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Synthesis and Structure−Activity Studies of Biphenyl Analogues of the Tuberculosis Drug (6*S*)-2-Nitro-6-{[4-(trifluoromethoxy)benzyl]oxy}-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (PA-824)

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A series of biphenyl analogues of the new tuberculosis drug PA-824 was prepared, primarily by coupling the known (6*S*)-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazin-6-ol with iodobenzyl halides, followed by Suzuki coupling of these iodides with appropriate arylboronic acids or by assembly of the complete biaryl side chain prior to coupling with the above alcohol. Antitubercular activity was determined under both replicating (MABA) and nonreplicating (LORA) conditions. *para*-Linked biaryls were the most active, followed by *meta*-linked and then *ortho*-linked analogues. A more detailed study of a larger group of *para*-linked analogues showed a significant correlation between potency (MABA) and both lipophilicity (CLOGP) and the electron-withdrawing properties of terminal ring substituents ($\sum\sigma$). Selected compounds were evaluated for their efficacy in a mouse model of acute *Mycobacterium tuberculosis* infection. *In vivo* activity correlated well with the stability of compounds to microsomal metabolism. Three compounds bearing combinations of lipophilic, electron-withdrawing groups achieved >200-fold higher efficacies than the parent drug.

Introduction

Tuberculosis (TB),[a] caused by *Mycobacterium tuberculosis* (*M. tb*), is of increasing concern due to the recent emergence and spread of multidrug-resistant strains.[1] While the majority of cases are potentially curable, complex and lengthy multidrug combination treatment is required, due largely to the presence of metabolically quiescent subpopulations of bacteria which can survive long periods of treatment with the conventional drugs. The current standard protocols employ combinations of isoniazid, rifampin, pyrazinamide, and ethambutol, given over a period of 6−9 months. This is a major barrier to full patient compliance and has contributed to the development of drug-resistant strains.[2]

Recently, there has been an intensive effort seeking drugs that hit new targets, or work in new ways, since these have the potential to overcome multidrug resistance. An exciting development has been the discovery of the 2-nitroimidazo-[2,1-*b*][1,3]oxazines, exemplified by **1** (PA-824),[3] and the related 6-nitroimidazo[2,1-*b*][1,3]oxazoles, exemplified by **2** (OPC-67683).[4,5] These drugs show activity against both active and persistent *M. tb* in animal models, suggesting a novel mechanism and the potential to shorten the long treatment cycles that are needed with the current agents.[6,7]

A study of **1** in susceptible and resistant strains of *M. tb* showed[3] that it was converted to more polar metabolites only by the former strains, suggesting a bioreductive activation step (it has a low one-electron reduction potential of −534 mV).[8] Genetic studies suggested the involvement of a protein similar to the F420-dependent glucose-6-phosphate dehydrogenase of *Mycobacterium smegmatis*.[3] A later paper[9] confirmed that while resistance to **1** is caused by loss of either a bacterial glucose-6-phosphate dehydrogenase (FGD1) or its deazaflavin cofactor (F420), some mutants wild type for both FGD1 and F420 were still resistant to **1** but not to the related compound **3** (CGI-17341). Sequencing of these mutants showed changes in an unknown gene product (Rv3547, a putative nitroreductase) and that complementation with this restored sensitivity to **1**. Both **1** and **2** are currently in phase II clinical trials for the treatment of drug-susceptible and drug-resistant TB,[10,11] following successful phase II early bactericidal activity (EBA) studies.[6,10] A recent mouse study showed that the combination of **1** at 100 mg/kg with pyrazinamide was as effective as the standard first-line combination of rifampin, isoniazid, and pyrazinamide, although the emergence of strains resistant to **1** was observed.[12]

To date, structure−activity (SAR) studies in the 2-nitroimidazo[2,1-*b*][1,3]oxazine series have been reasonably limited. Early work with **1** and analogues showed that there was a considerable difference between the enantiomers at C-6, with the *S* enantiomer being much more active than the *R*.[3,13]

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[a] Abbreviations: TB, tuberculosis; *M. tb*, *Mycobacterium tuberculosis*; FGD1, F420-dependent glucose-6-phosphate dehydrogenase; EBA, early bactericidal activity; SAR, structure−activity relationship; MIC, minimum inhibitory concentration; SD, standard deviation; HREIMS, high-resolution electron impact mass spectrometry; HRFABMS, high-resolution fast atom bombardment mass spectrometry; HRAPCIMS, high-resolution atmospheric pressure chemical ionization mass spectrometry; DMF, *N*,*N*-dimethylformamide; DMSO, dimethyl sulfoxide; THF, tetrahydrofuran; TBAF, tetra-*n*-butylammonium fluoride; DMA, *N*,*N*-dimethylacetamide; THP, tetrahydropyranyl; SRM, selected reaction monitoring; IS, internal standard; PAR, peak area ratio; CFU, colony forming unit; CMC, carboxymethylcellulose; TBDMS, *tert*-butyldimethylsilyl.

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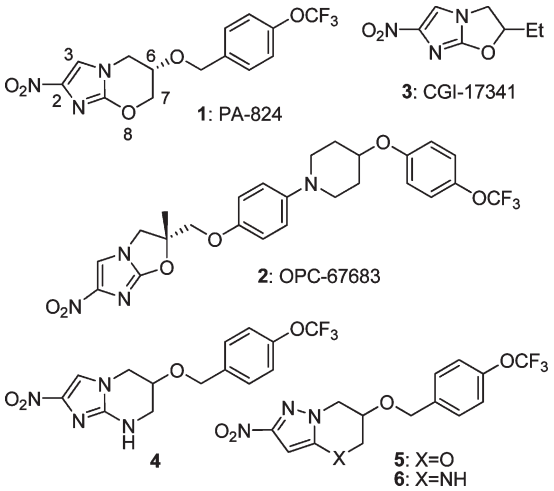
Some data have been reported for a small number of alternate linker analogues of **1**, with urea, carbamate, and amide variations all proving active in cultures of *Mycobacterium bovis*.[13] The synthesis and antitubercular activity of 7-methyl derivatives of **1** have also been described.[14] A recent broader study of the key determinants of aerobic activity in the general class of 2-nitroimidazo[2,1-*b*][1,3]oxazines showed that the required components were the bicyclic oxazine, the lipophilic tail, and an oxygen substituent in the 8-position.[15] Extending the linker between the 6-(*S*) position and the terminal aromatic ring also gave highly potent compounds for a more soluble 6-amino series.[16]

There are conflicting reports about the effects of other substituents at the 8-position. Two recent studies[8,16] have shown that replacing the 8-oxygen with sulfur (as thioether) is acceptable but that replacement with oxidized versions of the latter (sulfoxide, sulfone) or with carbon dramatically reduces activity. However, one report suggests that replacement of the 8-oxygen with nitrogen is also acceptable,[16] while in a study of heterocyclic analogues of **1** having similar one-electron reduction potentials we found the 2-nitroimidazo-[1,2-*a*]pyrimidine **4** (−568 mV), the 2-nitropyrazolo[5,1-*b*]-[1,3]oxazine **5** (−500 mV), and the 2-nitropyrazolo[1,5-*a*]pyrimidine **6** (−517 mV) to all be inactive.[8] A recent radiochemical study[17] has shown that the reduction chemistry of **1** is different to that of related nitroheterocycles, with imidazole ring reduction occurring in preference to nitro group reduction at the two-electron level. This is consistent with recent work[18] suggesting that **1** kills anaerobic bacteria through the release of reactive nitrogen species, including nitric oxide, with the level of formation (percent) of the concomitant denitro metabolite correlating with the amount of anaerobic killing of *M. tb* across a series of eight analogues.



Regarding the benzyl ether side chain of **1**, early studies had shown the utility of various lipophilic substituents, particularly at the 4-position on the benzyl ring (e.g., 4-*n*Bu, 4-*t*Bu, 4-CF_3, 4-OBn, 4-OCF_3, 4-OPh), to enhance potency against cultures of *M. bovis*.[13] To explain these and similar findings in the related 6-amino series, a recently described QSAR model predicted the presence of either two hydrophobic binding areas (one relatively close and one more distant from the imidazooxazine) or a single (proximal or more distant) hydrophobic pocket in the nitroreductase.[16] However, it is important to note that while a few analogues from some of the above studies display better potency than **1** *in vitro*, to date none of these have been shown to possess better efficacy than **1** *in vivo*.

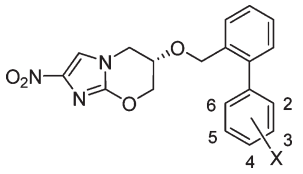
In considering the above SAR, we elected to retain the key imidazooxazine chromophore and probe the suggested hydrophobic pocket(s) by initially employing more conformationally rigid biphenyl-type side chains. The desired final goal of our studies was to develop a second generation analogue of **1** having an improved pharmacological profile over both **1** and **2** for potential advancement into clinical development for TB therapy. While **1** itself shows excellent pharmacokinetics, tolerability, and safety in current clinical trials,[19] the initial challenge for a second generation analogue, addressed in this study, was to first improve potency against both replicating and nonreplicating *M. tb* and to second demonstrate improved efficacy *in vivo*, compared to **1**. A more efficacious compound may help to reduce the cost of therapy and improve the therapeutic index, thereby delivering a safer, more affordable drug.

In this paper we therefore present SAR for biphenyl analogues of the nitroimidazooxazine **1**, with the aim of using this scaffold both to further delineate the nature of the preferred pharmacophore and to provide compounds of superior efficacy to **1**.

Chemistry and Methods

The bulk of the compounds listed in Tables 1, 2, and 3 were prepared from the known[13] chiral alcohol **122** by NaH-induced alkylations with iodobenzyl halides **123**, **125**, and **127** to give the iodobenzyl ethers **124**, **126**, and **128**[13] which then underwent Suzuki couplings with appropriate arylboronic acids (Scheme 1). Compounds **36**, **92**, **109**, **114**, and **120** were synthesized via the cyclic boronate esters **133** and **134**, which were conveniently prepared from the related bromobenzyl ethers **130** and **132** (Scheme 2). An alternative method, involving assembly of the biaryl side chain prior to coupling with alcohol **122**, was employed for compounds **98**, **100**, **106**, **107**, **111**, and **113** (Scheme 3). Novel iodobenzenes **137** and **138** were prepared from the commercial anilines **135** and **136**

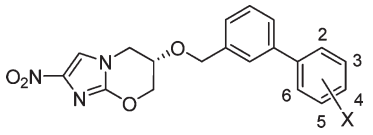
Table 1. MIC Data for *ortho*-Linked Biphenyl Analogues of **1**



compd	X	MIC (μM)[a] MABA	LORA
1		0.50 ± 0.30	2.6 ± 1.4
7	H	0.64 ± 0.32	2.5 ± 0.5
8	2-OCF_3	1.2 ± 0.4	6.2 ± 0.3
9	3-CN	0.80 ± 0.31	8.2 ± 2.9
10	3-F	0.62 ± 0.17	3.0 ± 0.6
11	3-OCF_3	1.9 ± 0.7	6.3 ± 0.7
12	3-SMe	0.81 ± 0.07	3.2 ± 0.3
13	4-COMe	17 ± 1	36 ± 13
14	4-CN	8.4 ± 1.4	19 ± 5
15	4-F	1.2 ± 0.3	3.4 ± 0.2
16	4-OCF_3	2.3 ± 0.4	4.3 ± 0.7
17	4-SMe	1.3 ± 0.4	3.4 ± 0.2
18	3-F, 4-OMe	1.8 ± 0.3	3.8 ± 0.2
19	3,4-benzo	1.6 ± 0.4	3.1 ± 0.8

[a] Minimum inhibitory concentration, determined under aerobic (MABA)[23] or anaerobic (LORA)[22] conditions. Each value is the mean of at least two independent determinations (29 determinations for **1**) ± SD.

Table 2. MIC Data for *meta*-Linked Biphenyl Analogues of **1**



		MIC (μM)[a]	
compd	X	MABA	LORA
1		0.50 ± 0.30	2.6 ± 1.4
20	H	0.095 ± 0.015	2.1 ± 0.8
21	2-OCF_3	0.46 ± 0.15	3.3 ± 0.1
22	3-CF_3	0.34 ± 0.10	2.9 ± 1.0
23	3-CN	0.17 ± 0.01	3.7 ± 2.1
24	3-F	0.18 ± 0.03	2.7 ± 0.8
25	3-OH	0.30 ± 0.16	3.7 ± 0.2
26	3-OCF_3	0.40 ± 0.07	3.5 ± 0.1
27	3-$O(CH_2)_3OH$	0.58 ± 0.33	2.5 ± 0.5
28	3-$O(CH_2)_3$Nmorph[b]	1.4 ± 0.4	11 ± 7
29	3-SMe	0.070 ± 0.037	1.4 ± 0.1
30	4-COMe	0.30 ± 0.19	1.9 ± 0.4
31	4-CF_3	0.12 ± 0.01	2.9 ± 1.4
32	4-CN	0.23 ± 0.01	4.8 ± 1.1
33	4-F	0.077 ± 0.034	3.8 ± 0.7
34	4-OH	0.54 ± 0.23	5.8 ± 0.1
35	4-OCF_3	0.11 ± 0.05	2.2 ± 0.5
36	4-OCF_2H	0.06 ± 0.01	2.8 ± 1.0
37	4-$O(CH_2)_3OH$	0.36 ± 0.12	1.5 ± 0.2
38	4-$O(CH_2)_3$Nmorph[b]	0.69 ± 0.25	11 ± 5
39	4-SMe	0.095 ± 0.025	0.93 ± 0.08
40	3-F, 4-OMe	0.19 ± 0.10	2.2 ± 0.5
41	3,4-benzo	0.12 ± 0.01	3.2 ± 0.4

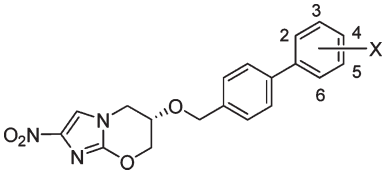
[a] As for Table 1. [b] *N*-Morpholinyl.

via diazotization. Suzuki coupling of these and other commercial halobenzenes (**139**−**141**) to 4-(hydroxymethyl)-phenylboronic acid (**142**) gave the required hydroxymethylbiphenyls (**143**−**147**), which were quantitatively converted to the bromides (**148**−**152**) by HBr/AcOH and coupled to **122** as above. This method was preferred for the larger scale synthesis of compounds when the required substituted phenylboronic acids were not commercially available. Compound **113** was similarly prepared via difluoromethylation[20] of the hydroxybiphenyl ester **154** (obtained via Suzuki coupling of 5-bromo-2-chlorophenol and 4-(methoxycarbonyl)phenylboronic acid **153**), followed by reduction to alcohol **156**, bromination with phosphorus tribromide, and coupling to **122**. Finally, to evaluate the acceptability of increasing aqueous solubility via alcohol and amino functionality linked to the terminal ring, several alkoxy-linked amino analogues (**28**, **38**, **66**, and **91**) were prepared from the corresponding phenols (**25**, **34**, **62**, and **85**, respectively) by reaction with protected hydroxyalkyl bromides, followed by elaboration of the deprotected alkoxy alcohols (**27**, **37**, **64**, and **90**) via their iodo derivatives (Scheme 4).

