column; (2) LiOH. (i) H_2, Pd/C, MeOH.

by introducing basic amine solubilizing groups exemplified by the known[17] benzylic amine **3b**. Therefore, we wanted to expand upon this class by introducing pyrrolidine and piperidine substituents on this phenyl ring in place of simple benzylic amines. We also hoped to increase potency by exploiting additional hydrogen-binding interactions with such residues as Asp766 and Glu763, as has been previously demonstrated.[16−18] Introduction of a pyridine, although weakly basic, improved both enzyme and cellular potencies significantly (**4a**−**c**). (Generally, a 1.5−2-fold difference in K_i and IC_{50} values are significant). However, these analogues typically had rather poor solubility properties; thus, we focused our attention on saturated, nitrogen-containing ring systems. 2-Pyrrolidine analogue **5** showed excellent enzyme potency and improved cellular potency vs pyridines **4a**−**c**. However, the 3-pyrrolidine analogue **6** demonstrated rather poor cellular activity. *N*-Methylation maintained good potency for 2-pyrrolidine **7**, while restoring cellular potency in 3-pyrrolidine **8**. Larger *N*-alkyl groups exemplified by isopropyl analogues **9** and **10** showed a modest decrease in both enzyme and cellular potencies. Within the piperidine series, 2-substituted analogue **11** demonstrated excellent PARP-1 enzyme and cellular potency, whereas the 3- and 4-substituted analogues **12** and **13** showed only modest cellular activity. As with the pyrrolidines, *N*-methylation maintained good potency for 2-substituted analogue **14**, while enhancing the potencies for **15** and **16**. Also, larger groups such as isopropyl tended to modestly decrease cellular potency (**17**−**19**). Additional SAR investigations focused on the most promising classes, the N-unsubstituted 2-pyrrolidine and 2-piperidine series. Our previous work[12a] demonstrated little tolerance for elaboration at the 5- or 6- positions of the benzimidazole scaffold. This was indeed confirmed, with 6-fluoro analogues **20** and **24** (Table 2) maintaining good potency, while 6-chloro analogue **21** showed a significant drop in potency. A fluorine was also incorporated into the 2-position of the phenyl ring and was well-tolerated, with both **22** and **25** maintaining good enzyme and cellular potencies. The addition of fluorine to both benzimidazole and phenyl rings typically improved both enzyme and cellular potency, with **23** and **26** demonstrating enzyme potencies of 2 nM and cellular potencies of 1 and 2 nM, respectively. The individual enantiomers of the racemic monofluoro pyrrolidine and piperidine analogues **22** and **25** were also evaluated. While there was little difference between the two enantiomers of **25**, the (*S*)-enantiomer of **22** (**22b**) showed superior potency in both PARP enzyme and cellular assays as compared to the respective (*R*)-enantiomer (**22a**), highlighted by the 1 nM K_i and EC_{50} values that exhibited by **22b**. This is one of the most potent PARP inhibitors that we have identified to date. To aid in the further differentiation of these enantiomers, the pharmacokinetic properties of **22a**,**b** and **25a**,**b** were studied (Table 3). In the CD-1 mouse, (*S*)-enantiomers **22b** and **25b** showed only modestly higher oral exposures, with AUCs of 1.2 and 1.7 μg h/mL vs 1.0 and 1.6 μg h/mL for **22a** and **25a**, respectively. (*S*)-Enantiomers **22b** and **25b** were

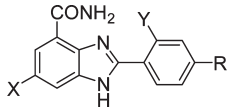
Table 1



Compd	R	PARP-1 (K_i, μM)	Cellular (EC_{50}, μM)[a]
3a	-H	0.017	0.075
3b	-CH_2NHMe	0.004	0.004
4a	2-pyridyl	0.002	0.008
4b	3-pyridyl	0.001	0.009
4c	4-pyridyl	0.002	0.012
5		0.003	0.003
6		0.003	0.036
7		0.005	0.004
8		0.005	0.004
9		0.011	0.006
10		0.013	0.007*
11		0.004	0.004
12		0.003	0.021
13		0.002	0.015
14		0.006	0.002
15		0.004	0.005
16		0.008	0.006
17		0.010	0.008*
18		0.012	0.026*
19		0.007	0.012*

[a] Average of ≥2 determinations unless noted by *.

Table 2



Compd	R	X	Y	PARP-1 (K_i, μM)	Cellular (EC_{50}, μM)[a]
20		-F	-H	0.002	0.002
21		-Cl	-H	0.009	0.026
22		-H	-F	0.003	0.004
22a		-H	-F	0.004	0.009
22b		-H	-F	0.001	0.001
23		-F	-F	0.002	0.001
24		-F	-H	0.003	0.001
25		-H	-F	0.006	0.006
25a		-H	-F	0.009	0.003
25b		-H	-F	0.006	0.006
26		-F	-F	0.002	0.002*

[a] Average of ≥2 determinations unless noted by *.

Table 3. Multispecies Pharmacokinetics of **22a**,**b** and **25a**,**b**

compd	species	dose[a]	% F	$T_{1/2}$ (iv)[b]	AUC (po)[c]	C_{max} (po)[d]	V_β[e]	CL[f]
22a	mouse	10	36	0.9	1.0	0.5	4.6	3.6
22b	mouse	10	36	0.8	1.2	0.6	3.3	2.9
	rat	5	55	1.9	1.7	0.3	4.1	1.7
	dog	2.5	72	1.7	1.1	0.5	3.6	1.7
	monkey	2.5	34	1.8	0.2	0.03	9.2	4.1
25a	mouse	10	100	1.4	1.6	0.3	15	7.3
25b	mouse	10	63	1.3	1.7	0.5	6.6	3.6
	rat	5	80	4.5	1.8	0.2	14	2.3
	dog	2.5	39	2.0	0.3	0.1	9.3	3.4
	monkey	2.5	61	5.0	0.75	0.1	15	2.2

[a] po, mg/kg. [b] h. [c] μg h/mL. [d] μg/mL. [e] L/kg. [f] L/h/kg.

further characterized in Sprague−Dawley rats, beagle dogs, and cynomolgus monkeys, with **22b** demonstrating oral bioavailabilities of 34−72% and half-lives of 1.7−1.9 h and **25b** with oral bioavailabilities of 39−80% and half-lives of 2−5 h. In addition, all four compounds **22a**,**b** and **25a**,**b** were shown to cross the blood−brain barrier in CD-1 mice (Table 4), with **22a**,**b** showing plasma-to-brain ratios of ∼2:1, while **25a**,**b** demonstrated approximately a 1:1 ratio. The ability of these compounds to penetrate the brain was considered a desirable property due to the role that TMZ plays in the treatment of gliomablastomas and the potential ability to enhance the efficacy of TMZ in a human clinical setting.

Selected compounds were also tested against the closely related PARP-2 enzyme, and all had similar K_i values (i.e., 1−26 nM). This is consistent with what we have previously reported that the majority of compounds within the benzimidazole class showed similar potencies against both enzymes.[12]

An X-ray cocrystal structure of PARP-1 with **25b** is shown in Figure 1. The key interactions of the benzimidazole carboxamide core in the PARP-1 active site, consistent with previous

literature reports, are highlighted. Both Ser-904 and Gly-863 are involved in key hydrogen-bond interactions with the carboxamido group of **25b**, with a π-stacking interaction between the benzimidazole ring and the Tyr-907. In addition, Glu-988 is involved in a water-mediated hydrogen bond with the −NH of the benzimidazole ring system. There is also an interaction of the piperidine nitrogen with Asp-766 and a water molecule, which may contribute to the enhanced potency of some of these analogues. The lack of this interaction due to steric interactions may also contribute to the somewhat reduced potency of the more hindered isopropyl substituted analogues. In addition, there are also two water molecules in the vicinity of the fluorine atom, which may result in favorable dipolar interactions.

Table 4. Mouse Plasma/Brain PK

		AUC (po)[b]	
compd	dose[a]	plasma	brain
22a	30	4.2	1.8
22b	30	9.8	4.2
25a	30	5.8	6.3
25b	30	9.9	9.1

[a] mg/kg. [b] μg h/mL.

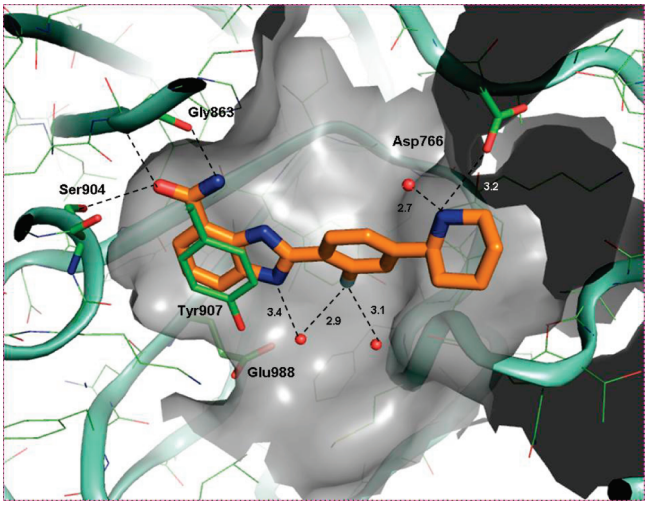
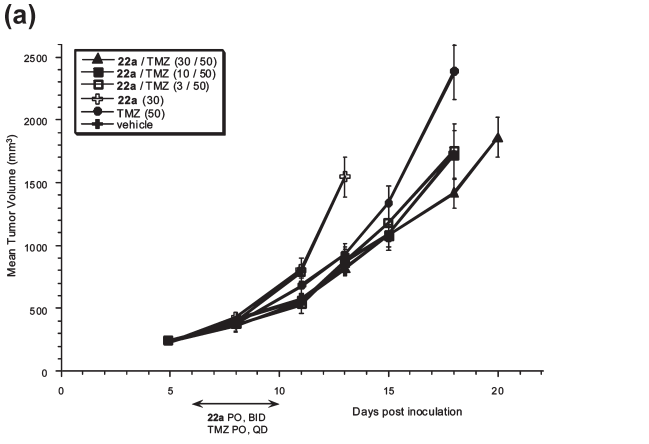


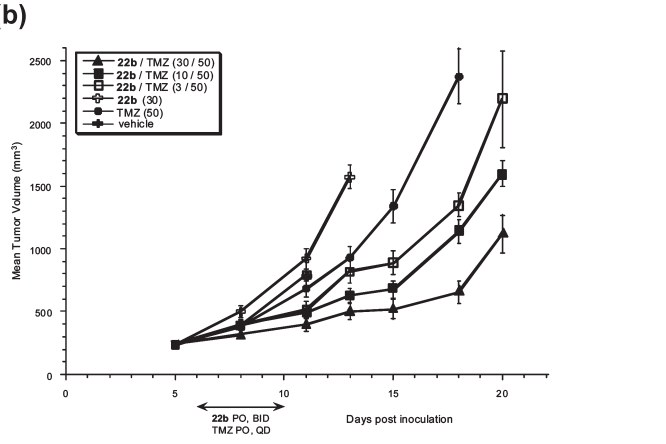
Figure 1. X-ray cocrystal structure of PARP-1 and **25b**.

