Benzimidazole-5-sulfonamides as Novel Nonpeptide Luteinizing Hormone Releasing Hormone (LHRH) Antagonists: Minimization of Mechanism-Based CYP3A4 Inhibition

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Herein we report the development of novel, potent and non-peptide luteinizing hormone releasing hormone (LHRH) antagonists. The optimization towards derivatives free from mechanism-based CYP3A4 inhibition is described. The identification of a main metabolite guided us towards structural modifications of the benzyl moiety, which resulted in significant improvements of the CYP3A4 profile, while maintaining potent LHRH antagonist activity.

Key words luteinizing hormone releasing hormone (LHRH); antagonist; irreversible CYP3A4 inhibition; mechanism-based; small molecule

Part of the effort to reduce attrition rates in drug discovery and development is the early detection of CYP inhibition.[1] Irreversible (mechanism-based) CYP inhibitors inhibit CYP activity through chemical modifications of the enzyme, for instance through reactive metabolites.[2—4] In these cases, enzyme activity cannot be recovered by the removal of inhibitors. Potentially leading to liver toxicity, these compounds are unfavorable drug candidates and often avoided in drug discovery and development. It is crucial to identify this undesired property at early stages in order to improve this parameter during lead optimisation.[5,6]

In previous communications we reported the discovery of potent non-peptide luteinizing hormone-releasing hormone (LHRH) antagonists **1** and **2** (Fig. 1) as possible therapeutics for the treatment of sex hormone-dependent disease states.[7,8] However, both compounds are plagued by time-dependent CYP3A4 inhibition that we wished to address early in the optimization process.

In this report we would like to disclose SAR studies that led to the discovery of derivatives with optimized CYP3A4 profile and maintained potency.

Chemistry

Benzimidazoles **7**, **10**, **11**, **14**—**18**, and **22**, **23** were prepared with benzylamines and *tert*-butyl isocyanates using methods described in previous communication.[8] The intermediates **7** were subjected to Sandmeyer reactions[9] in the presence of SO_2/HCl to prepare the sulfonylchloride derivatives **8** which were reacted with various substituted benzylamines to prepare the corresponding sulfonamides **12**, **13** and **19**—**21**.

Results and Discussion

Compounds **1** and **2** both show increased CYP3A4 inhibition after incrementing preincubation times (Fig. 1). This is also reflected by their strong inactivation constant (*e.g.* **2**, $k_{inact}=0.074\,min^{-1}$). To rule out the possibility of mere product inhibition, we diluted the microsomal assay system 50 fold after preincubation. However, **2**'s CYP3A4 kinetics remained unchanged, also after dilution ($k_{inact}=0.067\,min^{-1}$). These findings confirmed the suspicion that our compounds would irreversibly inhibit CYP3A4, possibly *via* a reactive or chelating metabolite.[10]

To gain insight into the metabolic fate of our compounds

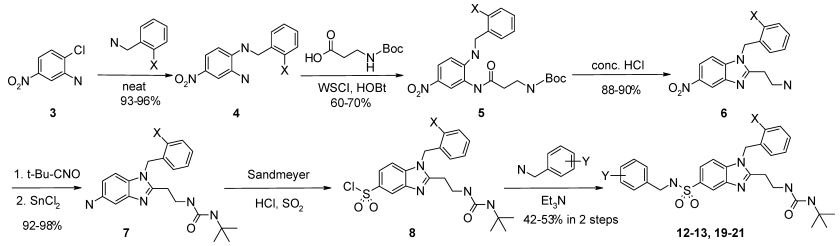
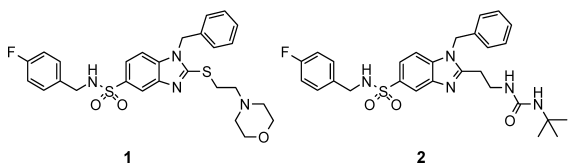


Chart 1. Synthesis of Benzimidazole Sulfonamide as LHRH Antagonists



	h-lhrh: IC_{50} (nM)	CYP3A4: IC_{50} (μM) -Preincubation time-	
		0 min.	30 min.
1	160	>30	4.4
2	20	>30	8.6

Fig. 1. CYP3A4 Inhibitory Activity of Compounds **1** and **2**

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we incubated compound **9** with human microsomes and identified the degradation products P-1 (main) and P-2 (trace, Fig. 2). This result suggested that the 1-benzyl group of the benzimidazole core is the major interaction and cleavage site.