The comparative lipophilicities of the compounds in Table 3 were estimated both by summing substituent π values and also by log *P* values calculated using ACD LogP/LogD prediction software (v.8.0, Advanced Chemistry Development Inc., Toronto, Canada). In calculating the electronic properties of multisubstituted compounds, 2′- and 4′-substituents were assigned σ_p values, and 3′- and 5′-substituents were assigned σ_m values, and the total was summed.

The *in vitro* activity of the compounds against *M. tb* (strain H37Rv) was quantified by MICs (minimum inhibitory concentrations; the lowest compound concentration effecting a growth inhibition of >90%). The values recorded represent

Table 3. MIC Data, Calculated log *P*, and Substituent Values for *para*-Linked Biphenyl Analogues of **1**



					MIC (μM)[d]	
compd	X	$\sum\pi$[a]	log *P*[b]	$\sum\sigma$[c]	MABA	LORA
1			2.70		0.50 ± 0.30	2.6 ± 1.4
42	H	0	3.51	0	0.045 ± 0.005	3.9 ± 2.0
43	2-CF_3	0.88	4.48	0.54	0.077 ± 0.031	1.6 ± 0.4
44	2-CHO	−0.65	2.63	0.42	0.08 ± 0.02	0.78 ± 0.16
45	2-F	0.14	4.02	0.06	0.19 ± 0.01	0.67 ± 0.12
46	2-Cl	0.71	3.99	0.23	0.15 ± 0.04	1.5 ± 0.3
47	2-OH	−0.67	2.47	−0.37	0.12 ± 0	4.6 ± 1.1
48	2-OMe	−0.02	3.17	−0.27	0.065 ± 0.005	1.4 ± 0.3
49	2-OEt	0.38	3.71	−0.24	0.03 ± 0	0.82 ± 0.19
50	2-$O(CH_2)_3OH$	−1.0	2.98	−0.20	0.34 ± 0.11	2.7 ± 0.9
51	2-OCF_3	1.04	4.21	0.35	0.035 ± 0.005	0.97 ± 0.08
52	2-OPh	2.08	5.50	−0.03	0.06 ± 0	1.8 ± 0.1
53	2-SMe	0.61	4.20	0.0	0.087 ± 0.025	1.4 ± 0.3
54	3-*i*Pr	1.53	4.85	−0.07	0.14 ± 0.02	4.2 ± 1.5
55	3-Ph	1.96	5.55	0.06	0.31 ± 0.09	0.88 ± 0.02
56	3-CF_3	0.88	4.48	0.43	0.067 ± 0.025	1.3 ± 0.4
57	3-CHO	−0.65	2.93	0.35	0.14 ± 0.02	0.57 ± 0.01
58	3-CN	−0.57	3.58	0.56	0.12 ± 0.01	1.3 ± 0.6
59	3-$CONH_2$	−1.49	2.03	0.28	2.8 ± 0.9	6.7 ± 1.9
60	3-F	0.14	3.53	0.34	0.045 ± 0.005	2.2 ± 0.7
61	3-Cl	0.71	4.08	0.37	0.06 ± 0.01	1.6 ± 0.5
62	3-OH	−0.67	2.76	0.12	0.14 ± 0.02	3.1 ± 1.4

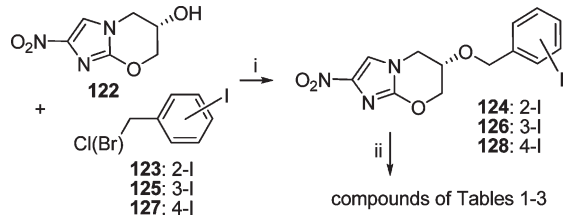
Table 3. Continued

compd	X	$\sum\pi^a$	log P^b	$\sum\sigma^c$	MIC (μM)[d] MABA	MIC (μM)[d] LORA
63	3-OMe	−0.02	3.27	0.12	0.27 ± 0.17	1.9 ± 0
64	3-O$(CH_2)_2$OH	−1.47	2.30	−0.02	0.46 ± 0.01	5.4 ± 2.3
65	3-O$(CH_2)_3$OH	−1.0	3.08	−0.12	0.18 ± 0.07	3.2 ± 1.2
66	3-O$(CH_2)_2NMe_2$		3.13	−0.12	1.5 ± 0.5	18 ± 8
67	3-OCF_3	1.04	4.31	0.38	0.077 ± 0.026	1.4 ± 0.6
68	3-OCH_2Ph	1.66	4.93	0.10	0.12 ± 0.06	0.88 ± 0
69	3-SMe	0.61	4.00	0.15	0.077 ± 0.026	0.95 ± 0.04
70	3-NH_2	−1.23	1.95	−0.16	0.12 ± 0	1.4 ± 0.1
71	3-NO_2	−0.28	3.04	0.71	0.13 ± 0.06	0.76 ± 0.18
72	4-*i*Pr	1.53	4.85	−0.15	0.10 ± 0.01	> 32
73	4-*t*Bu	1.98	5.20	−0.20	0.095 ± 0.015	> 32
74	4-Ph	1.96	5.17	−0.01	0.09 ± 0	> 32
75	4-CF_3	0.88	4.48	0.54	0.03 ± 0.01	1.4 ± 0.5
76	4-CH_2OH	−1.03	2.33	0.0	0.54 ± 0.12	3.7 ± 1.7
77	4-CH_2O*t*Bu	1.55	4.48	−0.32	0.077 ± 0.012	3.1 ± 2.0
78	4-CH_2NHPh	1.0	4.42	−0.15	0.060 ± 0.016	0.63 ± 0.30
79	4-CHO	−0.65	2.82	0.42	0.20 ± 0.07	1.0 ± 0.1
80	4-CN	−0.57	2.83	0.66	0.025 ± 0.005	0.58 ± 0.33
81	4-$CONH_2$	−1.49	1.92	0.36	2.1 ± 1.1	15 ± 5
82	4-COMe	−0.55	3.05	0.50	0.04 ± 0.01	0.73 ± 0.22
83	4-F	0.14	3.44	0.06	0.015 ± 0.005	1.4 ± 0.5
84	4-Cl	0.71	4.08	0.23	0.015 ± 0.005	2.7 ± 0.6
85	4-OH	−0.67	2.73	−0.37	0.64 ± 0.23	4.3 ± 2.1
86	4-OMe	−0.02	3.32	−0.27	0.065 ± 0.005	6.8 ± 3.2
87	4-O*i*Pr	0.85	4.20	−0.45	0.25 ± 0.03	3.8 ± 1.8
88	4-OPh	2.08	5.20	−0.03	0.04 ± 0.01	2.7 ± 1.2
89	4-O$(CH_2)_2$OH	−1.47	2.35	−0.16	0.55 ± 0.31	3.3 ± 1.6
90	4-O$(CH_2)_3$OH	−1.0	3.13	−0.17	0.60 ± 0.16	2.2 ± 1.3
91	4-O$(CH_2)_3$Nmorph[e]	−1.0	3.11	−0.17	0.097 ± 0.026	1.0 ± 0
92	4-OCF_2H	0.69	3.50	0.18	0.05 ± 0.01	0.77 ± 0.24
93	4-OCF_3	1.04	4.36	0.35	0.035 ± 0.015	1.3 ± 0.1
94	4-SMe	0.61	4.00	0.0	0.075 ± 0.015	0.95 ± 0.01
95	4-SO_2Me	−1.63	1.68	0.72	0.67 ± 0.20	14 ± 3
96	4-NH_2	−1.23	2.30	−0.66	0.49 ± 0.23	1.9 ± 1.0
97	2-Cl, 4-CF_3	1.59	5.15	0.77	0.03 ± 0	1.4 ± 0.1
98	2-Cl, 4-OCF_3	1.75	5.07	0.58	0.04 ± 0.01	0.78 ± 0.03
99	2-Cl, 6-OMe	0.69	3.89	−0.04	0.055 ± 0.005	0.74 ± 0.04
100	2-F, 4-OCF_3	1.18	4.92	0.41	0.045 ± 0.015	0.72 ± 0.29
101	2-F, 6-OMe	0.12	3.73	−0.21	0.13 ± 0.02	0.81 ± 0.24
102	2,6-diMe	1.12	4.43	−0.34	0.39 ± 0.06	0.93 ± 0.25
103	2,6-diOMe	−0.04	2.72	−0.54	0.31 ± 0.06	1.0 ± 0.1
104	3,4-diF	0.28	3.38	0.40	0.03 ± 0	1.2 ± 0.3
105	3-Cl, 4-CF_3	1.59	5.19	0.91	0.06 ± 0.03	0.97 ± 0.44
106	3-Cl, 4-OCF_3	1.75	4.88	0.72	0.03 ± 0	0.90 ± 0.12
107	3-OCF_3, 4-Cl	1.75	4.83	0.61	0.040 ± 0.014	0.95 ± 0.15
108	3-CF_3, 4-Cl	1.59	5.19	0.66	0.035 ± 0.005	1.9 ± 0.2
109	3-NO_2, 4-OCF_3	0.76	3.76	1.06	0.045 ± 0.015	1.6 ± 0.1
110	3-F, 4-OMe	0.12	3.30	0.07	0.040 ± 0.014	1.9 ± 0.8
111	3-F, 4-OCF_3	1.18	4.33	0.69	0.03 ± 0	0.34 ± 0.16
112	3-OMe, 4-F	0.12	3.16	0.18	0.045 ± 0.015	1.5 ± 0.1
113	3-OCF_2H, 4-Cl	1.40	3.97	0.54	0.03 ± 0	1.1 ± 0.4
114	3-OH, 4-Cl	0.04	3.30	0.35	0.20 ± 0.01	2.1 ± 0.5
115	3,5-diOMe	−0.04	2.92	0.24	0.28 ± 0.06	0.79 ± 0.24
116	2-OMe, 3,5-diF	0.26	3.28	0.41	0.04 ± 0.01	0.86 ± 0.22
117	2,6-diMe, 4-OMe	1.10	4.25	−0.61	0.58 ± 0.07	0.94 ± 0.44
118	3,4,5-triF	0.42	3.35	0.74	0.04 ± 0.01	0.76 ± 0.26
119	3,5-diMe, 4-OH	0.45	3.65	−0.51	0.30 ± 0.08	1.5 ± 0.6
120	3,5-diF, 4-OMe	0.26	3.31	0.41	0.045 ± 0.015	1.3 ± 0.4
121	3,4-benzo	1.16	4.74	0.10	0.045 ± 0.025	0.87 ± 0.13

[a] Sum of substituent π values. [b] log *P* calculated with ACD LogP/LogD prediction software (v.8.0; Advanced Chemistry Development Inc., Toronto, Canada). [c] Sum of substituent σ values. [d] Minimum inhibitory concentration, determined under aerobic (MABA)[23] or anaerobic (LORA)[22] conditions. Each value is the mean of at least two independent determinations (29 determinations for **1**) ± SD. [e] *N*-Morpholinyl.

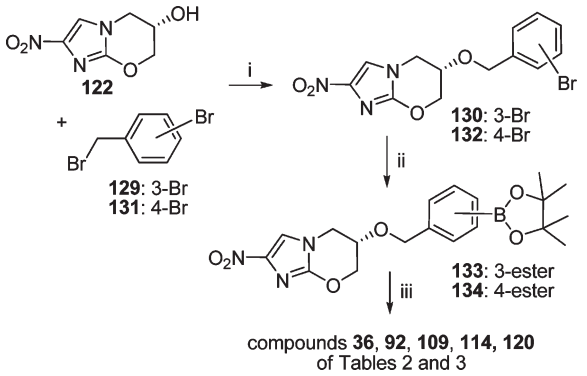
the mean of at least two separate determinations (±SD). Activity against replicating *M. tb* was measured using an 8 day microplate-based assay with Alamar blue readout (added on day 7) for determination of bacterial growth (MABA).[21] Screening for activity against nonreplicating *M. tb* employed an 11 day high-throughput, luminescence-based

Scheme 1[a]



[a] Reagents and conditions: (i) NaH, DMF, 5−20 °C, 1−2 h; (ii) $ArB(OH)_2$, toluene, EtOH, 2 M K_2CO_3, $Pd(dppf)Cl_2$ under N_2, reflux, 5−150 min.