In vivo, **22a**,**b** both demonstrated significant enhancement of the efficacy of TMZ in a murine B16F10 syngeneic melanoma model (Figure 2A,B), with the **22b** combination groups showing superior efficacy. The B16F10 model, while relatively resistant to most chemotherapeutics, is moderately sensitive to TMZ, and this sensitivity can be enhanced with PARP inhibitors. Compounds **22a**,**b** were administered orally on days 6−10 at doses of 3, 10, and 30 mg/kg/day, bid, while TMZ was administered orally at 50 mg/kg/day, qd, on days 6−10. Compound **22b** significantly enhanced the efficacy of TMZ in a dose-dependent manner. Significant potentiation was observed as early as day 12, with TGI (tumor growth inhibition) values (vs vehicle control) of 35, 38, and 49% for the 3, 10, and 30 mg/kg/day **22b** combination groups, respectively, as compared to 14% for TMZ alone. All three dosing groups continued to differentiate from the TMZ group out to day 18, with TGI values (vs TMZ control) of 43, 52, and 72% for the 3, 10, and 30 mg/kg/day **22b** combination groups, respectively. On the other hand, **22a** showed significant potentiation of TMZ at day 18 only with the 30 mg/kg/day dose, with a TGI value of 40%. Compounds **25a**,**b** also enhanced the efficacy of TMZ in the B16F10 model (Figure 3A,B). Both compounds were administered orally on days administered orally on days 6−10 at doses of 3, 10, and 30 mg/kg/day, bid, while TMZ was administered orally at 50 mg/kg/day, qd, on days 6−10. Both **25a**,**b** significantly enhanced the efficacy of TMZ in a dose-dependent manner. Significant potentiation was observed as early as day 12, with TGI values (vs vehicle control) of 46, 66, and 66% for the 3, 10, and 30 mg/kg/day **25a** combination groups, and 45, 68, and 72% for the 3, 10, and 30 mg/kg/day **25b** combination groups, respectively, as compared to 67% for TMZ alone. The 30 mg/kg/day **25a** combination group and the 10 and 30 mg/kg/day **25b** combination groups continued to differentiate



compd	dose (mg/kg/day)	tumor volume[a] (day 12)	%TGI[b] (day 12)	tumor volume[a] (day 18)	%TGI[c] (day 18)
22a/TMZ	30/50	568 ± 41	28	1418 ± 120	40*
	10/50	555 ± 55	30	--	--
	3/50	532 ± 68	33	1748 ± 220	27
vehicle/TMZ	0/50	681 ± 63	14	2379 ± 219	--
22a/vehicle	30/0	810 ± 91	0	--	--
combination vehicle	0/0	793 ± 111	--	--	--

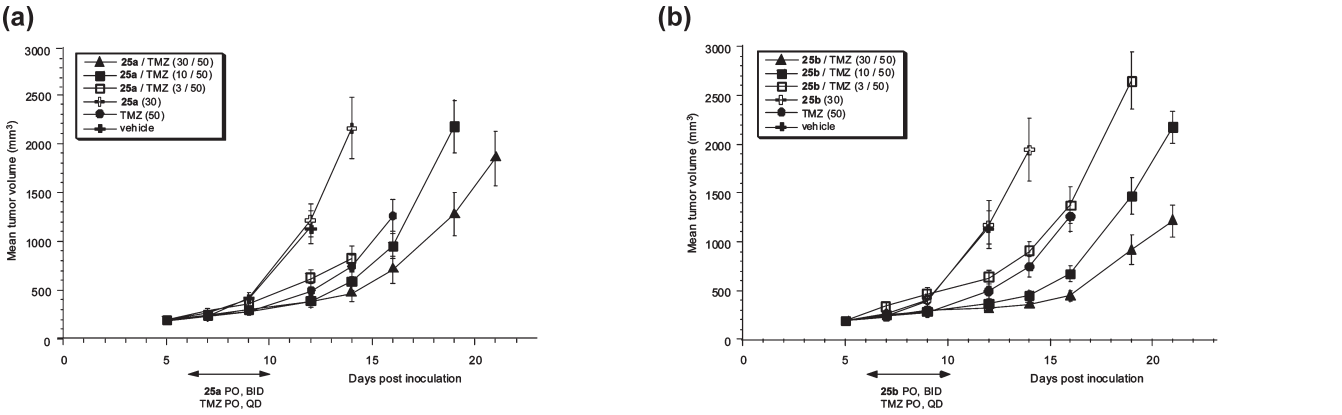
* p <0.001; [a] Mean (mm³) ± SEM of 9-10 mice/group; [b] vs. combo vehicle control; [c] vs. TMZ control.



compd	dose (mg/kg/day)	tumor volume[a] (day 12)	%TGI[b] (day 12)	tumor volume[a] (day 18)	%TGI[c] (day 18)
22b/TMZ	30/50	401 ± 57	49*	565 ± 90	72****
	10/50	493 ± 41	38**	1137 ± 95	52****
	3/50	514 ± 67	35***	1351 ± 94	43*****
vehicle/TMZ	0/50	681 ± 63	14	2379 ± 219	--
22b/vehicle	30/0	920 ± 82	0	--	--
combination vehicle	0/0	793 ± 111	--	--	--

* p =0.006, ** p =0.02, *** p =0.05, **** p <0.0001, ***** p <0.0004; [a] Mean (mm³) ± SEM of 9-10 mice/group; [b] vs. combo vehicle control; [c] vs. TMZ control.

Figure 2. B16F10 model: (a) **22a** and (b) **22b** in combination with TMZ.



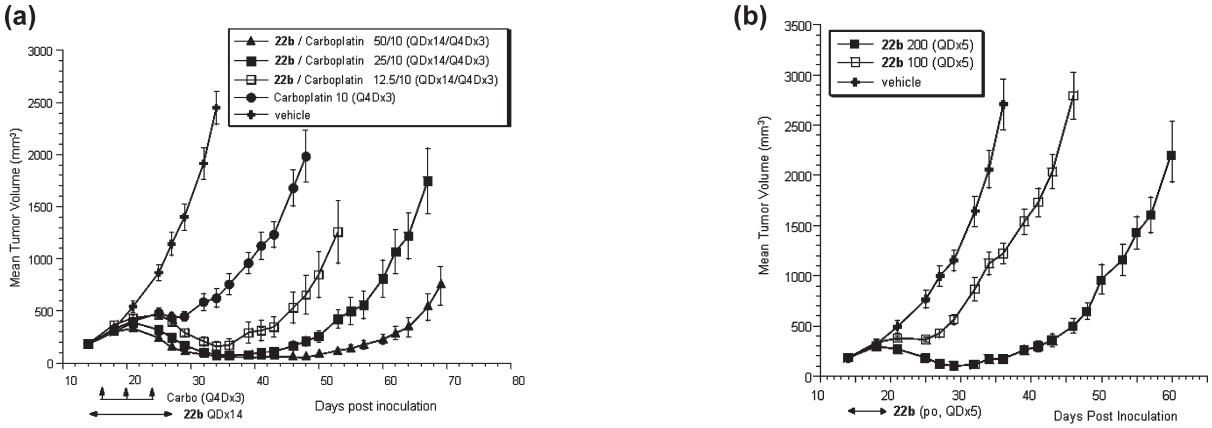
compd	dose (mg/kg/day)	tumor volume[a] (day 12)	%TGI[b] (day 12)	tumor volume[a] (day 16)	%TGI[c] (day 16)
25a/TMZ	30/50	386 ± 67	66*	710 ± 140	44****
	10/50	390 ± 47	66**	952 ± 133	25
	3/50	619 ± 88	46***	--	--
vehicle/TMZ	0/50	491 ± 75	67	1261 ± 161	--
25a/vehicle	30/0	1215 ± 167	0	--	--
combination vehicle	0/0	1147 ± 167	--	--	--

* p =0.0005, ** p =0.0004, *** p =0.01, ***** p =0.02; [a] Mean (mm^3) ± SEM of 9-10 mice/group; [b] vs. combo vehicle control; [c] vs. TMZ control.

compd	dose (mg/kg/day)	tumor volume[a] (day 12)	%TGI[b] (day 12)	tumor volume[a] (day 16)	%TGI[c] (day 16)
25b/TMZ	30/50	318 ± 34	72*	448 ± 54	64*
	10/50	363 ± 35	68**	670 ± 80	47****
	3/50	626 ± 84	45***	1375 ± 190	0
vehicle/TMZ	0/50	491 ± 75	67	1261 ± 161	--
25b/vehicle	30/0	1174 ± 248	0	--	--
combination vehicle	0/0	1147 ± 167	--	--	--

* p =0.0001, ** p =0.0002, *** p =0.01, **** p =0.004; [a] Mean (mm^3) ± SEM of 9-10 mice/group; [b] vs. combo vehicle control; [c] vs. TMZ control.