In order to separate LHRH antagonism and irreversible CYP inhibition, we measured the CYP3A4 inhibition constant of our benzimidazole derivatives at two time points (0, 30 min after preincubation) along with their LHRH antagonistic activity (Table 1).[11] All IC_{50}'s are means of at least two experiments each run in triplicates. Modification of **2**'s *tert*-butyl urea part by changing the alkyl group to *iso*-propyl (**10**) or 2-fluorophenyl (**11**) did not reduce mechanism-based CYP3A4 inhibition. Changing the substituent of benzylsulfonamide from 4-F to 3,4-F (**12**) or to 4-Cl (**13**) did not have any effects on time-dependent CYP3A4 inhibition either. We realized, however, that the weakly active *N*-debenzylated compound **23** was 'clean' in terms of time-dependent

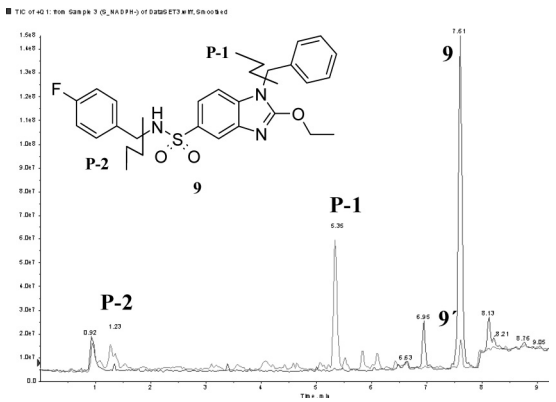
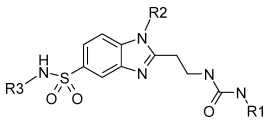


Fig. 2. Metabolite Identification of Compound **9**

Table 1. Functional Activity and CYP3A4 Inhibitory Effect of Compounds **10**—**23**



Compd.	R1	R2	R3	h-lhrh IC_{50} (nM)	CYP3A4 IC_{50} (μM)	
					Preincubation time 0 min	30 min
10	*i*-Pr			150	>30	5.5
11	2-F-Ph			66	23	11
12	*t*-Bu			170	>30	8.1
13	*t*-Bu			4.2	25	7.0
14	*t*-Bu			19	>30	15
15	*t*-Bu			9.0	>30	24
16	*t*-Bu			30	>30	7.2
17	*t*-Bu			26	>30	13
18	*t*-Bu			9.7	>30	27
19	*t*-Bu			27	>30	>30
20	*t*-Bu			10	>30	28
21	*t*-Bu			34	>30	>30
22	*t*-Bu			17	27	25
23	*t*-Bu	—		4500	>30	>30

CYP3A4 inhibition. It was thus tempting to speculate that the *N*-benzyl moiety of our leads could be the structural feature responsible for irreversible CYP3A4 inhibition. Therefore we focused our optimization strategy on modifying this molecular fragment.

Whereas the introduction of 2-F (**14**) only showed marginal improvement, the corresponding 2-Cl derivative (**15**) showed much better properties, inhibiting CYP3A4 only weakly after 30 min of preincubation (IC_{50}=24 μM). This effect was confirmed after introduction of an additional substituent in 4-position (**18**, IC_{50}=27 μM after 30 min). The 3- and 4-chloro-derivatives **16** and **17** however, were still potent time-dependent CYP3A4 inhibitors, suggesting the importance of a large substituent in *ortho* position. Indeed, keeping the 2-Cl substituent in the molecular structure led to the identification of compounds **19**—**21** with IC_{50} of 10—27 nM and no CYP3A4 inhibition activity up to 30 μM (**19**, **21**).[12] Confirming this hypothesis, the incorporation of steric bias in the adjacent methylene bridge also provided an LHRH inhibitor free from CYP3A4 interaction (**22**).