Scheme 2[a]



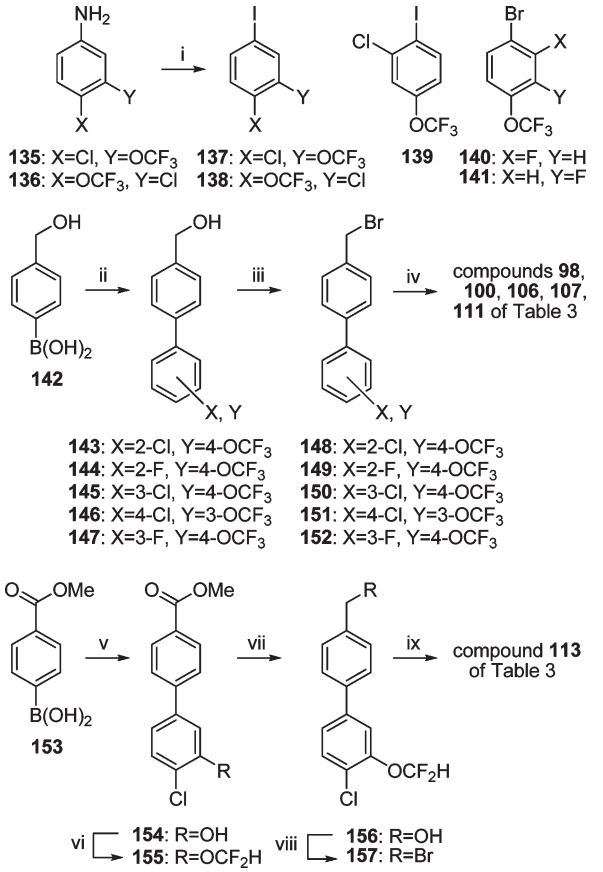
[a] Reagents and conditions: (i) NaH, DMF, 20 °C, 2 h; (ii) bis-(pinacolato)diboron, $Pd(dppf)Cl_2$, KOAc, DMSO, 90 °C, 1 h; (iii) 4-(HF_2CO)PhBr or (3-NO_2, 4-OCF_3)PhBr or (5-Br, 2-Cl)PhOH or (3,5-diF, 4-OMe)PhBr, toluene, EtOH, 2 M K_2CO_3, $Pd(dppf)Cl_2$ under N_2, reflux, 15−70 min.

low-oxygen-recovery assay (LORA), where the bacteria contained a plasmid with an acetamidase promoter driving a bacterial luciferase gene and were first adapted to low oxygen conditions by extended culture.[22] Mammalian cytotoxicity was also assessed[23] against VERO cells (CCL-81, American Type Culture Collection) in a 72 h exposure, using a tetrazolium dye assay.

Results and Discussion

Tables 1−3 provide data for 115 biphenyl analogues of **1**, including their activities against both replicating and non-replicating cultures of *M. tb*. All of the compounds were relatively nontoxic to mammalian VERO cells, with IC_{50}s >125 μM (data not shown). General inspection of Tables 1−3 shows that the *para*-linked biphenyl analogues are the most active, followed by the *meta*-linked ones, with the *ortho*-linked analogues being the least active. This relationship seems to hold regardless of the nature and positioning of substituents on the terminal benzene ring and suggests that the binding pocket is reasonably linear and restricted, at least around the inner benzyl ether ring (moderate steric bulk was well tolerated around the terminus of the second benzene ring, as shown by 3,4-benzo-fused compounds **19**, **41**, and **121** and by 3- and 4-Ph/OPh analogues, **55**, **74**, and **88**). This is demonstrated more quantitatively in Table 4, which shows a comparison of all of the *ortho*-linked analogues of Table 1 with their corresponding *meta*- and *para*-linked analogues bearing the same substituents on the terminal ring. The mean MICs for this representative set of compounds are 3.0, 0.19, and 0.05 μM in MABA and 7.9, 2.8, and 1.4 μM in LORA, respectively. The difference between the MICs in the LORA

Scheme 3[a]



[a] Reagents and conditions: (i) aqueous $NaNO_2$, 25% H_2SO_4, 0 °C, 10−12 min, then aqueous KI, 20 °C, 10−15 min, then 51 °C, 2 h; (ii) halobenzene (**137**, **138**, **139**, **140**, or **141**), toluene, EtOH, 2 M Na_2CO_3, $Pd(dppf)Cl_2$ under N_2, reflux, 1−3.5 h; (iii) 33% HBr/AcOH, 20 °C, 6−11 h; (iv) **122**, NaH, DMF, 0−20 °C, 3 h; (v) (5-Br, 2-Cl)PhOH, toluene, EtOH, 2 M K_2CO_3, $Pd(dppf)Cl_2$ under N_2, reflux, 30 min; (vi) $NaOCOCClF_2$, K_2CO_3, DMF, 80 °C, 14 h; (vii) $LiAlH_4$, Et_2O, 0−20 °C, 2 h; (viii) PBr_3, Et_2O, 0−20 °C, 3 h; (ix) **122**, NaH, DMF, 0−20 °C, 2 h.

and MABA assays is much smaller for the *ortho*-linked compounds as compared to their *meta*- and *para*-linked analogues (2.6-fold, compared to 14.5- and 28.4-fold, respectively), and there is little correlation between the two sets of assay data. This may indicate that a different mechanism of activation applies under aerobic and anaerobic conditions. Across this particular data set (Table 4), no single substituent gave consistently improved MIC results compared to the parent (H) compounds (**7**, **20**, and **42**); nevertheless, slightly improved potencies were observed in several *para*-linked examples (e.g., 4-CN, **80**; 4-F, **83**; 4-OCF_3, **93**).

In general, addition of a second aryl ring to the benzyl ether side chain of **1** led to analogues with lower aqueous solubilities than **1**, typically 0.5−5 μg/mL at pH = 7 for selected compounds from Tables 1−3 (compared with 19 μg/mL for **1** and 0.6 μg/mL for **2**; data not shown). The attachment of potentially solubilizing alcohol and amino functionality to the distal ring of *meta*- and *para*-linked biphenyls led to compounds (**27**, **28**, **37**, **38**, **50**, **64**, **65**, **66**, **76**, **89**, **90**, **91**) with *in vitro* activities generally comparable to **1**, although amino analogues **28**, **38**, and **66** were 4−7-fold less potent than **1** in the LORA assay. Overall, the *para*-linked biphenyl analogues of **1** were significantly more potent than **1** itself in both the MABA and LORA assays (an order of magnitude in MABA), as were

several of the *meta*-linked compounds. The set of *para*-linked biphenyl analogues was therefore expanded to explore the SAR in more detail.

Previous reports have suggested that a lipophilic side chain confers higher potency in both the nitroimidazooxazine and -oxazole classes, as shown[3] by the comparison of **1** (log *P* 2.70) with the less effective **3** (log *P* 0.70) and by the much greater inhibitory activity[2,5] of the even more lipophilic **2** (log *P* 4.75). In the present data set (Table 3) there were no clear relationships between MABA MIC values and measures of global lipophilicity (calculated log *P* values) for the (smaller) sets of *ortho*- and *meta*-substituted derivatives (**43**−**71**). However, in the larger set of *para*-linked biphenyls containing a 4-substituent (compounds **72**−**121**) there was a weak but significant and useful relationship (eq 1) between aerobic MIC (albeit having somewhat limited range) and the sum of π values of the substituents, suggesting that lipophilic compounds were more effective:

$$\log(\mathrm{MIC_{MABA}}) = -0.29(\pm 0.06)\sum\pi - 0.91(\pm 0.07) \quad (1)$$
$$n = 50 \quad R = 0.56 \quad F = 22.3$$

The addition of a second parameter ($\sum\sigma$) measuring the summed electronic properties of the ring B substituents gave an equation with an improved fit, showing that this variable was also significant across a broad range of values ($\sum\sigma$ from −0.66 to +1.06):

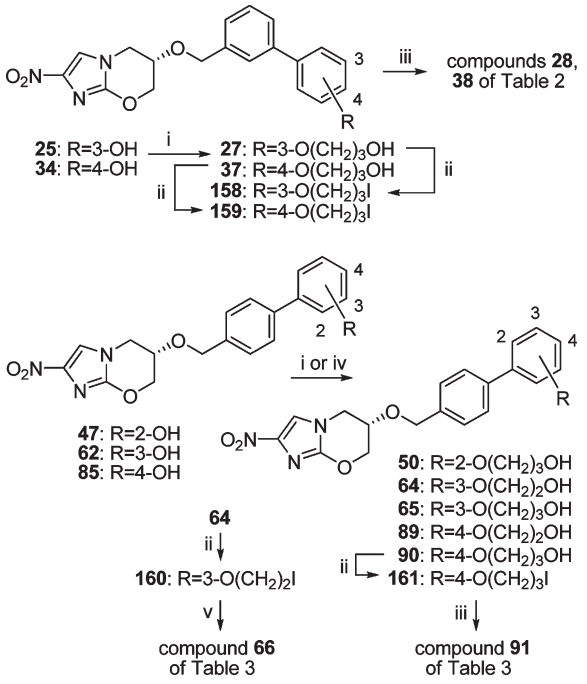
$$\log(\mathrm{MIC_{MABA}}) = -0.26(\pm 0.05)\sum\pi - 0.52(\pm 0.12)$$
$$\sum\sigma - 0.82(\pm 0.06)$$
$$n = 50 \quad R = 0.72 \quad F = 24.6$$
$$(\pi/\sigma \text{ cross-correlation coefficient } 0.15) \quad (2)$$

Attempted fits using other electronic-related (*F*, *R*) and lipophilicity-related (MR, PSA) parameters were examined but were not as good. Given the close cross-correlation between $\sum\pi$ for the ring B substituents and overall log *P* for the compounds ($R = 0.96$), the related eq 3 using overall log *P* values (which may be of more general utility for compounds that incorporate additional changes) was very similar:

$$\log(\mathrm{MIC_{MABA}}) = -0.25(\pm 0.06)\log P - 0.52(\pm 0.13)$$
$$\sum\sigma - 0.014(\pm 0.22)$$
$$n = 50 \quad R = 0.69 \quad F = 21.1$$
$$(\log P/\sigma \text{ cross-correlation coefficient } 0.15) \quad (3)$$

Equations 2 and 3 suggest that, in addition to the importance of overall higher compound lipophilicity, possibly to facilitate penetration of the exceptionally lipophilic cell wall of *M. tb*,[24] a relatively electron-deficient terminal ring is

Scheme 4[a]



[a] Reagents and conditions: (i) $Br(CH_2)_3OTBDMS$, Cs_2CO_3, DMF, 90 °C, 1−2 h, then TBAF, THF, 20 °C, 2 h; (ii) MsCl, Et_3N, THF, 0 °C, 1−2.5 h, then NaI, Me_2CO, reflux, 1−3 h; (iii) morpholine, DMA, 20 °C, 16 h; (iv) $Br(CH_2)_2OTHP$, Cs_2CO_3, DMF, 90 °C, 2 h, then MsOH, MeOH, 20 °C, 1 h; (v) aqueous Me_2NH, DMA, 20 °C, 16 h.

Table 4. Comparison of MICs for 13 Sets of *ortho*/*meta*/*para*-Linked Biphenyl Analogues

	ortho-link			*meta*-link			*para*-link		
		MIC (μM)			MIC (μM)			MIC (μM)	
substituent	no.	MABA	LORA	no.	MABA	LORA	no.	MABA	LORA
H	**7**	0.64	2.5	**20**	0.095	2.1	**42**	0.045	3.9
2-OCF_3	**8**	1.2	6.2	**21**	0.46	3.3	**51**	0.035	0.97
3-CN	**9**	0.80	8.2	**23**	0.17	3.7	**58**	0.12	1.3
3-F	**10**	0.62	3.0	**24**	0.18	2.7	**60**	0.045	2.2
3-OCF_3	**11**	1.9	6.3	**26**	0.40	3.5	**67**	0.077	1.4
3-SMe	**12**	0.81	3.2	**29**	0.07	1.4	**69**	0.077	0.95
4-COMe	**13**	17	36	**30**	0.30	1.9	**82**	0.04	0.73
4-CN	**14**	8.4	19	**32**	0.23	4.8	**80**	0.025	0.58
4-F	**15**	1.2	3.4	**33**	0.077	3.8	**83**	0.015	1.4
4-OCF_3	**16**	2.3	4.3	**35**	0.11	2.2	**93**	0.035	1.3
4-SMe	**17**	1.3	3.4	**39**	0.095	0.93	**94**	0.075	0.95
3-F, 4-OMe	**18**	1.8	3.8	**40**	0.19	2.2	**110**	0.04	1.9
3,4-benz	**19**	1.6	3.1	**41**	0.12	3.2	**121**	0.045	0.87
means		3.04	7.88		0.19	2.75		0.05	1.42

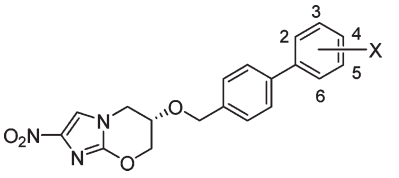
separately beneficial. This suggests that charge-transfer binding may be important at the binding site of the nitroreductase and is consistent with the empirical dominance of lipophilic, electron-withdrawing groups such as CF_3 and OCF_3 in the design of the more effective compounds. It also points to the potential use of halogens as suitable substituents.

Examining SAR for the expanded *para*-linked biphenyl class (Table 3) in more detail, there was only a slight potency advantage for 4-substitution over 3- or 2-substitution (6/9 cases in MABA), with 4-CF_3 (**75**) and 4-Cl (**84**) analogues proving particularly effective, in addition to the 4-CN (**80**), 4-F (**83**), and 4-OCF_3 (**93**) examples identified previously. However, 4-SO_2Me (**95**) and 3- or 4-$CONH_2$ (**59**, **81**) analogues were notably poor. The combination of two or more substituents (compounds **97**−**120**) generally retained or enhanced MIC potency, particularly in LORA, with **111** (3-F, 4-OCF_3: LORA MIC 0.34 μM) being the most potent of all the compounds evaluated in this assay.