Figure 3. B16F10 model: (a) **25a** and (b) **25b** in combination with TMZ.



compd	dose (mg/kg/day)	tumor volume[a] (day 34)	%TGI[b] (day 34)	tumor volume[a] (day 48)	%TGI[c] (day 48)
22b/carboplatin	50/10	67± 6	97*	54 ± 7	97*
	25/10	83 ± 7	97*	209 ± 45	89*
	12.5/10	166 ± 42	93*	657 ± 184	67*
vehicle/carboplatin	0/10	624 ± 89	69*	1983 ± 249	--
combination vehicle	0/0	2448 ± 157	--	--	--

* p< 0.001; [a] Mean (mm^3) ± SEM of 10 mice/group; [b] vs. combo vehicle control; [c] vs. carboplatin control.

22b dose (mg/kg/day)	tumor volume[a] (day 34)	%TGI[b] (day 34)
200	168 ± 37	92***
100	1122 ± 115	46***
0	2059 ± 184	--

p * p< 0.05, ** p< 0.01, *** p< 0.001; [a] Mean (mm^3) ± SEM of 8-9 mice/group; [b] vs. vehicle control.

Figure 4. (a) MX-1 model: **22b** in combination with carboplatin. (b) MX-1 model: Single agent **22b**.

from the TMZ group out to day 16, with TGI values (vs TMZ control) of 44% for the 30 mg/kg/day **25a** and 47 and 64% for the 10 and 30 mg/kg/day **25b** combination groups, respectively. The **22a**,**b** and **25a**,**b** TMZ combinations were all well-tolerated, with maximum body weight loss for all combination groups similar to the TMZ monotherapy group. Overall, both (*S*)-enantiomers **22b** and **25b** showed superior enhancement of the efficacy of TMZ (in terms of %TGI) in this model relative to their respective (*R*)-enanatiomers.

Because of a superior profile in the B16 model, **22b** was characterized further in vivo. Plasma and tumor levels of **22b** were assessed after 5 days of oral dosing in a separate B16F10 study using a 25 mg/kg/day, bid dose of **22b** in combination with TMZ (50 mg/kg/day, qd). Significant distribution of **22b** to the tumor was observed 6 h after the final dose, with a concentration of 21 μg/mL in the tumor vs 0.38 μg/mL in the plasma. Similar concentrations were obtained when **22b** was dosed alone (17.2 vs 0.22 μg/mL).

Compound **22b** was further characterized in a BRCA1-deficient MX-1 breast carcinoma model both in combination with carboplatin (Figure 4A) and as a single agent (Figure 4B), with **22b** dosed orally in a once a day dosing regimen in both studies. Female *SCID* mice were dosed with **22b** at doses of 12.5, 25, and 50 mg/kg/day, qd, for 14 days starting on day 14 post-tumor inoculation, while carboplatin was given as a single i.p. dose on days 16, 20, and 24 at 10 mg/kg. Significant potentiation was observed as early as day 34 with TGI values (vs vehicle control) of 93, 97, and 97% for the 12.5, 25, and 50 mg/kg/day **22b** combination groups, respectively, as compared to 69% TGI for carboplatin alone. The three combination groups continued to differentiate from the carboplatin group out to day 48, with TGI values (vs carboplatin control) of 67, 89, and 97% for the 12.5, 25, and

50 mg/kg/day **22b** combination groups, respectively. In addition, **22b** demonstrated significant single agent efficacy in this model (Figure 4B). Compound **22b** was dosed orally at 100 and 200 mg/kg/day, qd, for 5 days starting on day 15 post-tumor inoculation. Significant efficacy was observed at day 34 with TGI values (vs vehicle control) of 46 and 92% for the 100 and 200 mg/kg/day **22b** groups, respectively.

Conclusion

In summary, the discovery and characterization of a novel PARP inhibitor, **22b**, has been described. This exceptionally potent compound has demonstrated significant efficacy in two tumor models, enhancing the efficacy of both TMZ and carboplatin. In addition, this compound showed significant single agent activity in a BRCA-1-deficient MX-1 breast carcinoma model. This compound has excellent pharmacokinetic properties, is able to cross the blood−brain barrier, and appears to distribute well into tumor tissue. Compound **22b** represents a promising, structurally diverse benzimidazole analogue and is being further characterized preclinically.

Experimental Section

NMR spectra were obtained on Varian M-300, Bruker AMX-400, Varian U-400, or Varian Unity Inova 500 magnetic resonance spectrometers with indicated solvent and internal standard. Chemical shifts are given in delta (δ) values and coupling constants (J) in Hertz (Hz). The following abbreviations are used for peak multiplicities: s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet; and br, broadened. Mass spectra were performed as follows: ESI (electrospray ionization) was performed on a Finnigan SSQ7000 MS run as a flow injection acquisition; DCI (desorption chemical ionization) was performed on a Finnigan SSQ7000 MS using a direct exposure probe with ammonia gas; and APCI (atmospheric pressure chemical ionization) was performed on a Finnigan Navigator MS run as flow injection acquisition. Elemental analyses were performed by Quantitative Technologies Inc. (Whitehouse, NJ). All manipulations were performed under nitrogen atmosphere unless otherwise noted. All solvents and reagents were obtained from commercial sources and used without further purification. High-performance liquid chromatography (HPLC) purifications were carried out using a Zorbax C-18, 250 × 2.54 column and elution with a 0−100% gradient of mobile phase A [0.1% trifluoroacetic acid (TFA) in water] and mobile phase B (0.1% TFA in CH_3CN). Analytical liquid chromatography−mass spectrometry (LC-MS) was performed on a Finnigan Navigator mass spectrometer and Agilent 1100 HPLC system operating under positive APCI ionization conditions. The column used was a Phenomenex Luna Combi-HTS C8(2) 5 μm 100 Å (2.1 mm ×30 mm) with a gradient of 10−100% acetonitrile and 0.1% TFA in water or a gradient of 10−100% acetonitrile and 10 mM NH_4OAc in water. Analytical LC-MS or combustion analysis indicated that the purity of all compounds was not less than 95%, unless otherwise noted.

PARP Enzyme Assay.[4a] The enzyme assay was conducted in buffer containing 50 mM Tris, pH 8.0, 1 mM dithiothreitol (DTT), and 4 mM $MgCl_2$. PARP reactions contained 1.5 μM [^{3}H]-NAD^+ (1.6 μCi/mmol), 200 nM biotinylated histone H1, 200 nM slDNA, and 1 nM PARP-1 or 4 nM PARP-2 enzyme. Autoreactions utilizing SPA bead-based detection were carried out in 100 μL volumes in white 96-well plates. Reactions were initiated by adding 50 μL of 2X NAD^+ substrate mixture to 50 μL of 2X enzyme mixture containing PARP and DNA. These reactions were terminated by the addition of 150 μL of 1.5 mM benzamide (~1000-fold over its IC_{50}). A 170 μL amount of the stopped reaction mixtures was transferred to streptavidin-coated Flash Plates, incubated for 1 h, and counted using a TopCount microplate scintillation counter. K_i data were determined from inhibition curves at various substrate concentrations.

Cellular PARP Assay.[4a] C41 cells were treated with test compound for 30 min in a 96-well plate. PARP was activated by damaging DNA with 1 mM H_2O_2 for 10 min. Cells were washed with ice-cold phosphate-buffered saline (PBS) once and fixed with prechilled methanol/acetone (7:3) at −20 °C for 10 min. After they were air-dried, plates were rehydrated with PBS and blocked using 5% nonfat dry milk in PBS-tween (0.05%) (blocking solution) for 30 min at room temperature. Cells were incubated with anti-PAR antibody 10H (1:50) in blocking solution at room temperature for 60 min followed by washing with PBS-Tween20 five times, and incubation with goat antimouse fluorescein 5(6)-isothiocyanate (FITC)-coupled antibody (1:50) and 1 μg/mL 4′,6-diamidino-2-phenylindole (DAPI) in blocking solution at room temperature for 60 min. After washing with PBS-Tween20 5 times, analysis was performed using an fmax Fluorescence Microplate Reader set at the excitation and emission wavelength for FITC or the excitation and emission wavelength for DAPI. PARP activity (FITC signal) was normalized with cell numbers (DAPI).

Mouse Pharmacokinetic Analysis. Plasma samples were aliquoted into 96-well plates, and proteins were precipitated using acidified methanol. Tissue samples were prepared by homogenization with 2 volumes of saline followed by protein precipitation with acetonitrile. Supernatants were stored at −20 °C. Samples analyses were performed by LC-MS using a Shimadzu 10A-VP chromatography system with a Phenomenex Polar RP-5 cm column. The mobile phase consisted of mixtures of acetonitrile and 0.1% acetic acid in water with a flow rate of 0.4 mL/min. Mass detection was accomplished with an ESI equipped LCQ-Duo by ThermoFinnegan. External standards were prepared from spiked control plasma or tissue homogenate and used to generate a response factor for every study. Limits of detection were between 10 and 30 nM.

B16F10 Tumor Model.[4a] For B16F10 syngeneic studies, 6×10^4 cells were mixed with 50% matrigel (BD Biosciences, Bedford, MA) and inoculated by s.c. injection into the flank of 6−8 week old female C57BL/6 mice, 20 g (Charles River Laboratories, Wilmington, MA). Mice were injection-order allocated to treatment groups, and PARP inhibitor therapy was initiated on day 6 following inoculation, with TMZ treatment also starting on day 6.