In summary, we have discovered a novel class of benzimidazole LHRH receptor antagonists **19**—**22**, virtually free of time-dependent CYP3A4 inhibition. We have devised structure activity relationships indicating the *N*-benzyl group to be involved in this unwanted side effect. The structural conclusions drawn for avoiding irreversible CYP3A4 inhibition might be of value for the optimization of similar structural frameworks.

Experimental

General Methods Melting points (mp) were determined using a BÜCHI B-545 melting point apparatus and were uncorrected. The ^{1}H-NMR spectra were recorded on a Bruker DRX-300 (300 MHz for ^{1}H) spectrometer, Bruker 500 UltraShield™ (500 MHz for ^{1}H) spectrometer in $CDCl_3$, dimethyl sulfoxide-d_6 (DMSO-d_6) or CD_3OD using tetramethylsilane (TMS) as an internal standard. High-resolution mass spectra (HR-MS) were obtained using a Finnigan MAT 95. Liquid Chromatography-Mass spectroscopy (LC-MS) was measured using the Micromass Platform LC with a Shimadzu Phenomenex ODS column (4.6 mmϕ×30 mm), flushing a mixture of acetonitrile–water (9 : 1 to 1 : 9) at 1 ml/min flow rate. High performance liquid chromatographies (HPLC) were conducted by using Shimadzu LC-10A.

Measurement of LHRH Antagonistic Activity Intracelluar Ca mobilization data were obtained using recombinant CHO cells transfected with human LHRH receptor cDNA. The cells were pre-incubated with variable concentrations of test compounds for 20 min at 25 °C. Fluorescence changes indicating mobilization of cytoplasmic calcium (Flu-3 AM, Molecular Probes) were measured on a FDSS-3000 machine (Hamamatsu photonics) after stimulation with 10 nM LHRH. The inhibitory effect of the compounds was calculated by comparing the integral ratio of actual and intial fluorescence.

Metabolite Identification Study The study was carried out with 1-benzyl-2-ethoxy-1*H*-benzimidazole-5-sulfonamide **9** as a typical structural representative of our series. One hour after microsomal incubation the reaction was quenched with ether and the organic layer was analyzed by HPLC [Symmetry® C_{18}, 4.6 mmϕ×100 mm; acetonitrile–aqueous ammonium acetate (pH=6.8) 3 : 7 to 8 : 2 at 1 ml/min] showing two non-base line separated peaks [P-1 (72%); t_R=5.35 min, *m/z* (+1)=350.4, P-2 (17%); t_R=0.92 min, *m/z* (+1)=242.3], indicating a mixture of debenzylated and 4-F debenzylated metabolites with remaining starting material **9′** (11%).

Measurement of IC_{50} against CYP3A4 The incubation mixture [total amount: 0.2 ml, 0.2 mg/ml of microsomes, 1 mM EDTA, NADPH-generating system (25 mM glucose-6-phosphate, 2.5 mM $NADP^+$, and 2 units of glucose-6-phosphate dehydrogenase in 100 mM potassium phosphate buffer)], was pre-incubated with 0.1, 0.3, 1.0, 3.0, 10 and 30 μM of test compounds at 37 °C for 0 and 30 min. The CYP3A4 activity was assayed after initiation with testosterone (250 μM). The reaction was quenched with EtOAc, the organic layer was evaporated, and the residues were subjected to HPLC analysis [Symmetry® C_{18}, 4.6 mmϕ×100 mm, water–MeOH–THF (66 : 24 : 10) at 1 ml/min]. The time dependency of 6β-hydroxytestosterone concentrations was measured with and without test compounds. The IC_{50} value was calculated by linear interpolation. The initial-rate constant for inactivation (k, min^{-1}) was estimated from the slope of the log-linear regression line of residual CYP3A4 activity *vs.* the preincubation time profile. K_i and k_{inact} were determined using Eq. 1, where I is the test compound concentration, k_{inact} is the maximum rate constant inactivation, and K_i is the test compound concentration that produces half the maximal rate of inactivation.