Selected compounds were evaluated for their efficacy in a mouse model of acute *M. tb* infection, using a once daily oral dose of 100 mg/kg for 5 days a week for 3 weeks, following established protocols.[23] The choice of compounds was primarily based on their MIC potencies, with particular focus on compounds with good dual activity against both replicating and nonreplicating bacteria, but a variety of different types of substituents were also studied. The clinical trial drug **1** was employed as an internal standard, with activity recorded (Table 5) as the ratio of the fold decrease in colony forming units (CFUs) recovered from the lungs of compound-treated mice compared to the corresponding fold CFU decrease achieved by treatment with **1**, to allow interexperiment comparisons (see Supporting Information for raw CFU data for the compounds of Table 5). The data in Table 5 first illustrate the apparent liability incurred by alkoxy groups on the molecule, in that compounds **49**, **50**, and **91** (and, to a lesser extent, **68**) were markedly inferior to **1**. Even when a methoxy group was paired with a more electron-withdrawing group (**101**, **110**, and **116**) the *in vivo* results were still very poor. Similarly, anilino, ketone, and phenoxy-substituted analogues (**78**, **82**, and **88**) displayed almost no efficacy at all in this model. Conversely, compounds bearing lipophilic, electron-withdrawing groups (**75**, **93**, **98**, **100**, **106**, **111**, and **113**), particularly at the terminus (3- and 4-positions), were substantially more active than **1** (> 10-fold). The three best candidates were **93** (4-OCF_3), **106** (3-Cl, 4-OCF_3), and **111** (3-F, 4-OCF_3), which were all > 200-fold more efficacious than **1** in this model.

The strong *in vivo* efficacy of some of the most lipophilic analogues following oral dosing (as observed for **2**)[4,5] is consistent with significant oral bioavailability for this class, in addition to an improved binding to the nitroreductase. Thus, in order to further understand the divergent *in vivo* results above, the compounds were also evaluated for their stability in a metabolism screen with human and mouse liver microsome preparations. The compounds were incubated for 1 h at 37 °C, and the percentage of compound remaining was determined by LC-MS-MS. The results (Table 5) allow some SAR to be derived. The parent drug **1** showed good stability toward both human and mouse microsomes (82% and 94% remaining, respectively). Five analogues bearing *O*-alkyl groups (**49**, **50**, **91**, **101**, and **116**) had poor stabilities, especially in mouse microsomes (**68** and **110** also showed marginal stabilities in mouse microsomes). The remainder of the compounds were relatively stable in the mouse assay (less than 50% loss in 1 h), but a further three (**78**, **82**, and **88**) were markedly unstable toward human microsomes. Thus the lowest stability compounds were those bearing an alkoxy or phenoxy group or having a ketone substituent or a free NH function. Conversely, compounds that showed high stabilities in both assays were those with halogen and/or CF_3/OCF_3 substituents (**75**, **93**, **97**, **98**, **100**, **106**, **107**, **108**, and **111**), reinforcing the apparent utility of these substituents. Compounds **92** and **113** also showed the usefulness of the OCF_2H group, although these compounds appeared to be slightly less stable than their OCF_3 counterparts (**93** and **107**).

Table 5. Microsomal Stability and *in Vivo* Efficacy Data for Selected Analogues



compd	X	microsomes (% remaining at 1 h) H[a]	microsomes (% remaining at 1 h) M[b]	*in vivo* efficacy[c] (ratio vs **1**)
1		82	94	1.00
49	2-OEt	50	0.2	0.15
50	2-O$(CH_2)_3$OH	59	21	< 0.01
68	3-OCH_2Ph	64	50	0.66
75	4-CF_3	60	85	72
78	4-CH_2NHPh	12	52	0.03
80	4-CN	95	58	3.6
82	4-COMe	36	70	< 0.01
83	4-F	75	55	3.0
88	4-OPh	22	48	< 0.01
91	4-O$(CH_2)_3$Nmorph[d]	11	14	0.05
92	4-OCF_2H	94	77	8.5
93	4-OCF_3	97	96	> 205
97	2-Cl, 4-CF_3	92	93	7.2
98	2-Cl, 4-OCF_3	91	86	23
100	2-F, 4-OCF_3	88	89	46
101	2-F, 6-OMe	91	0	0.07
106	3-Cl, 4-OCF_3	90	84	281
107	3-OCF_3, 4-Cl	95	92	9.7
108	3-CF_3, 4-Cl	92	96	7.7
110	3-F, 4-OMe	80	50	0.02
111	3-F, 4-OCF_3	93	86	419
113	3-OCF_2H, 4-Cl	83	86	10
116	2-OMe, 3,5-diF	72	2	0.40

[a] Pooled human liver microsomes. [b] Pooled CD-1 mouse liver microsomes. [c] Fold reduction in lung CFU for compound compared with the fold CFU reduction for **1** in a mouse model of acute TB infection (see text). [d] *N*-Morpholinyl.

Across the whole data set, the microsomal stability results appear, at least for this group of compounds, to be usefully predictive for *in vivo* activity. If the microsomal assay data are crudely quantified as the sum of the two "% compound remaining" numbers, and the >, < modifiers are removed from the fold reduction numbers, eq 4 shows that there is a reasonable correlation between microsomal stability and *in vivo* efficacy:

$$\log(\text{fold reduction in CFU}) = 4.84(\pm 0.99) \log(\text{microsomal stability}) - 9.82$$
$$n = 23 \quad R = 0.73 \quad F = 23.8 \qquad (4)$$

Conclusions

This study has demonstrated the potential utility of particularly *para*-linked biphenyl-based side chains to provide highly potent analogues of **1**, having significantly improved activity *in vitro* against both replicating and nonreplicating *M. tb*. Seven of the compounds showed substantially (> 10-fold) improved efficacies over **1** when evaluated in a mouse model of acute *M. tb* infection, with three of these being > 200-fold more effective than **1**. This is the first study to report analogues of **1** with improved efficacy *in vivo*. Overall, there appears to be predictive correlations between MABA MIC data and the lipophilicity and electron-withdrawing properties of substituent groups on the distal ring of the side chain. The ability of the compounds to resist degradation by liver microsome preparations may also be a useful indicator of their potential *in vivo* activity.

Experimental Section

Analyses were performed by the Campbell Microanalytical Laboratory, University of Otago, Dunedin, New Zealand. Melting points were determined using an Electrothermal Model 9200 melting point apparatus and are as read. NMR spectra were measured in $(CD_3)_2SO$ (unless otherwise specified) on a Bruker Avance 400 spectrometer at 400 MHz for ^1H and are referenced to Me_4Si. Chemical shifts and coupling constants are recorded in units of parts per million and hertz, respectively. High-resolution electron impact (HREIMS) and fast atom bombardment (HRFABMS) mass spectra were determined on a VG-70SE mass spectrometer at nominal 5000 resolution. High-resolution atmospheric pressure chemical ionization (HRAPCIMS) mass spectra were determined on a Bruker micrOTOF-Q II mass spectrometer. Thin-layer chromatography was carried out on aluminum-backed silica gel plates (Merck 60 F_{254}) with visualization of components by UV light (254 nm) or exposure to I_2. Column chromatography was carried out on silica gel (Merck 230−400 mesh). Tested compounds were ≥95% pure, as determined by combustion analysis, or by HPLC conducted on an Agilent 1100 system, using a reversed-phase C8 column with diode array detection.

Preparation of Biphenyl Analogues from Iodobenzyl Ethers and Arylboronic Acids (Scheme 1). (6*S*)-6-[(2-Iodobenzyl)oxy]-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (124). A solution of (6*S*)-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazin-6-ol[13] (**122**) (3.188 g, 17.2 mmol) and 2-iodobenzyl chloride (**123**) (5.00 g, 19.8 mmol) in anhydrous DMF (60 mL) was treated at 5 °C with 60% NaH (0.893 g, 22.3 mmol). The mixture was warmed to room temperature, stirred for 1 h, and then quenched with water, and the precipitate was collected and triturated in Et_2O to give **124** (4.19 g, 61%): mp (Et_2O) 114−117 °C; ^1H NMR δ 8.04 (s, 1 H), 7.86 (d, J = 7.7 Hz, 1 H), 7.41−7.35 (m, 2 H), 7.10−7.04 (m, 1 H), 4.69−4.61 (m, 3 H), 4.51 (d, J = 11.6 Hz, 1 H), 4.35−4.23 (m, 3 H). Anal. ($C_{13}H_{12}IN_3O_4$) C, H, N, I. HPLC purity: 98.7%.

(6*S*)-6-[(3-Iodobenzyl)oxy]-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (126). Similar reaction of alcohol **122** with 3-iodobenzyl bromide (**125**) and NaH in DMF for 1 h at room temperature (as described for **124**) gave **126** (85%): mp (EtOAc/petroleum ether) 144−146 °C; ^1H NMR δ 8.01 (s, 1 H), 7.67−7.64 (m, 2 H), 7.33 (d, J = 7.6 Hz, 1 H), 7.16 (br t, J = 8.1 Hz, 1 H), 4.67−4.62 (m, 2 H), 4.60 (d, J = 12.3 Hz, 1 H), 4.47 (d, J = 12.0 Hz, 1 H), 4.28−4.20 (m, 3 H). Anal. ($C_{13}H_{12}IN_3O_4 \cdot 0.25$EtOAc) C, H, N.

(6*S*)-6-[(4-Iodobenzyl)oxy]-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (128). Similar reaction of alcohol **122** with 4-iodobenzyl bromide (**127**) and NaH in DMF for 2 h at room temperature (as described for **124**) gave **128**[13] (97%): mp (EtOAc/petroleum ether) 210−212 °C; ^1H NMR δ 8.01 (s, 1 H), 7.71 (dt, J = 8.3, 2.0 Hz, 2 H), 7.13 (br d, J = 8.3 Hz, 2 H), 4.67−4.60 (m, 2 H), 4.59 (d, J = 12.2 Hz, 1 H), 4.46 (d, J = 12.0 Hz, 1 H), 4.27−4.19 (m, 3 H). Anal. ($C_{13}H_{12}IN_3O_4$) C, H, N.

General Method for Suzuki Coupling. Iodides **124**, **126**, or **128** (1 equiv) and arylboronic acids (1.3 equiv) were suspended in toluene/EtOH (5 mL/2 mL per 100 mg of iodide), and then aqueous K_2CO_3 (1 mL of 2 M per 100 mg of iodide) was added. The resulting mixture was purged with N_2, treated with Pd(dppf)Cl_2 (5 mol %), and heated under reflux in an N_2 atmosphere for 20 min (or longer if noted). The mixture was diluted with water and extracted with EtOAc (3×). The dried ($MgSO_4$) organic layers were adsorbed onto silica gel and chromatographed, eluting first with EtOAc/hexane (1:1) and then with EtOAc to give the product. For more polar compounds, EtOAc/MeOH (95:5) or CH_2Cl_2/MeOH (95:5) was used. Trituration of the product in Et_2O gave the pure compounds.

Details of the preparation of compounds for Tables 1−3 by this method and their characterization (mp, ^1H NMR, and either combustion analysis or HPLC) are given in Supporting Information.

Preparation of Biphenyl Analogues via Boronic Acid Pinacol Esters and Aryl Halides (Scheme 2). (6*S*)-6-[(3-Bromobenzyl)oxy]-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (130). Reaction of alcohol **122** with 3-bromobenzyl bromide (**129**) and NaH in DMF for 2 h at room temperature (as described above for the preparation of **124**) gave **130** (93%): mp (EtOAc/petroleum ether) 59−62 °C; ^1H NMR δ 8.01 (s, 1 H), 7.50−7.47 (m, 2 H), 7.33−7.30 (m, 2 H), 4.69−4.65 (m, 2 H), 4.63 (d, J = 12.3 Hz, 1 H), 4.47 (d, J = 12.0 Hz, 1 H), 4.28−4.20 (m, 3 H). Anal. ($C_{13}H_{12}BrN_3O_4$) C, H, N.

(6*S*)-6-[(4-Bromobenzyl)oxy]-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (132). Similar reaction of alcohol **122** with 4-bromobenzyl bromide (**131**) and NaH in DMF for 2 h at room temperature (as described for **124**) gave **132** (88%): mp (Et_2O) 188−190 °C; ^1H NMR δ 8.01 (s, 1 H), 7.54 (dt, J = 8.4, 2.2 Hz, 2 H), 7.13 (dt, J = 8.5, 2.2 Hz, 2 H), 4.67−4.62 (m, 2 H), 4.61 (d, J = 12.2 Hz, 1 H), 4.46 (d, J = 12.0 Hz, 1 H), 4.28−4.19 (m, 3 H). Anal. ($C_{13}H_{12}BrN_3O_4$) C, H, N.

(6*S*)-2-Nitro-6-[3-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-benzyloxy]-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (133). A mixture of the 3-bromide **130** (1.50 g, 4.24 mmol) and bis(pinacolato)diboron (1.195 g, 4.70 mmol) and KOAc (2.55 g, 26.0 mmol) in DMSO (40 mL) was purged with N_2. Pd(dppf)Cl_2 (0.15 g, 0.20 mmol) was added, and the mixture was then stirred at 90 °C for 1 h while being continuously purged with N_2 (gas bubbled through the solution). The resulting cooled mixture was partitioned between EtOAc and water, and the organic extract was evaporated to dryness and then purified by chromatography on silica gel, eluting with EtOAc. The product was triturated in Et_2O to give **133** (1.033 g, 61%): mp 168−171 °C; ^1H NMR δ 8.01 (s, 1 H), 7.62−7.58 (m, 2 H), 7.44 (dt, J = 7.8, 1.5 Hz, 1 H), 7.36 (t, J = 7.5 Hz, 1 H), 4.68−4.61 (m, 3 H), 4.47 (d, J = 11.9 Hz, 1 H), 4.30−4.20 (m, 3 H), 1.28 (s, 12 H). Anal. ($C_{19}H_{24}BN_3O_6$) C, H, N.