MX-1 Tumor Model.[4a] A 0.2 cc amount of a 1:10 dilution of tumor brei in 45% Matrigel and 45% Spinner MEM (Life Technologies) was injected subcutaneously into the flank of female *SCID* mice (Charles River Laboratories) on study day 0. Tumors were allowed to grow to the indicated size and then randomized to therapy groups (N = 10 mice/group). PARP inhibitor therapy began on day 14, with cisplatin treatment starting on day 16. At various intervals following tumor inoculation, the individual tumor dimensions were serially measured using calibrated microcalipers, and the tumor volumes were calculated according to the formula $V = L \times W^2/2$ (V, volume; L, length; and W, width). Effects on tumor growth rate were assessed by determining %T/C [(mean tumor volume of treated group on day X/mean tumor volume of control group on day X) × 100] and %TGI (100 − %T/C) for a given treatment relative to vehicle or monotherapy treatment.

X-ray Crystallography Data. A crystallization attempt of apo-PARP-1 with a GST fusion h-PARP-1 (654−1014) expressed in *Escherichia coli* was not successful. Because crystals of PARP-1/2-(trifluoromethyl)-1*H*-benzimidazole-4-carboxamide complex could be obtained relatively easily, a ligand-exchange soaking technique was adopted for the structure of PARP-1/**25b** complex. The fact that the equilibrium constant K_i of 2-(trifluoromethyl)-1*H*-benzimidazole-4-carboxamide is 0.58 μM and that of **25b** is 0.006 μM helped the ligand-exchange experiment. Crystals of PARP-1/2-(trifluoromethyl)-1*H*-benzimidazole-4-carboxamide complex were obtained by the hanging drop

method at 17 °C. The protein solution was 60 mg/mL (0.874 mM) of PARP in 50 mM, pH 7.5, Tris buffer containing 150 mM NaCl and 1.5 mM DTT. The ligand (2-(trifluoromethyl)-1*H*-benzimidazole-4-carboxamide) concentration in the protein solution was 2 mM. The well solution had 0.8 M NaCl and 1.8 M ammonium sulfate in water, and the hanging drop was a 1:1 mixture of protein solution and well solution. The space group of PARP-1/2-(trifluoromethyl)-1*H*-benzimidazole-4-carboxamide complex crystal is *P*321 with cell dimensions of $a = b = 94.21$ Å, $c = 68.86$ Å, $\alpha = \beta = 90°$, and $\gamma = 120°$. The bound compound, 2-(trifluoromethyl)-1*H*-benzimidazole-4-carboxamide, in the crystal could be displaced with **25b** by addition of 1 mM **25b** into the crystal drop overnight. X-ray diffraction data were collected at Advanced Photon Source Beamline 17-ID of Argonne National Laboratory with ADSC CCD detector Quantum 210. Ethylene glycol (20%) in 1.2 M NaCl and 1.6 M ammonium sulfate solution was used as a cryoprotectant for data collection at 110 K. The complex crystal of PARP-1/**25b** diffracted up to 2.5 Å, and 12464 unique reflections were collected and scaled using HKL2000. The overall R_{sym} (I) is 0.096, and I/σ (I) is 12.0 with the overall completeness of 99%. The structure was solved using CCP4 molrep program with an in-house search model of PARP complex and refined with CNX2002 and BUSTER. The conventional and free *R*-factors after refinement were 0.205 and 0.275. The coordinates of the PARP-1/**25b** complex are deposited in the Protein Data bank with the code 3L3M.

General Procedure A. Preparation of 2-(4-Piperidin-4-ylphenyl)-1*H*-benzimidazole-4-carboxamide (13). A solution of *tert*-butyl 4-(4-carboxyphenyl)piperidine-1-carboxylate (1 g, 3.3 mmol) in pyridine (3 mL) and DMF (3 mL) at 40 °C was stirred for 30 min. Carbonyl diimidazole (CDI, 0.55 g, 3.4 mmol) was added, and the mixture was stirred for 1 h. 2,3-Diaminobenzamide dihydrochloride[12] (0.73 g, 3.3 mmol) was added, and the mixture was stirred for 1 h at ambient temperature. Isopropanol (10 mL) was added, and the mixture was stirred at 0 °C for 18 h and filtered. The solid was dissolved in water (10 mL), treated with 50% aqueous NaOH (0.26 mL), stirred for 3 h at ambient temperature, and filtered to give crude *tert*-butyl 4-(4-(2-amino-3-carbamoylphenylcarbamoyl)phenyl)piperidine-1-carboxylate (0.965 g, 69%). A solution of this solid (0.175 g, 0.4 mmol) in acetic acid (2 mL) was stirred at reflux for 90 min and concentrated. The concentrate was dissolved in water, treated with 50% aqueous NaOH (0.2 mL), and filtered. The filtrate was concentrated and purified by HPLC to give the title compound (87%). ^{1}H NMR (DMSO-d_6): δ 1.84 (qd, J = 13.0, 3.9 Hz, 2H), 2.00 (s, 2H), 2.97 (ddd, J = 12.0, 8.6, 3.4 Hz, 1H), 3.01−3.09 (m, 2H), 3.42 (d, J = 12.5 Hz, 2H), 7.35 (t, J = 7.8 Hz, 1H), 7.46 (d, J = 8.1 Hz, 2H), 7.74 (d, J = 7.2 Hz, 2H), 7.87 (d, J = 6.6 Hz, 1H), 8.22 (d, J = 8.4 Hz, 1H), 8.34 (s, 1H), 8.61 (s, 1H), 9.25 (s, 1H). Anal. ($C_{19}H_{20}N_4O\cdot 3.2$TFA) C, H, N.

General Procedure B. Preparation of 2-(4-(1-Methylpiperidin-2-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (14). To a solution of **11** (0.05 g, 0.2 mmol) and 36% formaldehyde in water (0.012 mL) in MeOH (1 mL) were added sodium cyanoborohydride (0.01 g, 0.2 mmol) and AcOH (0.2 mL), and the mixture was stirred at ambient temperature for 18 h. After concentration, the residue was stirred with TFA in dichloromethane and concentrated. Purification by flash chromatography on silica gel using 10% MeOH/dichloromethane afforded the title compound (0.043 mg, 83%). ^{1}H NMR (DMSO-d_6): δ 1.39 (m, 1H), 1.67 (s, 3H), 1.79 (d, J = 14.7 Hz, 2H), 1.91 (s, 5H), 2.82 (s, 1H), 2.99 (s, 1H), 7.35 (m, 1H), 7.53 (d, J = 7.8 Hz, 2H), 7.72 (d, J = 6.9 Hz, 2H), 7.87 (d, J = 7.5 Hz 1H), 8.20 (m, 2H), 9.35 (s, 1H). Anal. ($C_{20}H_{22}N_4O\cdot 1.15$TFA) C, H, N.

2-(4-Piperidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (4a). A mixture of 2,3-diaminobenzamide dihydrochloride[12] (1 g, 4.5 mmol), 4-pyridin-2-ylbenzaldehyde (0.82 g, 4.5 mmol), and 10% Pd/C (0.3 g) in MeOH (30 mL) was stirred at reflux for 18 h, cooled, filtered through Celite, and concentrated. The residue was crystallized from MeOH to provide the title compound (1.2 g, 86%). ^{1}H NMR (DMSO-d_6): δ 7.37 (t, J = 7.8 Hz, 1H), 7.42 (dd, J = 6.9, 4.7 Hz, 1H), 7.77 (d, J = 7.9 Hz, 1H), 7.80 (br, 1H), 7.89 (d, J = 7.3 Hz, 1H), 7.95 (dt, J = 7.8, 1.7 Hz, 1H), 8.10 (d, J = 7.9 Hz, 1H), 8.33 (d, J = 8.3 Hz, 2H), 8.37 (d, J = 8.3 Hz, 2H), 8.73 (d, J = 4.0 Hz, 1H), 9.39 (br, 1H). Anal. ($C_{19}H_{14}N_4O\cdot 1.9HCl\cdot 4.2H_2O$) C, H, N, Cl.

2-(4-Piperidin-3-ylphenyl)-1*H*-benzimidazole-4-carboxamide (4b). Step 1. Preparation of 2-(4-Bromophenyl)-1*H*-benzimidazole-4-carboxamide. The title compound was prepared from 4-bromobenzaldehyde using the procedure described for **4a** (158 mg, 52%).

Step 2. Preparation of 2-(4-Pyridin-3-ylphenyl)-1*H*-benzimidazole-4-carboxamide. A mixture of the product of step 1 (150 mg, 0.47 mmol), pyridin-3-ylboronic acid (70 mg, 0.57 mmol), Pd-(dppf)$_2$Cl$_2$ (40 mg), and Na_2CO_3 (55 mg) in dioxane (4 mL) was heated at 90 °C for 18 h. After it was cooled, filtered through Celite, and concentrated, the residue was purified by HPLC to give the title compound. ^{1}H NMR (DMSO-d_6): δ 7.38 (t, J = 7.8 Hz, 1H), 7.76 (br, 1H), 7.79 (dd, J = 7.9, 1.3 Hz, 1H), 7.87 (dd, J = 8.1, 5.4 Hz, 1H), 7.90 (dd, J = 7.7, 1.2 Hz, 1H), 8.05 (d, J = 8.6 Hz, 2H), 8.41 (d, J = 8.6 Hz, 2H), 8.60 (dt, J = 8.6, 1.8 Hz, 1H), 8.79 (dd, J = 5.3, 1.3 Hz, 1H), 9.20 (d, J = 2.2 Hz. 1H), 9.21 (br, 1H). Anal. ($C_{19}H_{14}N_4O\cdot 1.6$TFA) C, H, N.