$$k = \frac{k_{inact} \times I}{K_i + I} \quad (1)$$

1-(2-Chlorobenzyl)-2-[2-(3-*tert*-butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(3,4-difluorobenzyl)sulfonamide (19) A solution of *N*-(*tert*-butyl)-*N′*-(2-{1-[(2-chlorophenyl)amino]-5-amino-1*H*-benzimidazol-2-yl}ethyl)urea (**7**: X=Cl, 1.2 g, 2.9 mmol) was suspended in conc. HCl/HOAc and converted to a diazonium salt by adding a solution of $NaNO_2$/water (216 mg, 3.14 mmol) portion wise (1 h) at 0 °C. In a different flask, $CuCl_2$ (291 mg, 1.71 mmol) was added to SO_2-saturated acetic acid and stirred under continous bubbling of SO_2 for 20 min. The solution of the diazonium salt was added to the $CuCl_2$–SO_2 mixture at 0 °C and stirred for 30 min. After further stirring at room temperature for 30 min, the mixture was quenched with 10 ml of ice water. The whole mixture was extracted 3 times with EtOAc, and the organic layer was washed with water, saturated $NaHCO_3$ aq. and brine. The organic solvent was removed *in vacuo* to give the crude sulfonyl chloride **8** as a brownish precipitate (81%). LC-MS *m/z*: 483 (M^+), 421, 384.

The sulfonylchloride **8** (40 mg, 0.07 mmol) was reacted with 3,4-difluorobenzylamine (12.3 mg, 0.09 mmol) in CH_2Cl_2 with triethylamine for 2 h at room temperature. After removing the solvent, the residue was purified through preparative thin layer chromatography (CH_2Cl_2/MeOH=12/1) to give the desired 1-(2-chlorobenzyl-2-[2-(3-*tert*-butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(3,4-difluorobenzyl)sulfonamide **19** (32 mg, 73% yield) as a white powder. ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 1.62 (9H, s), 2.96 (2H, t, *J*=5.8 Hz), 3.71 (2H, q, *J*=5.8 Hz), 4.12 (2H, d, *J*=6.3 Hz), 4.58 (1H, s), 5.32 (1H, t, *J*=6.0 Hz), 5.49 (2H, s), 6.50 (2H, m), 7.00 (2H, m), 7.15 (1H, t, *J*=7.6 Hz), 7.27 (3H, m), 7.46 (1H, dd, *J*=7.9, 1.2 Hz), 7.72 (1H, dd, *J*=8.5, 1.7 Hz), 8.18 (1H, d, *J*=1.4 Hz). LC-MS *m/z*: 590 (M^+), 517, 491. HR-MS *m/z*: 590.1821 (M^+) (Calcd for $C_{28}H_{30}ClF_2N_5O_3S$: 590.1799). mp 157—158 °C.

1-Benzyl-2-[2-(3-*iso*-propyl-ureido)-ethyl]-1*H*-benzimidazole-5-(4-fluorobenzyl)sulfonamide (10) ^{1}H-NMR (500 MHz, DMSO) δ: 0.98 (3H, s), 0.99 (3H, s), 2.98 (2H, t, *J*=6.6 Hz), 3.48 (2H, q, *J*=6.6 Hz), 3.63 (1H, q, *J*=6.6 Hz), 3.93 (2H, s), 5.56 (2H, s), 5.90 (2H, m), 7.11 (4H, m), 7.29 (4H, m), 7.64 (2H, dd, *J*=24.0, 8.5 Hz), 8.02 (1H, s), 8.06 (1H, br s). LC-MS *m/z*: 524 (M^+), 465, 196. HR-MS *m/z*: 524.2123 (M^+) (Calcd for $C_{27}H_{30}FN_5O_3S$: 524.2127). mp 124—125 °C.