(6*S*)-2-Nitro-6-[4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-benzyloxy]-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (134). Similar reaction of the 4-bromide **132** with bis(pinacolato)-diboron, as described above for the preparation of **133**, followed by chromatography on silica gel (eluting with EtOAc) and trituration in Et_2O, gave **134** (51%): mp 150−153 °C; ^1H NMR δ 8.01 (s, 1 H), 7.65 (d, J = 8.0 Hz, 2 H), 7.32 (d, J = 8.0 Hz, 2 H), 4.70 (d, J = 12.5 Hz, 1 H), 4.67−4.63 (m, 2 H), 4.46 (d, J = 11.9 Hz, 1 H), 4.29−4.20 (m, 3 H), 1.29 (s, 12 H). Anal. ($C_{19}H_{24}BN_3O_6$) C, H, N.

(6*S*)-6-{[4′-(Difluoromethoxy)[1,1′-biphenyl]-3-yl]methoxy}-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (36). Reaction of **133** and 1-bromo-4-(difluoromethoxy)benzene under the general Suzuki conditions described above for 15 min gave **36** (54%) as a white solid: mp 113−115 °C; ^1H NMR δ 8.02 (s, 1 H), 7.67 (d, J = 8.8 Hz, 2 H), 7.60−7.53 (m, 2 H), 7.44 (t, J = 7.6 Hz, 1 H), 7.31 (br d, J = 7.6 Hz, 1 H), 7.27 (t, J_{H-F} = 74.1 Hz, 1 H),

7.25 (d, J = 8.8 Hz, 2 H), 4.75−4.63 (m, 3 H), 4.49 (d, J = 11.9 Hz, 1 H), 4.32−4.20 (m, 3 H). Anal. ($C_{20}H_{17}F_2N_3O_5$) C, H, N.

(6*S*)-6-{[4′-(Difluoromethoxy)[1,1′-biphenyl]-4-yl]methoxy}-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (92). Reaction of **134** and 1-bromo-4-(difluoromethoxy)benzene under the general Suzuki conditions described above for 15 min gave **92** (77%) as a cream solid: mp 188−191 °C; ^{1}H NMR δ 8.02 (s, 1 H), 7.71 (d, J = 8.8 Hz, 2 H), 7.63 (d, J = 8.3 Hz, 2 H), 7.40 (d, J = 8.3 Hz, 2 H), 7.26 (t, J_{H-F} = 74.1 Hz, 1 H), 7.25 (d, J = 8.8 Hz, 2 H), 4.73−4.64 (m, 3 H), 4.48 (d, J = 11.9 Hz, 1 H), 4.32−4.21 (m, 3 H). Anal. ($C_{20}H_{17}F_2N_3O_5$) C, H, N.

(6*S*)-2-Nitro-6-{[3′-nitro-4′-(trifluoromethoxy)[1,1′-biphenyl]-4-yl]methoxy}-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (109). A stirred mixture of **134** (50.1 mg, 0.125 mmol) and Pd(dppf)Cl_2 (9.2 mg, 0.0126 mmol) in toluene (2 mL) and EtOH (1 mL) was degassed for 4 min (vacuum pump), and then N_2 was added. Aqueous Na_2CO_3 (0.35 mL of 2 M, 0.70 mmol) and 4-bromo-2-nitro-1-(trifluoromethoxy)benzene (52 mg, 0.182 mmol) were sequentially added by syringe, the stirred mixture was again degassed for 4 min, and then N_2 was added. The resulting mixture was stirred at 90 °C for 70 min and then cooled, diluted with aqueous $NaHCO_3$ (50 mL), and extracted with CH_2Cl_2 (5 × 50 mL). The extracts were evaporated to dryness, and the residue was chromatographed on silica gel. Elution with 0−0.5% EtOAc/CH_2Cl_2 gave foreruns, and then elution with 1.5−2.5% EtOAc/CH_2Cl_2 gave **109** (48 mg, 80%) as a light yellow solid: mp (CH_2Cl_2/pentane) 165−168 °C; ^{1}H NMR ($CDCl_3$) δ 8.16 (d, J = 2.3 Hz, 1 H), 7.83 (dd, J = 8.6, 2.3 Hz, 1 H), 7.58 (dt, J = 8.3, 1.8 Hz, 2 H), 7.52 (dq, J = 8.6, 1.4 Hz, 1 H), 7.44 (br d, J = 8.3 Hz, 2 H), 7.39 (s, 1 H), 4.80 (d, J = 12.1 Hz, 1 H), 4.69 (d, J = 12.1 Hz, 1 H), 4.65 (ddd, J = 12.2, 3.7, 2.1 Hz, 1 H), 4.38 (dd, J = 12.1, 1.3 Hz, 1 H), 4.25−4.13 (m, 3 H). Anal. ($C_{20}H_{15}F_3N_4O_7$) C, H, N.

4-Chloro-4′-({[(6*S*)-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*]-[1,3]oxazin-6-yl]oxy}methyl)[1,1′-biphenyl]-3-ol (114). Reaction of **134** and 5-bromo-2-chlorophenol under the general Suzuki conditions described above for 1 h gave **114** (76%) as a light yellow powder: mp 92−95 °C; ^{1}H NMR δ 10.26 (br s, 1 H), 8.03 (s, 1 H), 7.55 (d, J = 8.3 Hz, 2 H), 7.42−7.37 (m, 3 H), 7.20 (d, J = 2.1 Hz, 1 H), 7.07 (dd, J = 8.3, 2.1 Hz, 1 H), 4.72−4.63 (m, 3 H), 4.48 (d, J = 11.9 Hz, 1 H), 4.32−4.21 (m, 3 H). Anal. ($C_{19}H_{16}ClN_3O_5$) C, H, N.

(6*S*)-6-[(3′,5′-Difluoro-4′-methoxy[1,1′-biphenyl]-4-yl)methoxy]-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (120). Reaction of **134** and 5-bromo-1,3-difluoro-2-methoxybenzene under the general Suzuki conditions described above for 30 min gave **120** (70%) as a white solid: mp 168−171 °C; ^{1}H NMR δ 8.02 (s, 1 H), 7.68 (d, J = 8.3 Hz, 2 H), 7.53−7.45 (m, 2 H), 7.39 (d, J = 8.3 Hz, 2 H), 4.70−4.64 (m, 3 H), 4.48 (d, J = 11.9 Hz, 1 H), 4.29−4.23 (m, 3 H), 3.95 (s, 3 H). Anal. ($C_{20}H_{17}F_2N_3O_5$) C, H, N.

Preparation of Biphenyl Analogues via Hydroxymethylbiphenyls (Scheme 3). 1-Chloro-4-iodo-2-(trifluoromethoxy)benzene (137). An ice-cold mixture of 98% H_2SO_4 (1.0 mL) and water (3.0 mL) was added to 4-chloro-3-(trifluoromethoxy)aniline (**135**) (1.00 g, 4.73 mmol), and the resulting salt was crushed (using a glass rod) and cooled in an ice bath. A solution of $NaNO_2$ (359 mg, 5.20 mmol) in cold water (0.75 mL, then 0.25 mL) was added dropwise (over 10 min with stirring), and then the mixture was stirred at 0 °C for 10 min. A solution of urea (43.5 mg, 0.724 mmol) in cold water (0.25 mL) was added, and the mixture was stirred at 0 °C for 3 min. Finally, a solution of KI (1.66 g, 10.0 mmol) in cold water (1.6 mL, then 0.2 mL) was added slowly, and the mixture was stirred at room temperature for 15 min and then at 51 °C for 2 h. The resulting mixture was cooled in ice, diluted with ice−water (45 mL), and extracted with pentane (4 × 50 mL). The extracts were sequentially washed with an aqueous solution of Na_2SO_3 (30 mL of 0.5%) and then with water (40 mL) and finally concentrated carefully under reduced pressure at 13 °C. The resulting oil was chromatographed on silica gel, eluting with pentane, to give **137** (1.33 g, 87%) as a colorless oil (a white solid on freezing): ^{1}H NMR ($CDCl_3$) δ 7.64 (quintet, J = 1.5 Hz, 1 H), 7.57 (dd, J = 8.4, 1.9 Hz, 1 H), 7.19 (d, J = 8.4 Hz, 1 H); HRAPCIMS calcd for $C_7H_3ClF_3IO$ m/z (M^+) 323.8834, 321.8864, found 323.8847, 321.8873.

2-Chloro-4-iodo-1-(trifluoromethoxy)benzene (138). Similar diazotization of 3-chloro-4-(trifluoromethoxy)aniline (**136**) (1.00 g) in a cold mixture of 98% H_2SO_4 (0.75 mL) and water (2.25 mL) (as described for **137**) and chromatography of the product on silica gel, eluting with pentane, gave **138** (1.24 g, 81%) as a colorless oil (a white solid on freezing): ^{1}H NMR ($CDCl_3$) δ 7.82 (d, J = 2.1 Hz, 1 H), 7.61 (dd, J = 8.6, 2.1 Hz, 1 H), 7.05 (dq, J = 8.6, 2.0 Hz, 1 H); HRAPCIMS calcd for $C_7H_3ClF_3IO$ m/z (M^+) 323.8834, 321.8864, found 323.8834, 321.8861.

Procedure A. [2′-Chloro-4′-(trifluoromethoxy)[1,1′-biphenyl]-4-yl]methanol (143). A stirred mixture of 4-(hydroxymethyl)-phenylboronic acid (**142**) (308 mg, 2.03 mmol) and Pd(dppf)Cl_2 (191 mg, 0.261 mmol) in toluene (22 mL) and EtOH (11 mL) was degassed for 8 min (vacuum pump), and then N_2 was added. Aqueous Na_2CO_3 (4.4 mL of 2 M, 8.8 mmol) was added by syringe, the stirred mixture was again degassed for 8 min, and then N_2 was added. 2-Chloro-1-iodo-4-(trifluoromethoxy)-benzene (**139**) (585 mg, 1.81 mmol) was added by syringe, and the resulting mixture was stirred at 88 °C for 60 min. The cooled mixture was then diluted with aqueous $NaHCO_3$ (100 mL) and extracted with CH_2Cl_2 (5 × 100 mL). The extracts were evaporated to dryness, and the residue was chromatographed on silica gel. Elution with 0−50% CH_2Cl_2/petroleum ether first gave foreruns, and then further elution with 50% CH_2Cl_2/petroleum ether gave **143** (537 mg, 98%) as a white solid: mp (pentane) 38−39 °C; ^{1}H NMR ($CDCl_3$) δ 7.46 (br d, J = 8.2 Hz, 2 H), 7.42 (dt, J = 8.3, 2.0 Hz, 2 H), 7.37 (br s, 1 H), 7.36 (d, J = 8.5 Hz, 1 H), 7.19 (m, 1 H), 4.77 (d, J = 5.9 Hz, 2 H), 1.70 (t, J = 5.9 Hz, 1 H); HREIMS calcd for $C_{14}H_{10}ClF_3O_2$ m/z (M^+) 304.0292, 302.0321, found 304.0294, 302.0317.

[2′-Fluoro-4′-(trifluoromethoxy)[1,1′-biphenyl]-4-yl]methanol (144). Reaction of **142** and 1-bromo-2-fluoro-4-(trifluoromethoxy)benzene (**140**) under the Suzuki conditions described in procedure A for 3.5 h, followed by chromatography of the product on silica gel, eluting with 0−40% CH_2Cl_2/petroleum ether (foreruns) and then 40% CH_2Cl_2/petroleum ether, gave **144** (73%) as a cream solid: mp (CH_2Cl_2/pentane) 72−74 °C; ^{1}H NMR ($CDCl_3$) δ 7.52 (dq, J = 8.3, 1.8 Hz, 2 H), 7.49−7.42 (m, 3 H), 7.13−7.03 (m, 2 H), 4.76 (d, J = 5.9 Hz, 2 H), 1.69 (t, J = 5.9 Hz, 1 H); HREIMS calcd for $C_{14}H_{10}F_4O_2$ m/z (M^+) 286.0617, found 286.0612.

[3′-Chloro-4′-(trifluoromethoxy)[1,1′-biphenyl]-4-yl]methanol (145). Reaction of **142** and **138** under the Suzuki conditions described in procedure A for 95 min, followed by chromatography of the product on silica gel, eluting with 0−40% CH_2Cl_2/petroleum ether (foreruns) and then 40% CH_2Cl_2/petroleum ether, gave **145** (84%) as a cream solid: mp (CH_2Cl_2/pentane) 65−66 °C; ^{1}H NMR ($CDCl_3$) δ 7.68 (d, J = 2.2 Hz, 1 H), 7.54 (dt, J = 8.3, 1.9 Hz, 2 H), 7.48 (dd, J = 8.5, 2.2 Hz, 1 H), 7.46 (br d, J = 8.4 Hz, 2 H), 7.38 (dq, J = 8.5, 1.4 Hz, 1 H), 4.76 (d, J = 5.9 Hz, 2 H), 1.70 (t, J = 5.9 Hz, 1 H); HREIMS calcd for $C_{14}H_{10}ClF_3O_2$ m/z (M^+) 304.0292, 302.0321, found 304.0300, 302.0319.

[4′-Chloro-3′-(trifluoromethoxy)[1,1′-biphenyl]-4-yl]methanol (146). Reaction of **142** and **137** under the Suzuki conditions described in procedure A for 120 min, followed by chromatography of the product on silica gel, eluting with CH_2Cl_2, gave **146** (77%) as a cream solid: mp (CH_2Cl_2/pentane) 70−71 °C; ^{1}H NMR ($CDCl_3$) δ 7.57−7.50 (m, 4 H), 7.49−7.43 (m, 3 H), 4.76 (d, J = 5.9 Hz, 2 H), 1.69 (t, J = 5.9 Hz, 1 H); HREIMS calcd for $C_{14}H_{10}ClF_3O_2$ m/z (M^+) 304.0292, 302.0321, found 304.0283, 302.0319.