2-(4-(Pyridin-4-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (4c). The title compound was prepared from 4-pyridin-4-ylbenzaldehyde using the procedure described for **4a** (0.40 g, 29%). ^{1}H NMR (DMSO-d_6): δ 7.41 (t, J = 7.7 Hz, 1H), 7.81 (d, J = 7.0 Hz, 1H), 7.82 (br, 1H), 7.91 (dd, J = 7.6, 1.2 Hz, 1H), 8.24 (d, J = 8.5 Hz, 2H), 8.36 (d, J = 6.7 Hz, 2H), 8.48 (d, J = 8.9 Hz, 2H), 8.96 (d, J = 6.7 Hz, 2H), 9.23 (br, 1H). MS (ESI): m/z 315 $(M + H)^+$.

2-(4-Pyrrolidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (5). Step 1. Preparation of *tert*-Butyl 2-(4-(4-carbamoyl-1*H*-benzimidazol-2-yl)phenyl)pyrrolidine-1-carboxylate. The title compound was prepared from *N*-Boc-4-pyrrolidin-2-ylbenzoic acid using general procedure A (0.24 g, 59%). ^{1}H NMR (DMSO-d_6): δ 1.12 (s, 5H), 1.41 (s, 4H), 1.80 (m, 3H), 2.36 (m, 1H), 3.55 (m, 2H), 4.79 (m, 1H), 7.34 (t, J = 7.8 Hz, 1H), 7.40 (br d, J = 8.5 Hz, 2H), 7.72 (d, J = 8.2 Hz, 1H), 7.75 (br, 1H), 7.87 (d, J = 7.4 Hz, 1H), 8.18 (br d, J = 7.8 Hz, 2H), 9.35 (br, 1H), 13.35 (br, 1H).

Step 2. Preparation of 2-(4-Pyrrolidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (5). The product of step 1 (0.23 g) in 1 M HCl in EtOH (5 mL) was stirred for 19 h. TLC analysis indicated incomplete reaction, so the mixture was treated with 12 M HCl (0.5 mL), stirred for 19 h, and then treated with additional 12 M HCl (0.5 mL) and stirred for 6 h. After it was concentrated, the residue was purified by flash chromatography on silica gel using a gradient of 95:5:1 to 80:20:1 dichloromethane/MeOH/NH_4OH to provide the title compound (0.10 g, 63%). ^{1}H NMR (DMSO-d_6): δ 1.56 (m, 1H), 1.80 (m, 2H), 2.20 (m, 1H), 3.01 (m, 2H), 4.18 (t, J = 7.6 Hz, 1H), 7.33 (t, J = 7.8 Hz, 1H), 7.59 (d, J = 7.8 Hz, 2H), 7.73 (d, J = 7.1 Hz, 1H), 7.74 (br, 1H), 7.85 (d, J = 7.4 Hz, 1H), 8.17 (d, J = 8.4 Hz, 2H), 9.34 (br, 1H).

3-(4-Pyrrolidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (6). The title compound was prepared as described for the synthesis of **5**, using *N*-Boc-4-pyrrolidin-3-ylbenzoic acid in place of *N*-Boc-4-pyrrolidin-2-ylbenzoic acid (0.112 g, 37%). ^{1}H NMR (DMSO-d_6): δ 1.86 (m, 1H), 2.28 (m, 1H), 2.89 (m, 1H), 3.09 (m, 1H), 3.22 (m, 1H), 3.32−3.46 (m, 2H), 7.33 (t, J = 7.8 Hz, 1H), 7.52 (d, J = 8.5 Hz, 2H), 7.73 (d, J = 7.9 Hz, 1H), 7.75 (br, 1H), 7.86 (d, J = 7.4 Hz, 1H), 8.19 (d, J = 8.5 Hz, 2H), 9.29 (br, 1H). Anal. ($C_{18}H_{18}FN_4O\cdot 2.1$TFA) C, H, N.

2-(4-(1-Methylpyrrolidin-2-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (7). The title compound was prepared from **5** and formaldehyde using general procedure B (66 mg, 56%). ^{1}H NMR (CD_3OD): δ 2.36 (m, 3H), 2.62 (m, 1H), 2.84 (s, 3H), 3.35 (m, 1H), 3.92 (m, 1H), 4.49 (m, 1H), 7.46 (m, 1H), 7.78 (d, J = 8.2 Hz, 2H), 7.82 (d, J = 8.2 Hz, 1H),

7.98 (d, J = 7.6 Hz, 1H), 8.31 (d, J = 8.5 Hz, 2H). MS (DCI): m/z 321 $(M + H)^+$.

2-(4-(1-Methylpyrrolidin-3-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (8). The title compound was prepared from **6** and formaldehyde using general procedure B (80 mg, 69%). ^{1}H NMR (CD_3OD): δ 2.29 (m, 1H), 2.61 (m, 1H), 3.06 (s, 3H), 3.29 (m, 1H), 3.71 (m, 2H), 3.96 (m, 2H), 7.52 (t, J = 7.9 Hz, 1H), 7.62 (d, J = 7.9 Hz, 2H), 7.86 (d, J = 7.9 Hz, 1H), 7.98 (d, J = 7.6 Hz, 1H), 8.17 (d, J = 8.2 Hz, 2H). MS (DCI): m/z 321 $(M + H)^+$.

2-(4-(1-Isopropylpyrrolidin-2-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (9). The title compound was prepared from **5** and acetone using general procedure B (34 mg, 30%). ^{1}H NMR (CD_3OD): δ 1.34 (dd, J = 6.6, 2.0 Hz, 6H), 2.30 (m, 3H), 2.60 (m, 1H), 3.51 (m, 2H), 3.70 (m, 1H), 4.74 (m, 1H), 7.49 (t, J = 7.9 Hz, 1H), 7.83 (m, 3H), 7.99 (d, J = 7.6 Hz, 1H), 8.32 (d, J = 8.5 Hz, 2H). MS (DCI): m/z 349 $(M + H)^+$.

2-(4-(1-Isopropylpyrrolidin-3-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (10). To a solution of **5** (100 mg, 0.33 mmol) in MeOH (10 mL) was added acetone (38 mg, 0.54 mmol), and the mixture was stirred at ambient temperature for 40 min. Sodium triacetoxyborohydride (253 mg, 1.2 mmol) and AcOH (100 μL) were added, and the mixture was stirred at ambient temperature for 18 h. Dichloromethane and water were added, and the organic layer was washed with dilute NaOH and water and concentrated. Purification by HPLC provided the title compound (30 mg, 27%). ^{1}H NMR (CD_3OD): δ 1.44 (d, J = 6.7 Hz, 6H), 2.25 (m, 1H), 2.59 (m, 1H), 3.30 (m, 1H), 3.57 (m, 2H), 3.69 (m, 1H), 3.77−3.88 (m, 2H), 4.02 (dd, J = 10.7, 7.0 Hz, 1H), 7.54 (t, J = 7.9 Hz, 1H), 7.65 (d, J = 7.9 Hz, 2H), 7.88 (d, J = 7.6 Hz, 1H), 7.99 (d, J = 7.6 Hz, 1H), 8.19 (d, J = 7.9 Hz, 2H). Anal. ($C_{21}H_{24}N_4O \cdot 2.5$TFA) C, H, N.

2-(4-Piperidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (11). A mixture of **4a** (0.905 g, 2.9 mmol) and PtO_2 (180 mg) in AcOH (20 mL) under hydrogen (60 psi) was stirred at ambient temperature for 4.5 h, filtered through a nylon membrane, and concentrated. The residue was purified by flash chromatography on silica gel using 10% MeOH/dichloromethane to provide the title compound (0.56 g, 55%). ^{1}H NMR (DMSO-d_6): δ 1.64 (m, 1H), 1.89 (m, 6H), 3.03 (td, J = 12.2, 4.2 Hz, 2H), 3.17 (s, 1H), 4.27 (dd, J = 11.7, 3.0 Hz, 1H), 7.36 (t, J = 7.6 Hz, 1H), 7.77 (m, 4H), 7.89 (d, J = 7.5 Hz, 1H), 8.33 (d, J = 8.1 Hz, 2H), 9.33 (s, 1H). MS (ESI): m/z 321 $(M + H)^+$.

2-(4-Piperidin-3-ylphenyl)-1*H*-benzimidazole-4-carboxamide (12). The title compound was prepared from **3** using the procedure described for **11** (0.96 g, 95%). ^{1}H NMR (DMSO-d_6): δ 1.52 (d, J = 12.2 Hz, 1H), 1.63 (dd, J = 12.2, 3.0 Hz, 1H), 1.68 (m, 1H), 1.91 (s, 1H), 2.57 (m, 2H), 2.67−2.75 (m, 1H), 2.97 (d, J = 12.2 Hz, 1H), 3.03 (d, J = 11.3 Hz, 1H), 7.32 (t, J = 7.8 Hz, 1H), 7.45 (d, J = 8.2 Hz, 2H), 7.73 (d, J = 8.2 Hz, 2H), 7.85 (d, J = 7.6 Hz 1H), 8.16 (d, J = 7.9 Hz, 1H), 9.33 (s, 1H). MS (ESI): m/z 321 $(M + H)^+$.

2-(4-(1-Methyl-piperidin-3-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (15). The title compound was prepared from **12** and formaldehyde using general procedure B (10 mg, 22%). ^{1}H NMR (DMSO-d_6): δ 1.43 (dd, J = 12.0, 4.0 Hz, 1H), 1.62 (m, 1H), 1.71 (m, 1H), 1.85 (s, 2H), 1.96 (m, 1H), 1.99 (m, 1H), 2.20 (s, 3H), 2.82 (m, 3H), 7.32 (t, J = 7.8 Hz, 1H), 7.48 (t, J = 7.3 Hz, 2H), 7.71 (d, J = 7.1 Hz, 2H), 7.85 (d, J = 7.4 Hz 1H), 8.15 (d, J = 8.0 Hz, 2H), 9.32 (s, 1H). Anal. ($C_{20}H_{22}N_4O \cdot$ 4.35TFA) C, H, N.