1-Benzyl-2-[2-(2-fluoro-phenyl)-ethyl]-1*H*-benzimidazole-5-(4-fluorobenzyl)sulfonamide (11) ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 1.57 (9H, s), 3.12 (2H, t, *J*=6.1 Hz), 3.98 (4H, m), 5.08 (1H, t, *J*=6.5 Hz), 5.43 (2H, s), 6.84 (4H, m), 7.03 (4H, m), 7.31 (4H, m), 7.55 (2H, m), 7.81 (1H, d, *J*=1.5 Hz), 8.11 (1H, t, *J*=8.5 Hz). LC-MS *m/z*: 575 (M^+), 465, 439. HR-MS *m/z*: 576.1881 (M^+) (Calcd for $C_{30}H_{27}F_2N_5O_3S$: 576.1876). mp 152—153 °C.

1-Benzyl-2-[2-(3-*tert*-butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(3,4-difluorobenzyl)sulfonamide (12) ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 1.20 (9H, s), 2.99 (2H, br s), 3.70 (2H, q, *J*=5.7 Hz), 4.09 (2H, d, *J*=6.0 Hz), 4.72 (1H, s), 5.40 (3H, m), 7.00 (5H, m), 7.33 (4H, m), 7.71 (1H, dd, *J*=8.5, 1.6 Hz), 8.12 (1H, d, *J*=1.4 Hz). LC-MS *m/z*: 555 (M^+), 483, 457. HR-MS *m/z*: 555.7147 (M^+) (Calcd for $C_{28}H_{31}F_2N_5O_3S$: 555.7153).

1-Benzyl-2-[2-(3-*tert*-butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(4-chlorobenzyl)sulfonamide (13) ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 1.23 (9H, s), 2.95 (2H, t, *J*=5.9 Hz), 3.69 (2H, q, 5.8 Hz), 4.12 (2H, d, *J*=6.2 Hz), 4.59 (1H, s), 5.33 (1H, t, *J*=5.9 Hz), 5.40 (2H, s), 6.37 (1H, br s), 7.03 (2H, m), 7.16 (4H, q, *J*=5.3 Hz), 7.35 (4H, m), 7.72 (1H, dd, *J*=8.5, 1.6 Hz), 8.19 (1H, d, 1.5 Hz). LC-MS *m/z*: 554 (M^+), 481, 455. HR-MS *m/z*: 554.1967 (M^+) (Calcd for $C_{28}H_{32}ClN_5O_3S$: 554.1988).

1-(2-Fluorobenzyl)-2-[2-(3-*tert*-butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(4-fluorobenzyl)sulfonamide (14) ^{1}H-NMR (300 MHz, MeOD) δ: 1.24 (9H, s), 3.13 (2H, t, *J*=6.4 Hz), 3.60 (2H, t, *J*=6.5 Hz), 4.02 (2H, s), 5.61 (2H, s), 6.61 (4H, s), 6.87 (2H, m), 6.97 (1H, t, *J*=7.6 Hz), 7.15 (4H, m), 7.36 (1H, m), 7.50 (1H, d, *J*=8.7 Hz), 7.67 (1H, dd, *J*=8.6, 1.7 Hz), 8.08 (1H, d, *J*=1.3 Hz). LC-MS *m/z*: 556 (M^+), 483, 475.

1-(2-Chlorobenzyl)-2-[2-(3-*tert*-butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(4-fluorobenzyl)sulfonamide (15) ^{1}H-NMR (300 MHz, DMSO) δ: 1.17 (9H, s), 2.91 (2H, t, J=6.3 Hz), 3.45 (2H, q, J=6.2 Hz), 3.95 (2H, d, J=5.5 Hz), 5.62 (2H, s), 5.76 (1H, d, 1.9 Hz), 5.81 (1H, t, J=6.0 Hz), 6.55 (2H, s), 7.06 (2H, t, J=8.9 Hz), 7.24 (3H, m), 7.35 (1H, t, J=8.9 Hz), 7.56 (3H, m), 8.06 (2H, br s), 8.60 (1H, s). LC-MS m/z: 572 (M^+), 516, 499. HR-MS m/z: 572.1898 (M^+) (Calcd for $C_{28}H_{31}ClFN_5O_3S$: 572.1893). mp 139—140 °C.