[3′-Fluoro-4′-(trifluoromethoxy)[1,1′-biphenyl]-4-yl]methanol (147). Reaction of **142** and 4-bromo-2-fluoro-1-(trifluoromethoxy)benzene (**141**) under the Suzuki conditions described

in procedure A for 2.5 h, followed by chromatography of the product on silica gel, eluting with 0−40% CH_2Cl_2/petroleum ether (foreruns) and then 40% CH_2Cl_2/petroleum ether, gave **147** (73%) as a cream solid: mp (CH_2Cl_2/pentane) 70−71 °C; ^{1}H NMR ($CDCl_3$) δ 7.54 (dt, J = 8.3, 1.8 Hz, 2 H), 7.46 (br d, J = 8.2 Hz, 2 H), 7.41 (br d, J = 11.2 Hz, 1 H), 7.40−7.32 (m, 2 H), 4.76 (d, J = 5.9 Hz, 2 H), 1.69 (t, J = 5.9 Hz, 1 H); HREIMS calcd for $C_{14}H_{10}F_4O_2$ m/z (M^+) 286.0617, found 286.0616.

Procedure B. 4-(Bromomethyl)-2′-chloro-4′-(trifluoromethoxy)-1,1′-biphenyl (148). HBr in AcOH (5 mL of 33% w/w) was added to a solution of alcohol **143** (618 mg, 2.04 mmol) in glacial AcOH (2.5 mL), and the mixture was stirred at room temperature for 11 h. The resulting orange solution was added slowly to ice−water (50 mL) with stirring, and then the mixture was extracted with pentane (6 × 50 mL). The extracts were washed with ice−water (50 mL) and then evaporated to give **148** (743 mg, 100%) as an oil: ^{1}H NMR ($CDCl_3$) δ 7.47 (dt, J = 8.3, 1.9 Hz, 2 H), 7.39 (dt, J = 8.3, 1.9 Hz, 2 H), 7.37 (m, 1 H), 7.35 (d, J = 8.5 Hz, 1 H), 7.19 (m, 1 H), 4.55 (s, 2 H); HREIMS calcd for $C_{14}H_9BrClF_3O$ m/z (M^+) 367.9427, 365.9457, 363.9477, found 367.9428, 365.9453, 363.9485.

4-(Bromomethyl)-2′-fluoro-4′-(trifluoromethoxy)-1,1′-biphenyl (149). Bromination of alcohol **144** using procedure B for 9 h gave **149** (100%) as a cream solid that was used directly in the next step: ^{1}H NMR ($CDCl_3$) δ 7.52−7.42 (m, 5 H), 7.13−7.03 (m, 2 H), 4.54 (s, 2 H); HRAPCIMS calcd for $C_{14}H_9F_4O$ m/z [M − Br]$^+$ 269.0584, found 269.0574.

4-(Bromomethyl)-3′-chloro-4′-(trifluoromethoxy)-1,1′-biphenyl (150). Bromination of alcohol **145** using procedure B for 10 h gave **150** (100%) as a cream solid that was used directly in the next step: ^{1}H NMR ($CDCl_3$) δ 7.67 (d, J = 2.2 Hz, 1 H), 7.52 (dt, J = 8.5, 2.2 Hz, 2 H), 7.48 (dt, J = 8.6, 2.3 Hz, 2 H), 7.48 (dd, J = 8.6, 2.2 Hz, 1 H), 7.38 (dq, J = 8.5, 1.4 Hz, 1 H), 4.54 (s, 2 H); HRAPCIMS calcd for $C_{14}H_9ClF_3O$ m/z [M − Br]$^+$ 287.0260, 285.0289, found 287.0264, 285.0291.

4-(Bromomethyl)-4′-chloro-3′-(trifluoromethoxy)-1,1′-biphenyl (151). Bromination of alcohol **146** using procedure B for 6 h gave **151** (100%) as a cream solid that was used directly in the next step: ^{1}H NMR ($CDCl_3$) δ 7.55−7.46 (m, 6 H), 7.45 (dd, J = 8.3, 2.1 Hz, 1 H), 4.54 (s, 2 H); HRAPCIMS calcd for $C_{14}H_9ClF_3O$ m/z [M − Br]$^+$ 287.0260, 285.0289, found 287.0249, 285.0289.

4-(Bromomethyl)-3′-fluoro-4′-(trifluoromethoxy)-1,1′-biphenyl (152). Bromination of alcohol **147** using procedure B for 6 h gave **152** (100%) as a cream solid that was used directly in the next step: ^{1}H NMR ($CDCl_3$) δ 7.52 (dt, J = 8.5, 2.2 Hz, 2 H), 7.48 (dt, J = 8.5, 2.2 Hz, 2 H), 7.43−7.32 (m, 3 H), 4.54 (s, 2 H); HRAPCIMS calcd for $C_{14}H_9F_4O$ m/z [M − Br]$^+$ 269.0584, found 269.0572.

Procedure C. (6*S*)-6-{[2′-Chloro-4′-(trifluoromethoxy)[1,1′-biphenyl]-4-yl]methoxy}-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*]-[1,3]oxazine (98). A stirred solution of (6*S*)-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazin-6-ol[13] (**122**) (342 mg, 1.85 mmol) and bromide **148** (741 mg, 2.03 mmol) in anhydrous DMF (7 mL) under N_2 at 0 °C was treated with 60% NaH (111 mg, 2.78 mmol) and then quickly degassed and resealed under N_2. After stirring at room temperature for 3 h, the reaction was cooled (CO_2/acetone), quenched with ice/aqueous $NaHCO_3$ (20 mL), added to brine (80 mL), and extracted with CH_2Cl_2 (6×80 mL). The combined extracts were evaporated to dryness, and the residue was chromatographed on silica gel, eluting with CH_2Cl_2, to give **98** (694 mg, 80%) as a light yellow solid: mp (CH_2Cl_2/pentane) 80−82 °C; ^{1}H NMR ($CDCl_3$) δ 7.45−7.35 (m, 6 H), 7.34 (d, J = 8.5 Hz, 1 H), 7.20 (m, 1 H), 4.79 (d, J = 12.0 Hz, 1 H), 4.68 (d, J = 12.1 Hz, 1 H), 4.65 (ddd, J = 12.1, 2.9, 2.5 Hz, 1 H), 4.37 (br d, J = 11.8 Hz, 1 H), 4.24−4.12 (m, 3 H). Anal. ($C_{20}H_{15}ClF_3N_3O_5$) C, H, N.

(6*S*)-6-{[2′-Fluoro-4′-(trifluoromethoxy)[1,1′-biphenyl]-4-yl]-methoxy}-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (100). Reaction of alcohol **122** with bromide **149** (0.99 equiv) using procedure C, followed by chromatography of the product on silica gel, eluting with 0−1% EtOAc/CH_2Cl_2 (foreruns) and then 1−2% EtOAc/CH_2Cl_2, gave **100** (79%) as a light yellow solid: mp (CH_2Cl_2/hexane) 149−151 °C; ^{1}H NMR ($CDCl_3$) δ 7.52 (dq, J = 8.3, 1.7 Hz, 2 H), 7.44 (t, J = 8.5 Hz, 1 H), 7.40 (br d, J = 8.4 Hz, 2 H), 7.38 (s, 1 H), 7.13−7.04 (m, 2 H), 4.79 (d, J = 12.1 Hz, 1 H), 4.67 (d, J = 12.1 Hz, 1 H), 4.64 (ddd, J = 12.1, 3.7, 2.1 Hz, 1 H), 4.37 (dd, J = 12.1, 1.5 Hz, 1 H), 4.23−4.11 (m, 3 H). Anal. ($C_{20}H_{15}F_4N_3O_5$) C, H, N.

(6*S*)-6-{[3′-Chloro-4′-(trifluoromethoxy)[1,1′-biphenyl]-4-yl]-methoxy}-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (106). Reaction of alcohol **122** with bromide 150 (0.99 equiv) using procedure C, followed by chromatography of the product on silica gel, eluting with CH_2Cl_2 (foreruns) and then 0−2% EtOAc/CH_2Cl_2, gave **106** (70%) as a pale yellow solid: mp (CH_2Cl_2/hexane) 150−151 °C; ^{1}H NMR ($CDCl_3$) δ 7.67 (d, J = 2.2 Hz, 1 H), 7.54 (dt, J = 8.2, 1.7 Hz, 2 H), 7.47 (dd, J = 8.5, 2.2 Hz, 1 H), 7.43−7.36 (m, 4 H), 4.78 (d, J = 12.0 Hz, 1 H), 4.67 (d, J = 12.0 Hz, 1 H), 4.63 (ddd, J = 12.1, 3.6, 2.1 Hz, 1 H), 4.37 (dd, J = 12.1, 1.1 Hz, 1 H), 4.23−4.11 (m, 3 H). Anal. ($C_{20}H_{15}ClF_3N_3O_5$) C, H, N.

(6*S*)-6-{[4′-Chloro-3′-(trifluoromethoxy)[1,1′-biphenyl]-4-yl]-methoxy}-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (107). Reaction of alcohol **122** with bromide **151** (1.00 equiv) using procedure C, followed by chromatography of the product on silica gel, eluting with CH_2Cl_2 (foreruns) and then 2% EtOAc/CH_2Cl_2, gave **107** (78%) as a cream solid: mp (CH_2Cl_2/pentane) 168−171 °C; ^{1}H NMR ($CDCl_3$) δ 7.57−7.51 (m, 3 H), 7.50 (m, 1 H), 7.44 (dd, J = 8.3, 2.1 Hz, 1 H), 7.40 (br d, J = 8.2 Hz, 2 H), 7.38 (s, 1 H), 4.78 (d, J = 12.0 Hz, 1 H), 4.67 (d, J = 12.2 Hz, 1 H), 4.63 (ddd, J = 12.1, 3.7, 2.1 Hz, 1 H), 4.36 (br d, J = 12.1 Hz, 1 H), 4.23−4.11 (m, 3 H). Anal. ($C_{20}H_{15}ClF_3N_3O_5$) C, H, N.

(6*S*)-6-{[3′-Fluoro-4′-(trifluoromethoxy)[1,1′-biphenyl]-4-yl]-methoxy}-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (111). Reaction of alcohol **122** with bromide **152** (0.99 equiv) using procedure C, followed by chromatography of the product on silica gel, eluting with 0−2% EtOAc/CH_2Cl_2 (foreruns) and then 2% EtOAc/CH_2Cl_2, gave **111** (76%) as a cream solid: mp (CH_2Cl_2/pentane) 169−171 °C; ^{1}H NMR ($CDCl_3$) δ 7.54 (dt, J = 8.3, 1.8 Hz, 2 H), 7.43−7.32 (m, 6 H), 4.78 (d, J = 12.0 Hz, 1 H), 4.67 (d, J = 11.9 Hz, 1 H), 4.64 (ddd, J = 12.1, 3.7, 2.1 Hz, 1 H), 4.37 (dd, J = 12.1, 1.3 Hz, 1 H), 4.23−4.12 (m, 3 H). Anal. ($C_{20}H_{15}F_4N_3O_5$) C, H, N.

(6*S*)-6-{[4′-Chloro-3′-(difluoromethoxy)[1,1′-biphenyl]-4-yl]-methoxy}-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (113). A mixture of 4-(methoxycarbonyl)phenylboronic acid (**153**) (1.00 g, 5.56 mmol) and 5-bromo-2-chlorophenol (1.15 g, 5.54 mmol) in aqueous K_2CO_3 (5 mL of 2 M, 10 mmol), EtOH (15 mL), and toluene (25 mL) was purged with N_2. Pd(dppf)Cl_2 (50 mg, 0.068 mmol) was added, and the mixture was refluxed under N_2 for 0.5 h and then partitioned between EtOAc and 0.1 M HCl. Column chromatography of the organic extract (eluting with CH_2Cl_2) gave methyl 4′-chloro-3′-hydroxy[1,1′-biphenyl]-4-carboxylate (**154**) (1.177 g, 81%) as a white solid: mp 162−164 °C; ^{1}H NMR [$(CD)_3SO$] δ 10.36 (br s, 1 H), 8.03 (d, J = 8.5 Hz, 2 H), 7.73 (d, J = 8.5 Hz, 2 H), 7.44 (d, J = 8.3 Hz, 1 H), 7.27 (d, J = 2.2 Hz, 1 H), 7.17 (dd, J = 8.3, 2.2 Hz, 1 H), 3.88 (s, 3 H). APCI MS m/z 263, 265 [M + H]$^+$.

A mixture of **154** (1.119 g, 4.26 mmol), K_2CO_3 (0.735 g, 5.32 mmol), and sodium chlorodifluoroacetate (1.30 g, 8.52 mmol) in anhydrous DMF (10 mL) was stirred at 80 °C under N_2 for 14 h. The mixture was partitioned between EtOAc and water, and the organic fraction was dried and evaporated. Column chromatography (eluting with 1:1 hexanes:CH_2Cl_2) gave methyl 4′-chloro-3′-(difluoromethoxy)[1,1′-biphenyl]-4-carboxylate (**155**) (0.493 g, 37%) as a white solid: mp 80−81 °C; ^{1}H NMR ($CDCl_3$) δ 8.12 (d, J = 8.5 Hz, 2 H), 7.61 (d, J = 8.5 Hz, 2 H), 7.54 (d, J = 8.3 Hz, 1 H), 7.49−7.47 (m, 1 H), 7.43 (dd, J = 8.3, 2.1 Hz, 1 H), 6.60 (t, J_{H-F} = 73.4 Hz, 1 H), 3.95 (s, 3 H). APCI MS m/z 313, 315 [M + H]$^+$.