2-(4-(1-Methyl-piperidin-4-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (16). The title compound was prepared from **13** and formaldehyde using general procedure B (7 mg, 8%). MS (ESI): m/z 335 $(M + H)^+$. ^{1}H NMR (DMSO-d_6): δ 1.23 (s, 1H), 1.95 (m, 2H), 2.07 (m, 2H), 2.83 (s, 3H), 2.92 (m, 1H), 3.11 (m, 1H), 3.51 (s, 2H), 7.35 (t, J = 7.8 Hz, 1H), 7.48 (d, J = 7.9 Hz, 2H), 7.76 (m, 2H), 7.87 (d, J = 7.3 Hz, 1H), 8.22 (d, J = 8.2 Hz, 2H), 9.35 (d, J = 2.7 Hz, 1H).

2-(4-(1-Isopropyl-piperidin-2-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (17). The title compound was prepared using general procedure B, using acetone in place of formaldehyde (9 mg, 13%). ^{1}H NMR (DMSO-d_6): δ 0.76 (s, 2H), 0.95 (s, 2H), 1.11 (m, 1H), 1.26 (d, J = 6.1 Hz, 2H), 1.48 (s, 2H), 1.73 (s, 3H), 2.16 (s, 1H), 2.93 (s, 1H), 7.33 (m, 1H), 7.52 (s, 1H), 7.73 (s, 3H), 7.86 (d, J = 7.7 Hz 1H), 8.17 (m, 2H), 9.35 (s, 1H). MS (ESI): m/z 363 $(M + H)^+$.

2-(4-(1-Isopropyl-piperidin-3-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (18). The title compound was prepared from **12** and acetone using general procedure B (41 mg, 72%). ^{1}H NMR (DMSO-d_6): δ 1.27 (d, J = 4.6 Hz, 6H), 1.84 (m, 1H), 1.98 (m, 2H), 2.98 (m, 1H), 3.16 (d, J = 5.2 Hz, 2H), 3.43 (m, 4H), 7.34 (t, J = 7.8 Hz, 1H), 7.55 (d, J = 8.3 Hz, 2H), 7.73 (d, J = 7.7 Hz, 2H), 7.87 (d, J = 7.4 Hz, 1H), 8.21 (d, J = 8.3 Hz, 2H), 9.32 (s, 1H). MS (ESI): m/z 363 $(M + H)^+$.

2-(4-(1-Isopropyl-piperidin-4-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (19). The title compound was prepared from **13** and acetone using general procedure B (86 mg, 76%). ^{1}H NMR (DMSO-d_6): δ 1.30 (d, J = 6.7 Hz, 6H), 1.95 (m, 2H), 2.11 (d, J = 13.7 Hz, 2H), 8.97 (s, 1H), 3.00 (ddd, J = 12.1, 8.8, 3.8 Hz, 1H), 3.26 (s, 1H), 3.13 (m, 3H), 3.53 (d, J = 9.5 Hz, 1H), 7.36 (t, J = 7.8 Hz, 1H), 7.47 (d, J = 8.2 Hz, 2H), 7.77 (m, 2H), 7.87 (d, J = 7.6 Hz, 1H), 8.23 (d, J = 8.2 Hz, 2H). MS (DCI): m/z 363 $(M + H)^+$.

6-Fluoro-2-(4-pyrrolidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (20). The title compound was prepared as described for the synthesis of **21**, using 2,3-diamino-5-fluorobenzamide[12] in place of 2,3-diamino-5-chlorobenzamide (120 mg, 57%). ^{1}H NMR (DMSO-d_6): δ 2.12 (m, 3H), 2.47 (m, 1H), 3.41 (m, 2H), 4.69 (s, 1H), 7.62 (m, 2H), 7.72 (d, J = 8.2 Hz, 2H), 7.99 (s, 1H), 8.33 (d, J = 8.2 Hz, 2H), 8.96 (s, 1H), 9.21 (s, 1H), 9.77 (s, 1H). Anal. ($C_{18}H_{17}FN_4O \cdot 2.2$TFA) C, H, N.

6-Chloro-2-(4-pyrrolidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (21). Step 1. Preparation of *tert*-Butyl 2-(4-(4-Carbamoyl-6-chloro-1*H*-benzimidazol-2-yl)phenyl)pyrrolidine-1-carboxylate. To a solution of *tert*-butyl 2-(4-carboxyphenyl)-pyrrolidine-1-carboxylate (500 mg, 1.7 mmol) in dichloromethane (10 mL) were added oxalyl chloride (0.15 mL, 1.7 mmol) and DMF (1 drop), and the mixture was stirred at ambient temperature for 1 h. After concentration, the residue was dissolved in dichloromethane (20 mL), and the solution was added to a solution of 2,3-diamino-5-chlorobenzamide[12] (316 mg, 1.7 mmol) in THF (10 mL), followed by triethylamine (2 mL). The mixture was stirred at ambient temperature for 18 h and concentrated. The residue was dissolved in AcOH (10 mL), heated at 80 °C for 2 h, and concentrated. The residue was dissolved in EtOAc, washed with sodium bicarbonate solution and brine, and concentrated. Purification by flash chromatography on silica gel using EtOAc afforded the title compound (370 mg, 55%).

Step 2. Preparation of 6-Chloro-2-(4-pyrrolidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (21). To a solution of the product of step 1 (410 mg, 0.93 mmol) in dichloromethane (20 mL) was added TFA (4 mL), and the mixture was stirred at ambient temperature for 1 h. After concentration, the residue was purified by HPLC to provide the title compound (40 mg, 32%). ^{1}H NMR (DMSO-d_6): δ 2.12 (m, 3H), 2.46 (m, 2H), 3.39 (m, 2H), 4.68 (m, 1H), 7.72 (d, J = 8.3 Hz, 2H), 7.82 (m, 1H), 7.95 (s, 1H), 8.34 (d, J = 8.3 Hz, 2H), 8.94 (s, 1H), 9.14 (s, 1H), 9.73 (s, 1H). Anal. ($C_{18}H_{17}ClN_4O \cdot 2$TFA) C, H, N, Cl.

2-(4-(1-Ethylpyrrolidin-2-yl)-2-fluorophenyl)-1*H*-benzimidazole-4-carboxamide (22). Step 1. Preparation of *tert*-Butyl 2-(3-fluoro-4-methoxycarbonylphenyl)pyrrole-1-carboxylate. A mixture of methyl 4-bromo-2-fluorobenzoate (4 g, 17.2 mmol), 1-(Boc)pyrrole-2-boronic acid (5.44 g, 25.8 mmol), and dichlorobis(triphenylphosphine)palladium(II) (1.2 g, 1.72 mmol) in 7:3:2 DME/water/EtOH (300 mL) and 2 M aqueous sodium carbonate (17.2 mL) was stirred for 140 min at 80 °C. The mixture was cooled and concentrated, and the residue was dissolved in EtOAc. The solution was washed with brine and concentrated, and the residue was purified by flash chromatography on silica gel using 1:4 EtOAc/hexane to afford the title compound (5.51 g, 100%). ^{1}H NMR ($CDCl_3$): δ 1.41 (s, 9H),

3.94 (s, 3H), 6.22−6.31 (m, 2H), 7.10−7.22 (m, 2H), 7.38 (dd, J = 3.4, 1.7 Hz, 1H), 7.92 (t, J = 8.0 Hz, 1H). MS (DCI): m/z 320 $(M + H)^+$.

Step 2. Preparation of *tert*-Butyl 2-(3-fluoro-4-methoxycarbonylphenyl)pyrrolidine-1-carboxylate. A mixture of the product of step 1 (5.5 g, 17.2 mmol) and 5% Pt/C (20 mg) in acetic acid (200 mL) was hydrogenated at 60 psi for 12 h and filtered. The filtrate was concentrated, and the residue was partitioned between EtOAc and sodium bicarbonate solution. The organic phase was separated and concentrated, and the residue was purified by flash chromatography on silica gel using a gradient of 10−30% EtOAc/hexanes to give the title compound (5.5 g, 98%). ^{1}H NMR ($CDCl_3$): δ 1.21 (s, 9H), 1.46 (m, 1H), 1.75−1.96 (m, 2H), 2.35 (m, 1H), 3.63 (m, 2H), 3.92 (s, 3H), 4.73−4.82 (m, 1H), 6.95 (d, J = 11.9 Hz, 1H), 7.03 (d, J = 8.1 Hz, 1H), 7.88 (t, J = 7.8 Hz, 1H). MS (DCI): m/z 324 $(M + H)^+$.

Step 3. Preparation of *tert*-Butyl 2-(4-carboxy-3-fluorophenyl)pyrrolidine-1-carboxylate. A mixture of the product of step 2 (5.5 g, 17 mmol) and lithium hydroxide monohydrate (1.43 g, 34 mmol) in THF (50 mL) and water (50 mL) was titrated with MeOH until transparent. After it was stirred at ambient temperature for 2 h, the mixture was brought to pH 2 with 2 M HCl, concentrated to ∼40 mL, and filtered to provide the crude product (4.87 g, 92%). MS (DCI): m/z 310 $(M + H)^+$.