1-(3-Chlorobenzyl)-2-[2-(3-*tert*-butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(4-fluorobenzyl)sulfonamide (16) ^{1}H-NMR (300 MHz, DMSO) δ: 1.18 (9H, s), 2.96 (2H, t, J=6.3 Hz), 3.46 (2H, q, J=6.3 Hz), 3.94 (2H, d, J=6.0 Hz), 5.59 (2H, s), 5.79 (1H, s), 5.86 (1H, t, J=6.1 Hz), 7.03 (3H, m), 7.25 (3H, m), 7.36 (2H, m), 7.65 (2H, q, J=5.0 Hz), 8.04 (2H, m). LC-MS m/z: 572 (M^+), 499, 473. HR-MS m/z: 572.1910 (M^+) (Calcd for $C_{28}H_{31}ClFN_5O_3S$: 572.1893). mp 135—136 °C.

1-(4-Chlorobenzyl)-2-[2-(3-*tert*-butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(4-fluorobenzyl)sulfonamide (17) ^{1}H-NMR (300 MHz, DMSO) δ: 1.18 (9H, s), 2.95 (2H, t, J=6.4 Hz), 3.45 (2H, q, J=6.1 Hz), 3.93 (2H, d, J=6.2 Hz), 5.57 (2H, s), 5.78 (1H, s), 5.83 (1H, t, J=5.9 Hz), 7.07 (4H, m), 7.25 (2H, m), 7.39 (2H, m), 7.63 (2H, q, J=4.0 Hz), 8.05 (2H, m). LC-MS m/z: 572 (M^+), 499, 473. HR-MS m/z: 572.1888 (M^+) (Calcd for $C_{28}H_{31}ClFN_5O_3S$: 572.1893). mp 180—181 °C.

1-(2,4-Dichlorobenzyl)-2-[2-(3-*tert*-butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(4-fluorobenzyl)sulfonamide (18) ^{1}H-NMR (300 MHz, DMSO) δ: 1.16 (9H, s), 2.90 (2H, t, J=6.3 Hz), 3.45 (2H, q, J=5.9 Hz), 3.94 (2H, d, 6.1 Hz), 5.60 (2H, s), 5.74 (1H, d, J=5.6 Hz), 5.80 (1H, t, J=6.3 Hz), 7.07 (2H, t, J=6.6 Hz), 7.28 (3H, m), 7.58 (2H, q, J=8.6 Hz), 7.74 (1H, d, J=2.1 Hz), 8.07 (2H, m). LC-MS m/z: 606 (M^+), 533, 507. HR-MS m/z: 606.1486 (M^+) (Calcd for $C_{28}H_{30}Cl_2FN_5O_3S$: 606.1504). mp 171—172 °C.

1-(2-Chlorobenzyl)-2-[2-(3-*tert*-butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(2,4-difluorobenzyl)sulfonamide (20) ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 1.19 (9H, s), 2.17 (1H, s), 2.95 (2H, s), 3.70 (2H, d, J=4.0 Hz), 4.18 (2H, d, J=5.6 Hz), 4.76 (1H, s), 5.42 (1H, br s), 5.49 (2H, s), 6.60 (3H, m), 7.23 (4H, m), 7.46 (1H, d, J=7.6 Hz), 7.71 (1H, d, J=7.8 Hz), 8.17 (1H, s). LC-MS m/z: 590 (M^+), 517, 491. HR-MS m/z: 590.1780 (M^+) (Calcd for $C_{28}H_{30}ClF_2N_5O_3S$: 590.1799).