$LiAlH_4$ (80 mg, 2.11 mmol) was added to a solution of **155** (0.314 g, 1.00 mmol) in Et_2O (20 mL) at 0 °C. The mixture was stirred at room temperature for 2 h, cooled to 0 °C, quenched with ice, and then filtered through Celite. The organic fraction was dried and evaporated, and then column chromatography (eluting with 9:1 CH_2Cl_2:Et_2O) gave [4′-chloro-3′-(difluoromethoxy)[1,1′-biphenyl]-4-yl]methanol (**156**) (0.286 g, 100%) as a white solid: mp 50−51 °C; ^{1}H NMR ($CDCl_3$) δ 7.54 (d, J = 8.3 Hz, 2 H), 7.50 (d, J = 8.3 Hz, 1 H), 7.47−7.42 (m, 3 H), 7.39 (dd, J = 8.3, 2.1 Hz, 1 H), 6.58 (t, J_{H-F} = 73.5 Hz, 1 H), 4.75 (d, J = 5.9 Hz, 2 H), 1.68 (t, J = 5.9 Hz, 1 H). APCI MS m/z 285, 287 $[M + H]^+$.

PBr_3 (45 μL, 0.48 mmol) was added to a solution of **156** (0.271 g, 0.952 mmol) in Et_2O (10 mL) at 0 °C. The mixture was stirred at room temperature for 3 h, cooled to 0 °C, and quenched with ice. The organic layer was washed with aqueous $NaHCO_3$, dried, and evaporated. Column chromatography (eluting with 1:1 hexanes:CH_2Cl_2) gave 4-(bromomethyl)-4′-chloro-3′-(difluoromethoxy)-1,1′-biphenyl (**157**) (0.271 g, 82%) as white flakes, which was used directly in the next step: mp (hexanes) 60−61 °C; ^{1}H NMR ($CDCl_3$) δ 7.54−7.43 (m, 6 H), 7.38 (dd, J = 8.3, 2.1 Hz, 1 H), 6.58 (t, J_{H-F} = 73.5 Hz, 1 H), 4.54 (s, 2 H).

NaH (60%) (20 mg, 0.5 mmol) was added to a solution of **157** (0.120 g, 0.35 mmol) and **122** (0.064 g, 0.35 mmol) in anhydrous DMF (7 mL) at 0 °C. The mixture was stirred at room temperature for 2 h and then quenched with water and partitioned between EtOAc and water. The organic fraction was dried and evaporated, and then column chromatography using a gradient (1:1 hexanes:EtOAc to EtOAc), followed by Et_2O trituration, gave **113** (0.117 g, 75%) as a white solid: mp 159−161 °C; ^{1}H NMR δ 8.02 (s, 1 H), 7.71−7.64 (m, 3 H), 7.62−7.55 (m, 2 H), 7.43 (d, J = 8.2 Hz, 2 H), 7.40 (t, J_{H-F} = 73.3 Hz, 1 H), 4.75−4.64 (m, 3 H), 4.48 (d, J = 11.9 Hz, 1 H), 4.32−4.21 (m, 3 H). Anal. ($C_{20}H_{16}ClF_2N_3O_5$) C, H, N.

Preparation of Substituted Alkoxy Biphenyl Analogues (Scheme 4). 3′-({[(6*S*)-2-Nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*]-[1,3]oxazin-6-yl]oxy}methyl)[1,1′-biphenyl]-3-ol (25). Reaction of iodide **126** and 3-hydroxyphenylboronic acid under the Suzuki conditions described above gave **25** (83%) as a tan solid: mp 80−83 °C; ^{1}H NMR δ 9.48 (s, 1 H), 8.02 (s, 1 H), 7.53−7.49 (m, 2 H), 7.42 (t, J = 7.5 Hz, 1 H), 7.29 (d, J = 7.5 Hz, 1 H), 7.24 (t, J = 7.8 Hz, 1 H), 7.04−6.98 (m, 2 H), 6.77 (ddd, J = 8.1, 2.4, 0.8 Hz, 1 H), 4.75−4.64 (m, 3 H), 4.48 (d, J = 11.9 Hz, 1 H), 4.32−4.21 (m, 3 H). Anal. ($C_{19}H_{17}N_3O_5 \cdot 0.25$EtOAc) C, H, N.

Procedure D. 3-{[3′-({[(6*S*)-2-Nitro-6,7-dihydro-5*H*-imidazo-[2,1-*b*][1,3]oxazin-6-yl]oxy}methyl)[1,1′-biphenyl]-3-yl]oxy}-1-propanol (27). A mixture of phenol **25** (0.737 g, 2.01 mmol), (3-bromopropoxy)(*tert*-butyl)dimethylsilane (0.740 g, 2.92 mmol), and Cs_2CO_3 (3.2 g, 9.8 mmol) in anhydrous DMF (20 mL) was stirred at 90 °C for 2 h. The resulting mixture was partitioned between EtOAc and water, the organic fraction was dried, and the solvent was removed under reduced pressure to give an oil, which was dissolved in THF (100 mL) and treated with TBAF (1 M in THF, 10 mL). The solution was stirred at room temperature for 2 h, then the solvent was removed, and the residue was partitioned between EtOAc and water. The organic portion was washed with water, dried, and evaporated, and the resulting oil was purified by column chromatography (eluting with EtOAc) to give **27** (0.477 g, 56%) as a gum: ^{1}H NMR δ 8.01 (s, 1 H), 7.60−7.55 (m, 2 H), 7.43 (t, J = 8.2 Hz, 1 H), 7.37−7.29 (m, 2 H), 7.20−7.13 (m, 2 H), 6.93 (ddd, J = 8.2, 2.5, 0.7 Hz, 1 H), 4.75−4.63 (m, 3 H), 4.52 (t, J = 5.2 Hz, 1 H), 4.47 (d, J = 11.7 Hz, 1 H), 4.33−4.20 (m, 3 H), 4.10 (t, J = 6.4 Hz, 2 H), 3.58 (td, J = 5.9, 5.2 Hz, 2 H), 1.89 (tt, J = 6.4, 5.9 Hz, 2 H); HRFABMS calcd for $C_{22}H_{24}N_3O_6$ m/z $[M + H]^+$ 426.1665, found 426.1673. HPLC purity: 93.2%.

Procedure E. (6*S*)-6-{[3′-(3-Iodopropoxy)[1,1′-biphenyl]-3-yl]-methoxy}-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (158). Mesyl chloride (0.16 mL, 2.04 mmol) was added to a solution of alcohol **27** (0.436 g, 1.02 mmol) and Et_3N (0.43 mL, 3.09 mmol) in THF (40 mL) at 0 °C. The mixture was stirred at 0 °C for 1 h, then diluted with EtOAc, washed with saturated aqueous $NaHCO_3$, and dried, and the solvent was removed to give a crude mesylate. This mesylate was then refluxed with NaI (1.54 g, 10.3 mmol) in Me_2CO (100 mL) for 3 h. The solvent was removed under reduced pressure, and the residue was partitioned between EtOAc and water. The organic portion was dried and evaporated, and the residue was chromatographed (eluting with EtOAc) to give **158** (0.426 g, 78%) as a pale yellow glass, which was used directly in the next step: ^{1}H NMR δ 8.01 (s, 1 H), 7.61−7.57 (m, 2 H), 7.43 (t, J = 7.8 Hz, 1 H), 7.36 (t, J = 7.9 Hz, 1 H), 7.32 (d, J = 7.6 Hz, 1 H), 7.23−7.16 (m, 2 H), 6.96 (dd, J = 8.1, 4.9 Hz, 1 H), 4.76−4.65 (m, 3 H), 4.48 (d, J = 11.8 Hz, 1 H), 4.32−4.21 (m, 3 H), 4.10 (t, J = 6.0 Hz, 2 H), 3.42 (t, J = 6.9 Hz, 2 H), 2.23 (tt, J = 6.9, 6.0 Hz, 2 H). APCI MS m/z 536 $[M + H]^+$.

Procedure F. (6*S*)-6-({3′-[3-(4-Morpholinyl)propoxy][1,1′-biphenyl]-3-yl}methoxy)-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*]-[1,3]oxazine (28). A solution of iodide **158** (85 mg, 0.16 mmol) and morpholine (70 μL, 0.80 mmol) in DMA (3 mL) was stirred at room temperature for 16 h. The resulting mixture was partitioned (EtOAc/water), the organic fraction was dried, and then removal of the solvent gave a solid, which was triturated in Et_2O to give **28** (76 mg, 97%) as a pale yellow solid: mp 92−95 °C; ^{1}H NMR δ 8.01 (s, 1 H), 7.60−7.55 (m, 2 H), 7.43 (br t, J = 7.7 Hz, 1 H), 7.37−7.29 (m, 2 H), 7.17 (br d, J = 7.7 Hz, 1 H), 7.16 (t, J = 2.2 Hz, 1 H), 7.14 (ddd, J = 8.2, 2.5, 0.7 Hz, 1 H), 4.75−4.64 (m, 3 H), 4.48 (d, J = 11.8 Hz, 1 H), 4.32−4.20 (m, 3 H), 4.08 (t, J = 6.3 Hz, 2 H), 3.57 (t, J = 4.6 Hz, 4 H), 2.44 (t, J = 7.1 Hz, 2 H), 2.41−2.33 (m, 4 H), 1.89 (tt, J = 7.1, 6.3 Hz, 2 H). Anal. ($C_{26}H_{30}N_4O_6 \cdot 0.5H_2O$) C, H, N.

3-{[3′-({[(6*S*)-2-Nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazin-6-yl]oxy}methyl)[1,1′-biphenyl]-4-yl]oxy}-1-propanol (37). Reaction of phenol **34** (prepared by Suzuki coupling; see Supporting Information) with (3-bromopropoxy)(*tert*-butyl)dimethylsilane and Cs_2CO_3 in DMF, followed by desilylation with TBAF, using procedure D, gave **37** (51%) as a white solid: mp 160−162 °C; ^{1}H NMR δ 8.02 (s, 1 H), 7.57−7.49 (m, 4 H), 7.40 (t, J = 7.6 Hz, 1 H), 7.25 (br d, J = 7.6 Hz, 1 H), 7.01 (d, J = 8.8 Hz, 2 H), 4.74−4.63 (m, 3 H), 4.53 (t, J = 5.2 Hz, 1 H), 4.48 (d, J = 11.7 Hz, 1 H), 4.32−4.19 (m, 3 H), 4.08 (t, J = 6.2 Hz, 2 H), 3.57 (td, J = 5.9, 5.2 Hz, 2 H), 1.88 (tt, J = 6.2, 5.9 Hz, 2 H). Anal. ($C_{22}H_{23}N_3O_6$) C, H, N.

(6*S*)-6-{[4′-(3-Iodopropoxy)[1,1′-biphenyl]-3-yl]methoxy}-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (159). Mesylation of alcohol **37**, followed by reaction with NaI in Me_2CO, using procedure E, gave **159** (84%) as a glass, which was used directly in the next step: ^{1}H NMR δ 8.02 (s, 1 H), 7.58−7.50 (m, 4 H), 7.40 (t, J = 7.6 Hz, 1 H), 7.25 (br d, J = 7.6 Hz, 1 H), 7.02 (d, J = 8.8 Hz, 2 H), 4.73−4.62 (m, 3 H), 4.48 (d, J = 11.7 Hz, 1 H), 4.33−4.20 (m, 3 H), 4.07 (t, J = 6.0 Hz, 2 H), 3.41 (t, J = 6.8 Hz, 2 H), 2.22 (tt, J = 6.8, 6.0 Hz, 2 H). APCI MS m/z 536 $[M + H]^+$.

(6*S*)-6-({4′-[3-(4-Morpholinyl)propoxy][1,1′-biphenyl]-3-yl}-methoxy)-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (38). Reaction of iodide **159** with morpholine in DMA using procedure F gave **38** (91%) as a white solid: mp 203−206 °C; ^{1}H NMR δ 8.02 (s, 1 H), 7.56−7.49 (m, 4 H), 7.40 (t, J = 7.6 Hz, 1 H), 7.24 (br d, J = 7.6 Hz, 1 H), 6.99 (d, J = 8.8 Hz, 2 H), 4.73−4.63 (m, 3 H), 4.48 (d, J = 11.8 Hz, 1 H), 4.32−4.20 (m, 3 H), 4.05 (t, J = 6.4 Hz, 2 H), 3.58 (t, J = 4.6 Hz, 4 H), 2.43 (t, J = 7.3 Hz, 2 H), 2.40−2.33 (m, 4 H), 1.89 (tt, J = 7.3, 6.4 Hz, 2 H). Anal. ($C_{26}H_{30}N_4O_6$) C, H, N.

3-{[4′-({[(6*S*)-2-Nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazin-6-yl]oxy}methyl)[1,1′-biphenyl]-2-yl]oxy}-1-propanol (50). Reaction of phenol **47** (prepared by Suzuki coupling; see Supporting Information) with (3-bromopropoxy)(*tert*-butyl)dimethylsilane and Cs_2CO_3 in DMF, followed by desilylation with TBAF (2.4 equiv), using procedure D, gave **50** (52%) as a gum: ^{1}H NMR δ 8.03 (s, 1 H), 7.48 (d, J = 8.3 Hz, 2 H), 7.33 (d, J = 8.3 Hz, 2 H),

7.31−7.25 (m, 2 H), 7.09 (br d, J = 7.6 Hz, 1 H), 7.00 (td, J = 7.4, 1.0 Hz, 1 H), 4.72−4.63 (m, 3 H), 4.49 (d, J = 11.9 Hz, 1 H), 4.45 (t, J = 5.2 Hz, 1 H), 4.33−4.21 (m, 3 H), 4.04 (t, J = 6.2 Hz, 2 H), 3.48 (td, J = 5.9, 5.2 Hz, 2 H), 1.78 (tt, J = 6.2, 5.9 Hz, 2 H); HRFABMS calcd for $C_{22}H_{24}N_3O_6$ m/z $[M + H]^+$ 426.1665, found 426.1659. HPLC purity: 95.5%.