Step 4. Preparation of *tert*-Butyl 2-(4-(4-carbamoyl-1*H*-benzimidazol-2-yl)-3-fluorophenyl)pyrrolidine-1-carboxylate. To a solution of the product of step 3 (1.48 g, 4.8 mmol) in pyridine (5 mL) and DMF (5 mL) was added CDI (0.856 g, 5.28 mmol), and the mixture was stirred at 45 °C for 2 h. 2,3-Diaminobenzamide dihydrochloride[12] (1.08 g, 4.8 mmol) was added, and the mixture was stirred at ambient temperature for 18 h and concentrated. The residue was dissolved in AcOH (30 mL), heated at 80 °C for 3 h, and concentrated. The residue was dissolved in EtOAc, washed with sodium bicarbonate solution and brine, and concentrated. Purification by flash chromatography on silica gel using a gradient of 0−15% MeOH in 2:1 EtOAc/hexane provided the title compound (1.56 g, 77%). ^{1}H NMR ($CDCl_3$): δ 1.25 (s, 6H), 1.51 (s, 3H), 1.80−1.96 (m, 3H), 2.32−2.45 (m, 1H), 3.50 (s, 3H), 3.67 (m, 2H), 4.81−4.98 (m, 1H), 6.05 (s, 1H), 6.98−7.22 (m, 2H), 7.39 (s, 1H), 7.77 (s, 1H), 8.17 (s, 1H), 8.46 (d, J = 1.5 Hz, 1H). MS (APCI): m/z 425 $(M + H)^+$.

Step 5. Preparation of 2-(2-Fluoro-4-pyrrolidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (22). To a solution of the product of step 4 (1.5 g, 3.5 mmol) in dichloromethane (50 mL) was added TFA (10 mL), and the mixture was stirred at ambient temperature for 1 h. After it was concentrated, the residue was purified by HPLC to provide the title compound (1.42 g, 73%). ^{1}H NMR (CD_3OD): δ 2.28 (m, 3H), 2.58 (m, 1H), 3.53 (m, 2H), 4.76 (dd, J = 9.5, 7.1 Hz, 1H), 7.43 (t, J = 7.8 Hz, 1H), 7.50−7.55 (m, 2H), 7.84 (d, J = 7.4 Hz, 1H), 7.98 (d, J = 7.7 Hz, 1H), 8.42 (t, J = 8.1 Hz, 1H). MS (APCI): m/z 325 $(M + H)^+$.

(*R*)-2-(2-Fluoro-4-(pyrrolidin-2-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (22a). Step 1. Preparation of (*S*)-*tert*-Butyl 2-(3-fluoro-4-(methoxycarbonyl)phenyl)pyrrolidine-1-carboxylate. The racemic product of **22**, step 3 (6.3 g), was resolved by chiral HPLC (Whelk O, 95:2.5:2.5 hexane/EtOH/MeOH) to give the 2.6 g of the title compound as the faster-eluting fraction (100% e.e., *S*-enantiomer), MS (DCI): m/z 324 $(M + 1)^+$, and 2.7 g of a slower-eluting fraction (97.5% e.e., *R*-enantiomer), MS (DCI): m/z 324 $(M + 1)^+$.

Step 2. Preparation of (*R*)-4-(1-(Boc)pyrrolidin-2-yl)-2-fluorobenzoic Acid. To a solution of the slower-eluting fraction (*R*-enantiomer) of step 1 (2.65 g, 8.2 mmol) in THF (20 mL) was added a solution of lithium hydroxide monohydrate (688 mg, 16.4 mmol) in 20 mL of water. MeOH (10 mL) was added until a transparent solution formed, and the solution was stirred at room temperature for 2 h. The mixture was brought to pH 2 with 2 N HCl and concentrated to ∼10 mL, diluted with water, and allowed to stand at room temperature for 16 h. The white solid was collected by filtration, washed with water, and dried to give the crude title compound (2.21 g, 87%). Recrystallization from MeOH and water gave the title compound (1.61 g, 63%). MS (DCI): m/z 310 $(M + H)^+$.

Step 3. Preparation of (*R*)-2-(2-Fluoro-4-(pyrrolidin-2-yl)-phenyl)-1*H*-benzimidazole-4-carboxamide (22a). The title compound was prepared using the product of step 2 according to the procedure for **22**, step 6. To a solution of the TFA salt in MeOH and dichloromethane was added 1 M HCl in ether. Concentration afforded the title compound as the HCl salt. $[\alpha]^{589}$ = +7.3 (c = 0.6 in MeOH). ^{1}H NMR (CD_3OD): δ 2.29 (m, 3H), 2.63 (m, 1H), 3.54 (m, 2H), 4.83 (m, 1H), 7.73 (m, 3H), 8.05 (d, J = 8.2 Hz, 1H), 8.12 (d, J = 7.6 Hz, 1H), 8.26 (t, J = 7.8 Hz, 1H). MS (DCI): m/z 325 $(M + H)^+$. Anal. ($C_{18}H_{17}FN_4O \cdot 2.6HCl$) C, H, N; calcd, 13.37; found, 12.96.

(*S*)-2-(2-Fluoro-4-(pyrrolidin-2-yl)phenyl)-1*H*-benzimidazole-4-carboxamide (22b). The title compound was prepared as described for **22a**, using the faster-eluting fraction (*S*-enantiomer) from step 1. $[\alpha]^{589}$ = −6.8 (c = 0.7 in MeOH). ^{1}H NMR (CD_3OD): δ 2.30 (m, 3H), 2.62 (m, 1H), 3.55 (m, 2H), 4.83 (m, 1H), 7.74 (m, 3H), 8.05 (d, J = 7.6 Hz, 1H), 8.12 (d, J = 7.6 Hz, 1H), 8.26 (t, J = 7.8 Hz, 1H). MS (DCI): m/z 325 $(M + H)^+$. Anal. ($C_{18}H_{17}FN_4O \cdot 2.3HCl$) C, H, N.

6-Fluoro-2-(2-fluoro-4-pyrrolidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (23). Step 1. Preparation of *tert*-Butyl 2-(4-(4-carbamoyl-6-fluoro-1*H*-benzimidazol-2-yl)-3-fluorophenyl)pyrrolidine-1-carboxylate. To a solution of **22**, step 4 (700 mg, 2.26 mmol), in dichloromethane (8 mL) were added oxalyl chloride (296 μL, 3.5 mmol) and DMF (1 drop), and the mixture was stirred at ambient temperature for 1 h. The mixture was concentrated, and the residue was dissolved in dichloromethane (8 mL). This solution was added to a solution of 2,3-diamino-5-fluorobenzamide[12] (382 mg, 2.26 mmol) and triethylamine (378 μL, 2.71 mmol) in THF (8 mL), and the mixture was stirred at ambient temperature for 18 h. The mixture was concentrated, and the residue was dissolved in AcOH (15 mL), heated at 80 °C for 3 h, and concentrated. The residue was partitioned between EtOAc and sodium bicarbonate solution, and the organic layer was washed with sodium bicarbonate solution and concentrated. The residue was purified by flash chromatography on silica gel using 3:2 EtOAc/hexanes to afford the title compound (367 mg, 37%). ^{1}H NMR (CD_3OD): δ 1.23 (s, 9H), 1.86−1.97 (m, 3H), 2.41−2.48 (m, 1H), 3.57−3.69 (m, 2H), 4.90−4.98 (m, 1H), 7.15−7.27 (m, 2H), 7.49 (s, 1H), 7.70 (d, J = 10.7 Hz, 1H), 8.32 (s, 1H). MS (APCI): m/z 443 $(M + H)^+$.

Step 2. Preparation of 6-Fluoro-2-(2-fluoro-4-pyrrolidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (23). A solution of the product of step 1 (360 mg, 0.81 mmol) in dichloromethane (25 mL) and TFA (5 mL) was stirred at ambient temperature for 1 h and concentrated. The residue was purified by HPLC to provide the title compound (326 mg, 70%). ^{1}H NMR (CD_3OD): δ 2.20−2.37 (m, 3H), 2.56−2.62 (m, 1H), 3.47−3.57 (m, 2H), 4.76 (dd, J = 9.3, 7.2 Hz, 1H), 7.50−7.54 (m, 3 H), 7.71 (dd, J = 10.4, 2.4 Hz, 1H), 8.43 (t, J = 7.9 Hz, 1 H). Anal. ($C_{18}H_{16}F_2N_4O \cdot 2.1TFA$) C, H, N.

6-Fluoro-2-(4-piperidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (24). The title compound was prepared from *tert*-butyl 2-(4-carboxyphenyl)piperidine-1-carboxylate and 2,3-diamino-5-fluorobenzamide[12] using general procedure A (89 mg, 54%). ^{1}H NMR (DMSO-d_6): δ 1.68 (s, 1H), 1.83 (d, J = 2.5 Hz, 2H), 1.91 (m, 2H), 1.98 (s, 1H), 3.07 (s, 1H), 3.38 (s, 2H), 4.33 (s, 1H), 7.61 (m, 2H), 7.76 (d, J = 8.3 Hz, 2H), 7.94 (s, 1H), 8.32 (d, J = 8.6 Hz, 2H), 9.21 (s, 1H). MS (ESI): m/z 339 $(M + H)^+$.

2-(2-Fluoro-4-piperidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (25). Step 1. Preparation of 2-(4-Bromo-2-fluorophenyl)-1*H*-benzimidazole-4-carboxamide. The title compound was prepared from 4-bromo-2-fluorobenzoic acid using general procedure A (91%). ^{1}H NMR (DMSO-d_6): δ 7.42 (t, J = 7.5 Hz, 1H), 7.66 (d, J = 8.2 Hz, 1H), 7.85 (m, 3H), 7.96 (d, J = 7.3 Hz, 1H), 8.29 (t, J = 7.9 Hz, 1H), 9.29 (s, 1H), 13.15 (s, 1H).