1-(2-Chlorobenzyl)-2-[2-(3-*tert*-butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(2,4-dichlorobenzyl)sulfonamide (21) ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 1.21 (9H, s), 2.97 (2H, s), 3.71 (2H, d, J=6.5 Hz), 4.22 (2H, d, J=5.9 Hz), 4.80 (1H, br s), 5.48 (2H, s), 6.50 (1H, d, J=7.7 Hz), 7.22 (6H, m), 7.45 (1H, d, J=7.7 Hz), 7.67 (1H, d, J=8.4 Hz), 8.17 (1H, s). LC-MS m/z: 621 (M^+), 549, 525. HR-MS m/z: 622.1204 (M^+) (Calcd for $C_{28}H_{30}Cl_3N_5O_3S$: 622.1208).

1-(1-Phenylethyl)-2-[2-(3-*tert*-butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(4-chlorobenzyl)sulfonamide (22) ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 1.97 (9H, s), 2.00 (3H, d, J=8.4 Hz), 3.08 (2H, m), 3.81 (2H, q, J=6.0 Hz), 4.11 (2H, d, J=6.2 Hz), 5.53 (1H, br s), 5.82 (1H, q, J=7.1 Hz), 6.76 (1H, br s), 7.26 (9H, m), 7.56 (1H, dd, J=6.9, 1.7 Hz), 8.19 (1H, d, J=1.5 Hz). LC-MS m/z: 568 (M^+), 495, 469. HR-MS m/z: 568.2151 (M^+) (Calcd for $C_{29}H_{34}ClN_5O_3S$: 568.2144).

2-[2-(3-*tert*-Butyl-ureido)-ethyl]-1*H*-benzimidazole-5-(4-chlorobenzyl)sulfonamide (23) ^{1}H-NMR (300 MHz, MeOD) δ: 1.25 (9H, s), 3.07 (2H, t, J=6.6 Hz), 3.58 (2H, t, J=6.7 Hz), 4.02 (2H, s), 7.16 (4H, m), 7.65 (2H, m), 7.95 (1H, br s). LC-MS m/z: 464 (M^+), 391, 365.

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References and Notes

1) Lin J. H., Lu A. Y. H., *Clin. Pharmacokinet.*, **35**, 361—390 (1998).
2) Guengrich F. P., *Mol. Pharmacol.*, **33**, 500—508 (1988).
3) Roberts E. S., Hopkins N. E., Alworth W. L., Hollenberg P. F., *Chem. Res. Toxicol.*, **6**, 470—479 (1993).
4) Lopez-Garcia M. P., Dansette P. M., Mansuy D., *Biochemistry*, **33**, 166—175 (1993).
5) Nakamura T., Sato M., Kakinuma H., Miyata N., Taniguchi K., Bando K., Koda A., Kameo K., *J. Med. Chem.*, **46**, 5416—5427 (2003).
6) Wu Y. -J., He H., Hu S., Huang Y., Scola P. M., Grant-Young K., Bertekap R. L., Wu D., Gao Q., Li Y., Klakouski C., Westphal R. S., *J. Med. Chem.*, **46**, 4834—4837 (2003).
7) Hashimoto K., Tatsuta M., Kataoka M., Yasoshima K., Shogase Y., Shimazaki M., Yura T., Li Y., Yamamoto N., Gupta J. B., Urbahns K., *Bioorg. Med. Chem. Lett.*, **15**, 799—803 (2005).
8) Li Y., Kataoka M., Tatsuta M., Yasoshima K., Yura T., Urbahns K., Kiba A., Yamamoto N., Gupta J. B., Hashimoto K., *Bioorg. Med. Chem. Lett.*, **15**, 805—807 (2005).
9) Mortlock A. A., Bath C., Butlin R. J., Heys C., Hunt S. J., Reid A. C., Sumner N. F., Tang E. K., Whiting E., Wilson C., Wright N. D., *Bioorg. Med. Chem. Lett.*, **7**, 1399—1402 (1997).
10) Naritomi Y., Teramura Y., Terashita S., Kagayama A., *Drug Metab. Pharmacokinet.*, **19**, 55—61 (2004).
11) Yamaoka K., Tanigawara Y., Nakagawa T., Uno T., *J. Pharmacobio-Dyn.*, **4**, 879—885 (1981).
12) It is not clear how the structural modification on *N*-benzyl group affects a metabolic fate of these compounds.