2-{[4′-({[(6*S*)-2-Nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazin-6-yl]oxy}methyl)[1,1′-biphenyl]-3-yl]oxy}ethanol (64). A mixture of phenol **62** (prepared by Suzuki coupling; see Supporting Information) (0.440 g, 1.20 mmol), 2-(2-bromoethoxy)tetrahydro-2*H*-pyran (0.500 g, 2.39 mmol), and Cs_2CO_3 (1.95 g, 5.98 mmol) in anhydrous DMF (10 mL) was stirred at 90 °C for 2 h. The resulting mixture was partitioned between EtOAc and water, the organic fraction was evaporated, and the residue was chromatographed (eluting with EtOAc) to give a crude THP ether. This ether was dissolved in MeOH (100 mL), treated with MsOH (5 drops), and then stirred at room temperature for 1 h. The resulting solution was filtered through a plug of $NaHCO_3$, and the solvent was removed under reduced pressure. Chromatography of the residue (eluting with EtOAc) gave **64** (0.400 g, 67%) as a pale yellow glass: ^{1}H NMR δ 8.03 (s, 1 H), 7.63 (d, J = 8.3 Hz, 2 H), 7.38 (d, J = 8.3 Hz, 2 H), 7.36 (t, J = 8.0 Hz, 1 H), 7.20 (br d, J = 7.7 Hz, 1 H), 7.17 (t, J = 2.2 Hz, 1 H), 6.93 (ddd, J = 8.2, 2.5, 0.7 Hz, 1 H), 4.83 (t, J = 5.5 Hz, 1 H), 4.73−4.63 (m, 3 H), 4.48 (d, J = 11.9 Hz, 1 H), 4.33−4.21 (m, 3 H), 4.06 (t, J = 4.9 Hz, 2 H), 3.74 (dt, J = 5.5, 4.9 Hz, 2 H); HRFABMS calcd for $C_{21}H_{22}N_3O_6$ m/z $[M + H]^+$ 412.1509, found 412.1514. HPLC purity: 96.8%.

3-{[4′-({[(6*S*)-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazin-6-yl]oxy}methyl)[1,1′-biphenyl]-3-yl]oxy}-1-propanol (65). Reaction of phenol **62** with (3-bromopropoxy)(*tert*-butyl)dimethylsilane (2.5 equiv) and Cs_2CO_3 in DMF for 1 h, followed by desilylation with TBAF (2.1 equiv), using procedure D, and chromatography of the product on silica gel, eluting with a gradient of 1:1 hexanes:EtOAc to EtOAc, gave **65** (66%) as an oil: ^{1}H NMR δ 8.03 (s, 1 H), 7.64 (d, J = 8.3 Hz, 2 H), 7.39 (d, J = 8.3 Hz, 2 H), 7.35 (dd, J = 8.2, 7.9 Hz, 1 H), 7.19 (br d, J = 7.9 Hz, 1 H), 7.16 (t, J = 2.2 Hz, 1 H), 6.92 (ddd, J = 8.2, 2.2, 0.7 Hz, 1 H), 4.73−4.64 (m, 3 H), 4.52 (t, J = 5.2 Hz, 1 H), 4.48 (d, J = 11.9 Hz, 1 H), 4.32−4.21 (m, 3 H), 4.10 (t, J = 6.4 Hz, 2 H), 3.57 (td, J = 5.9, 5.2 Hz, 2 H), 1.88 (tt, J = 6.4, 5.9 Hz, 2 H); HRFABMS calcd for $C_{22}H_{24}N_3O_6$ m/z $[M + H]^+$ 426.1665, found 426.1659. HPLC purity: 95.7%.

(6*S*)-6-{[3′-(2-Iodoethoxy)[1,1′-biphenyl]-4-yl]methoxy}-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (160). Mesylation of alcohol **64**, followed by reaction with NaI in Me_2CO, using procedure E, gave **160** (80%) as a foam, which was used directly in the next step: ^{1}H NMR δ 8.03 (s, 1 H), 7.65 (d, J = 8.3 Hz, 2 H), 7.42−7.35 (m, 3 H), 7.24 (br d, J = 8.3 Hz, 1 H), 7.18 (m, 1 H), 6.95 (ddd, J = 8.2, 2.5, 0.7 Hz, 1 H), 4.72−4.65 (m, 3 H), 4.48 (d, J = 11.9 Hz, 1 H), 4.34 (t, J = 6.1 Hz, 2 H), 4.32−4.21 (m, 3 H), 3.54 (t, J = 6.1 Hz, 2 H). APCI MS m/z 522 $[M + H]^+$.

***N*,*N*-Dimethyl-2-{[4′-({[(6*S*)-2-nitro-6,7-dihydro-5*H*-imidazo-[2,1-*b*][1,3]oxazin-6-yl]oxy}methyl)[1,1′-biphenyl]-3-yl]oxy}ethanamine (66).** A solution of iodide **160** (0.105 g, 0.202 mmol) and dimethylamine (~0.1 mL of a 40% aqueous solution) in DMA (2 mL) was stirred at room temperature for 16 h in a sealed tube. The resulting mixture was partitioned (EtOAc/water), the organic fraction was dried, and then removal of the solvent gave a solid, which was triturated in Et_2O to give **66** (58 mg, 65%) as a white solid: mp 93−96 °C; ^{1}H NMR δ 8.03 (s, 1 H), 7.64 (d, J = 8.3 Hz, 2 H), 7.39 (d, J = 8.3 Hz, 2 H), 7.35 (t, J = 8.0 Hz, 1 H), 7.19 (br d, J = 7.7 Hz, 1 H), 7.17 (t, J = 2.2 Hz, 1 H), 6.93 (ddd, J = 8.2, 2.5, 0.7 Hz, 1 H), 4.73−4.64 (m, 3 H), 4.48 (d, J = 11.9 Hz, 1 H), 4.32−4.21 (m, 3 H), 4.11 (t, J = 5.8 Hz, 2 H), 2.64 (t, J = 5.8 Hz, 2 H), 2.22 (s, 6 H). Anal. ($C_{23}H_{26}N_4O_5 \cdot 0.5H_2O$) C, H, N.

2-{[4′-({[(6*S*)-2-Nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazin-6-yl]oxy}methyl)[1,1′-biphenyl]-4-yl]oxy}ethanol (89). A mixture of phenol **85** (prepared by Suzuki coupling; see Supporting Information) (0.600 g, 1.20 mmol), 2-(2-bromoethoxy)tetrahydro-2*H*-pyran (0.68 g, 3.25 mmol), and Cs_2CO_3 (2.66 g, 8.16 mmol) in DMF (15 mL) was stirred at 90 °C for 2 h. The resulting mixture was partitioned (EtOAc/water), the organic fraction was evaporated, and the residue was chromatographed (eluting with EtOAc) to give a crude THP ether (0.649 g, 80%). This ether (0.573 g) was dissolved in MeOH (100 mL), treated with MsOH (10 drops), and then stirred at room temperature for 1 h. The resulting solution was filtered through a plug of $NaHCO_3$, and the solvent was removed under reduced pressure. Chromatography of the residue (eluting with EtOAc) gave **89** (0.410 g, 86%) as a white solid: mp 209−212 °C; ^{1}H NMR δ 8.02 (s, 1 H), 7.61−7.55 (m, 4 H), 7.36 (d, J = 8.3 Hz, 2 H), 7.01 (d, J = 8.8 Hz, 2 H), 4.85 (t, J = 5.5 Hz, 1 H), 4.71−4.62 (m, 3 H), 4.48 (d, J = 11.9 Hz, 1 H), 4.32−4.21 (m, 3 H), 4.02 (t, J = 4.9 Hz, 2 H), 3.73 (dt, J = 5.5, 4.9 Hz, 2 H). Anal. ($C_{21}H_{21}N_3O_6$) C, H, N.

3-{[4′-({[(6*S*)-2-Nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazin-6-yl]oxy}methyl)[1,1′-biphenyl]-4-yl]oxy}-1-propanol (90). Reaction of phenol **85** with (3-bromopropoxy)(*tert*-butyl)dimethylsilane (2.0 equiv) and Cs_2CO_3 in DMF for 1 h, followed by desilylation with TBAF (2.0 equiv), using procedure D, gave **90** (61%) as a white solid: mp 206−209 °C; ^{1}H NMR δ 8.02 (s, 1 H), 7.61−7.55 (m, 4 H), 7.36 (d, J = 8.3 Hz, 2 H), 7.00 (d, J = 8.8 Hz, 2 H), 4.71−4.62 (m, 3 H), 4.52 (t, J = 5.2 Hz, 1 H), 4.47 (d, J = 11.9 Hz, 1 H), 4.31−4.21 (m, 3 H), 4.07 (t, J = 6.4 Hz, 2 H), 3.56 (td, J = 5.9, 5.2 Hz, 2 H), 1.87 (tt, J = 6.4, 5.9 Hz, 2 H). Anal. ($C_{22}H_{23}N_3O_6$) C, H, N.

(6*S*)-6-{[4′-(3-Iodopropoxy)[1,1′-biphenyl]-4-yl]methoxy}-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (161). Mesylation of alcohol **90** for 2.5 h, followed by reaction with NaI in Me_2CO for 1 h, using procedure E, gave **161** (75%) as a white solid: mp 146−148 °C; ^{1}H NMR δ 8.02 (s, 1 H), 7.61−7.55 (m, 4 H), 7.36 (d, J = 8.3 Hz, 2 H), 7.03 (d, J = 8.8 Hz, 2 H), 4.70−4.62 (m, 3 H), 4.47 (d, J = 11.9 Hz, 1 H), 4.30−4.20 (m, 3 H), 4.07 (t, J = 6.0 Hz, 2 H), 3.40 (t, J = 6.8 Hz, 2 H), 2.21 (tt, J = 6.8, 6.0 Hz, 2 H). APCI MS m/z 536 $[M + H]^+$.

(6*S*)-6-({4′-[3-(4-Morpholinyl)propoxy][1,1′-biphenyl]-4-yl}-methoxy)-2-nitro-6,7-dihydro-5*H*-imidazo[2,1-*b*][1,3]oxazine (91). Reaction of iodide **161** with morpholine using procedure F gave **91** (89%) as a white solid: mp 221−224 °C; ^{1}H NMR δ 8.02 (s, 1 H), 7.60−7.54 (m, 4 H), 7.36 (d, J = 8.3 Hz, 2 H), 7.00 (d, J = 8.8 Hz, 2 H), 4.70−4.62 (m, 3 H), 4.47 (d, J = 11.9 Hz, 1 H), 4.32−4.20 (m, 3 H), 4.05 (t, J = 6.4 Hz, 2 H), 3.57 (t, J = 4.6 Hz, 4 H), 2.42 (t, J = 7.3 Hz, 2 H), 2.40−2.33 (m, 4 H), 1.88 (tt, J = 7.3, 6.4 Hz, 2 H). Anal. ($C_{26}H_{30}N_4O_6 \cdot 0.25H_2O$) C, H, N.

Biology. MABA and LORA Tuberculosis Assays. These were carried out according to the published protocols.[21−23]

Microsomal Stability Assays. Compounds (1 μM) were incubated with pooled human or CD-1 mouse liver microsome preparations (0.5 mg/mL final protein concentration) and an NADPH regenerating system ($MgCl_2$, 3.3 mM; G6P, 3.3 mM; G6PD, 0.4 unit/mL; $NADP^+$, 1.3 mM) in phosphate buffer (75 mM, pH 7.4), with a final volume of 200 μL. Compounds were dissolved in DMSO such that the final DMSO concentration was 0.5%. Positive controls (warfarin, propranolol, and testosterone, incubated as a cocktail) were treated similarly. Incubation was at 37 °C in a 96-well plate in a humidified incubator, and reactions were stopped at 0 and 60 min by the addition of MeCN (100 μL) containing metoprolol (0.2 μM) as an internal standard. Samples were diluted 10× and centrifuged prior to analysis by LC-MS/MS using electrospray ionization and SRM monitoring using a gradient LC method (isocratic for 0.5 min, followed by a 5% to 91% MeCN linear gradient in water containing 0.1% HCOOH over 1 min, with a 2.5 min hold at 91% MeCN; total run time 6.5 min). An XDB-C18 2.1 × 50 mm column (Agilent) was employed, with an eluent flow rate of 0.5 mL/min. LC peak areas were integrated and expressed as analyte/IS peak area ratios (PAR), and a mean value for each

time point was calculated from the duplicates. The percent remaining value was calculated as

$$\% \text{ remaining} = 100(\text{mean PAR}_{T60}/\text{mean PAR}_{T0})$$

The assays were conducted by MDS Pharma Services, 22011 30th Drive SE, Bothell, WA 98021.

***In Vivo* Acute TB Infection Assay.** BALB/c mice were infected via aerosol with a suspension of $\sim 2 \times 10^6$ colony forming units (CFU) of *M. tuberculosis* Erdman/mL.[23] Each compound was given orally to a group of seven or eight mice at 100 mg/kg daily for 5 days a week for 3 weeks, beginning on day 11 postinfection. Compounds were administered as a suspension in 0.5% CMC/0.08% Tween 80 in water. Mice were sacrificed on day 31, and the numbers of CFU in the lungs were determined and compared with the CFU for vehicle alone-treated mice at this time.[23] The parent drug **1** was employed as a positive control in each experiment, and the results are recorded as the ratio of the average reduction in CFU in the compound-treated mice/the average CFU reduction in the mice treated with **1**. In this assay, **1** itself caused up to 2.5−3 log reductions in CFU.

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Supporting Information Available: Additional experimental procedures and characterizations for compounds in Tables 1−3; CFU data for compounds in Table 5; combustion analytical data. This material is available free of charge via the Internet at http://pubs.acs.org.

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