Step 2. Preparation of 2-(2-Fluoro-4-pyridin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide. To the product of step 1 (200 mg,0.6 mmol), $Pd_2(dba)_3$ (55 mg, 0.06 mmol), and tri-*o*-tolylphosphine (55 mg, 0.028 mmol) were added DMF (10 mL), 2-(tri-*n*-butylstannyl)pyridine (220 mg, 0.6 mmol), and triethylamine (238 μL, 1.7 mmol). The mixture was purged with nitrogen, heated at 75 °C for 18 h, cooled, and concentrated. The residue was purified by flash chromatography on silica gel using 5% MeOH/20% EtOAc/75% hexanes, followed by recrystallization from MeOH to give the title compound (110 mg, 55%). ^{1}H NMR (DMSO-d_6): δ 7.40 (m, 2H), 7.51 (m, 2H), 7.78 (m, 1H), 7.93 (t, J = 5.8 Hz, 1H), 8.00 (m, 1H), 8.20 (m, 3H), 8.43 (t, J = 8.1 Hz, 1H), 8.76 (d, J = 3.7 Hz, 1H), 9.15 (s, 1H).

Step 3. Preparation of 2-(2-Fluoro-4-piperidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (25). The product of step 2 (80 mg, 0.25 mmol) and catalytic 5% Pt/C (53 mg, 0.025 mmol) in MeOH (10 mL) were hydrogenated under 60 psi of hydrogen until starting material was consumed. After filtration, the filtrate was concentrated, and the residue was purified by HPLC to afford the title compound (95%). ^{1}H NMR (CD_3OD): δ 1.82 (m, 2H), 2.03 (m, 3H), 2.18 (d, J = 15.9 Hz, 1H), 3.24 (t, J = 10.0, 1H), 3.54 (d, J = 12.8 Hz, 1H), 4.39 (m, 1H), 7.44 (t, J = 7.9 Hz, 1H), 7.53 (d, J = 10.0, 2H), 7.85 (d, J = 7.9 Hz, 1H), 7.99 (d, J = 7.6 Hz, 1H), 8.44 (t, J = 7.8 Hz, 1H). MS (DCI): m/z 339 $(M + H)^+$.

(*R*)-2-(2-Fluoro-4-piperidin-2-yl-phenyl)-1*H*-benzimidazole-4-carboxamide (25a). Step 1. Preparation of Methyl 2-Fluoro-4-pyridin-2-yl-benzoate. To a mixture of methyl 4-bromo-2-fluorobenzoate (5.0 g, 21.5 mmol), $Pd_2(dba)_3$ (1.5 g, 1.6 mmol), and tri-2-furylphosphine (1.5 g, 6.4 mmol) in DMF (100 mL) were added 2-trimethylstannyl pyridine (9.5 g, 25.75 mmol) and triethylamine (2 mL) under nitrogen, and the mixture was stirred at 80 °C for 10 h. After it was cooled, the mixture was partitioned between EtOAc and brine, and the organic phase was concentrated. The residue was purified by flash chromatography using 1/5 EtOAc/hexane to give the title compound (2.6 g, 52%). ^{1}H NMR (CD_3OD): δ 3.93 (s, 3H), 7.40 (m, 1H), 7.88 (m, 4H), 8.05 (m, 1H), 8.65 (m, 1H). MS (DCI): m/z 232 $(M + H)^+$.

Step 2. Preparation of Benzyl 2-(3-Fluoro-4-(methoxycarbonyl)-phenyl)piperidine-1-carboxylate. The product of step 1 (7.0 g, 30 mmol) was stirred under 60 psi of hydrogen with 5% Pt/C (350 mg, 0.16 mmol) in 100 mL of MeOH for 10 h to give 7.0 g of crude product. This was dissolved in dioxane (100 mL) and water (50 mL) and treated with potassium carbonate (5 g) and benzyl chlorofomate (5.2 mL, 35 mmol). After the mixture was stirred at ambient temperature for 4 h, piperazine (86 mg, 1 mmol) was added, and the mixture was stirred for 30 min. The mixture was concentrated, and the residue was partitioned between EtOAc and dilute HCl. The organic phase was washed with water and concentrated, and the residue was purified by flash chromatography using 1/4 EtOAc/hexane to give the title compound (7.5 g, 93%). ^{1}H NMR (CD_3OD): δ 1.39 (m, 1H), 1.66 (m, 3H), 1.94 (m, 1H), 2.32 (d, J = 2.7 Hz, 1H), 2.87 (m, 1H), 3.90 (s, 3H), 4.15 (m, 1H), 5.17 (d, J = 5.1 Hz, 2H), 5.45 (d, J = 3.7 Hz, 1H), 7.04 (d, J = 12.2 Hz, 1H), 7.12 (d, J = 8.5 Hz, 1H), 7.32 (m, 5H), 7.89 (t, J = 7.8 Hz, 1H). MS (DCI): m/z 372 $(M + H)^+$.

Step 3. Preparation of Benzyl (*R*)-3-(4-Methoxycarbonyl-3-fluorophenyl)piperidine-1-carboxylate. The product of step 2 (1 g, 3.7 mmol) was resolved by chiral HPLC (Chiralcel OJ, 85:7.5:7.5 hexane/EtOH/MeOH) to afford a faster-eluting fraction (448 mg, 48%; 100% e.e., *R*-enantiomer) and a slower-eluting fraction (460 mg, 46%; 98% e.e., *S*-enantiomer). ^{1}H NMR (CD_3OD): δ 1.38 (m, 1H), 1.55 (m, 3H), 1.92 (m, 1H), 2.32 (d, J = 2.7 Hz, 1H), 2.80 (m, 1H), 3.90 (s, 3 H), 4.10 (m, 1H), 5.17 (d, J = 5.1 Hz, 2H), 5.45 (d, J = 3.7 Hz, 1H), 7.04 (d, J = 12.2 Hz, 1H), 7.12 (d, J = 8.5 Hz, 1H), 7.31 (m, 5H), 7.89 (t, J = 7.8 Hz, 1H). MS (DCI): m/z 372 $(M + H)^+$.

Step 4: Preparation of Benzyl (*R*)-2-(4-Carboxy-3-fluorophenyl)piperidine-1-carboxylate. The title compound was prepared according to the procedure for **22**, step 4, using the fast-eluting fraction (*R*-enantiomer) of step 3 (390 mg, 88%). ^{1}H NMR (CD_3OD): δ 1.63 (m, 1H), 1.78 (m, 3H), 2.02 (m, 1H), 2.76 (m, 1H), 2.93 (s, 1H), 4.17 (dd, J = 12.9, 4.1 Hz, 2H), 5.14 (s, 2H), 7.14 (s, 1H), 7.32 (d, J = 3.7 Hz, 1H), 7.36 (m, 5H), 7.87 (t, J = 7.8 Hz, 1H). MS (DCI): m/z 358 $(M + H)^+$.

Step 5. Preparation of Benzyl (*R*)-2-[4-(2-Amino-3-carbamoyl-phenylcarbamoyl)-3-fluorophenyl]piperidine-1-carboxylate. The title compound was prepared from the product of step 4 using general procedure A (440 mg, 85%). MS (DCI/NH_3): m/z 473 $(M + H)^+$.

Step 6. Preparation of (*R*)-2-(2-Fluoro-4-piperidin-2-yl-phenyl)-1*H*-benzimidazole-4-carboxamide (25a). The title compound was prepared as described for the synthesis of **37**, step 3, using the product of step 5 (305 mg, 73%). $[\alpha]^{589}$ = +3.0 (c = 1.0 in MeOH). ^{1}H NMR (CD_3OD): δ 1.87 (m, 2H), 2.05 (m, 3H), 2.20 (m, 1H), 3.25 (m, 1H), 3.56 (d, J = 12.9 Hz, 1H), 4.49 (d, J = 11.7 Hz, 1H), 7.69 (m, 3H), 8.02 (d, J = 8.3 Hz, 1H), 8.11 (d, J = 7.7 Hz, 1H), 8.30 (t, J = 7.8 Hz, 1H). MS (DCI): m/z 373 $(M + H)^+$. Anal. ($C_{19}H_{19}FN_4O \cdot 3HCl$) C, N, H; calcd, 4.95; found, 5.39.

(*S*)-2-(2-Fluoro-4-piperidin-2-yl-phenyl)-1*H*-benzimidazole-4-carboxamide (25b). The title compound was prepared as described for **25a**, using the slower-eluting fraction (*S*-enantiomer) produced in **25a**, step 3. $[\alpha]^{589}$ = −3.03 (c = 1.0 in MeOH). ^{1}H NMR (CD_3OD): δ 1.84 (m, 2H), 2.10 (m, 4H), 3.22 (m, 1H), 3.56 (d, J = 12.9 Hz, 1H), 4.49 (d, J = 11.7 Hz, 1H), 7.70 (m, 3H), 8.02 (d, J = 8.3 Hz, 1H), 8.11 (d, J = 7.7 Hz, 1H), 8.30 (t, J = 7.8 Hz, 1H). Anal. ($C_{19}H_{19}FN_4O \cdot 3HCl$) C, H, N.

6-Fluoro-2-(2-fluoro-4-piperidin-2-ylphenyl)-1*H*-benzimidazole-4-carboxamide (26). The title compound was prepared as described for the synthesis of **25**, using 2,3-diamino-5-fluorobenzamide[12] in place of 2,3-diaminobenzamide (400 mg, 80%). ^{1}H NMR (CD_3OD): δ 1.83 (m, 2H), 2.04 (m, 3H), 2.18 (m, 1H), 3.21−3.26 (m, 1H), 3.54 (d, J = 12.5 Hz, 1H), 4.39 (dd, J = 12.2, 2.4 Hz, 1H), 7.51 (m, 3H), 7.70 (dd, J = 10.4, 2.4 Hz, 1H), 8.43 (t, J = 7.8 Hz, 1H). MS (DCI): m/z 357 $(M + H)^+$.

Supporting Information Available: PARP-2 data for select compounds and microanalytical data for compounds **4a**,**b**, **6**, **10**, **13**−**15**, **20**, **21**, **22a**,**b**, **23**, and **25a**,**b**. This material is available free of charge via the Internet at http://pubs.acs.org.

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