Electronic Supplementary Material (ESI) for Medicinal Chemistry Communications

Electric supplementary information

Novel Candesartan Derivatives as Indoleamine-2,3-Dioxygenase Inhibitors

Kenji Matsuno,[a] Hiroshi Yamazaki,[a] Yoshinobu Isaka,[a] Kazushige Takai,[a] Yuka Unno,[a] Naohisa Ogo,[a] Yoshinobu Ishikawa,[a] Satoshi Fujii,[a] Osamu Takikawa[b] and Akira Asai*[a]

[a]Graduate School of Pharmaceutical Sciences, University of Shizuoka, 52-1 Yada, Suruga-ku, Shizuoka 422-8526, Japan. [b]National Institute for Longevity Sciences, National Center for Geriatrics and Gerontology, 36-3 Gengo, Morioka, Obu, Aichi 474-8522, Japan

Contents

1. Chemistry S-2
2. Molecular modelling S-12
3. Biological evaluations S-13

1. Chemistry

^{1}H NMR spectra were recorded with JEOL ECA-500 or JEOL A-500 spectrometers in the indicated solvent. Chemical shifts (δ) are expressed in parts per million (ppm) relative to the internal standard tetramethylsilane. Coupling constants (*J* values) are reported in Hz. High performance liquid chromatography (HPLC) and electrospray mass spectra were recorded in positive or negative mode on a Waters Quattro micro system with 2795 separations module and 2996 photodiode array detector. The sample was applied on a Waters XBrige™ C_{18} column (2.1 mm x 50 mm, 5 μm) with a Waters XBrige™ C_{18} guard cartridge (size 2.1×10 mm, 5 μm) and eluted at 1 mL/min with 4 min gradient (10 % B to 90% B) and then maintaining the final condition for 1 min, where solvent A was water (0.1% AcOH buffer) and solvent B was acetonitrile. Flash column chromatography by Biotage FLASH 40+ system with M or SNAP KP-SIL cartridges, open column chromatography with Wakogel C-200 (100-200 mesh, Wako) and preparative thin layer chromatography (PTLC) with Kiesel gel 60 F254 (1.0 mm, Merck) were performed for purification. Silica gel TLC plates from Merck (Kiesel gel 60 F_{254}: 0.25 mm silica gel precoated glass plates with fluorescent indicator visualizable at 254 nm) were used for thin layer chromatography (TLC). Reagents were used without further purification. The purity of tested compounds was ≥95%.

2-Ethoxy-*N*-methyl-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazole-carboxamide (5a): prepared from tritylcandesartan **5** and NH_3 in 1,4-dioxane according to the synthesis of compound **5b** in 5% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 7.60 (1H, d, *J* = 8.0 Hz), 7.58 (1H, d, *J* = 8.0 Hz), 7.50 (2H, m), 7.43 (1H, d, *J* = 8.0 Hz), 7.12-7.08 (2H, m), 6.96 (2H, d, *J* = 8.0 Hz), 6.91 (2H, d, *J* = 8.0 Hz), 5.41 (2H, s), 4.60 (2H, q, *J* = 6.9 Hz), 1.39 (3H, t, *J* = 6.9 Hz). HPLC: >99% purity, RT 2.08 min. ES-MS (*m/z*): 440 $(M + H)^+$.

2-Ethoxy-*N*-methyl-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazole-carboxamide (5b)

To a solution of commercially available tritylcandesartan **5** (75 mg, 0.11 mmol) in CH_2Cl_2 (1 mL) were added Hünig base (77 μL, 0.66 mmol), HATU (63 mg, 0.17 mmol) and methylamine hydrochloride (11 mg, 0.17 mmol). After being stirred at room temperature overnight under Ar atmosphere, aqueous NH_4Cl solution was added. The mixture was extracted with CH_2Cl_2 and the organic phase was evaporated under reduced pressure. The residue was dissolved in EtOH (2mL) solution of boric acid (3.4 mg, 0.055 mmol). After being refluxed for 7 h, the reaction mixture was evaporated. Brine was added to the residue and the mixture was extracted with CH_2Cl_2. Evaporation and purification by PTLC (CH_2Cl_2 : MeOH = 10 : 1) afforded the title compound (13 mg, 0.029 mmol) in 26% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.15 (1H, q, *J* = 4.5 Hz), 7.61 (1H, d, *J* = 8.0 Hz), 7.58 (1H, d, *J* = 8.0 Hz), 7.51 (2H, m), 7.43 (1H, d, *J* = 8.0 Hz), 7.12-7.08 (2H, m), 6.96 (2H, d, *J* = 8.0 Hz), 6.91 (2H, d, *J* = 8.0 Hz), 5.41 (2H, s), 4.60 (2H, q, *J* = 6.9 Hz), 2.62 (3H, d, *J* = 4.5 Hz), 1.39 (3H, t, *J* = 6.9 Hz). HPLC: >99% purity, RT 2.20 min. ES-MS (*m/z*): 454 $(M + H)^+$.

The following compounds (**5c**-**5f**) were synthesized from tritylcandesartan **5** and the corresponding amine in the same manner of compound **5b**.

***N*-Butyl-2-ethoxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazole-carboxamide (5c)** in 52% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.32 (1H, t, *J* = 5.7 Hz), 7.66-7.62 (2H, m), 7.56-7.50 (2H, m), 7.44 (1H, d, *J* = 7.9 Hz), 7.14-7.10 (2H, m), 6.97 (2H, d, *J* = 7.9 Hz), 6.93 (2H, d, *J* = 7.9 Hz), 5.41 (2H, s), 4.56 (2H, q, *J* = 6.8 Hz), 3.10 (2H, m), 1.38 (3H, t, *J* = 6.8 Hz), 1.32 (2H, m), 1.22 (2H, m), 0.81 (3H, t, *J* = 7.4 Hz). HPLC: >99% purity, RT 2.67 min. ES-MS (*m/z*): 496 $(M + H)^+$.

***N*-Cyclohexyl-2-ethoxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazole-carboxamide (5d)** in 41% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.29 (1H, d, *J* = 7.4 Hz),

7.67-7.61 (2H, m), 7.56-7.51 (2H, m), 7.43 (1H, d, *J* = 7.9 Hz), 7.13-7.10 (2H, m), 7.09-6.96 (4H, m), 5.41 (2H, s), 4.55 (2H, q, *J* = 6.8 Hz), 3.61 (1H, m), 1.66-1.60 (4H, m), 1.54 (1H, m), 1.37 (3H, t, *J* = 6.8 Hz), 1.24-1.22 (2H, m), 1.20-1.05 (3H, m). HPLC: 99% purity, RT 2.82 min. ES-MS (*m/z*): 522 $(M + H)^+$.

***N*-Cyclohexylmethyl-2-ethoxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5e)** in 28% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.35 (1H, t, *J* = 6.3 Hz), 7.66-7.61 (2H, m), 7.55 (1H, d, *J* = 8.0 Hz), 7.51 (1H, m), 7.43 (1H, d, *J* = 8.0 Hz), 7.12-7.11 (2H, m), 6.97 (2H, d, *J* = 8.6 Hz), 6.92 (2H, d, *J* = 8.6 Hz), 5.40 (2H, s), 4.54 (2H, q, *J* = 6.9 Hz), 2.97 (2H, t, *J* = 6.3 Hz), 1.64-1.61 (5H, m), 1.42 (1H, m), 1.34 (3H, t, *J* = 6.9 Hz), 1.08-1.07 (3H, m), 0.87-0.84 (2H, m). HPLC: >99% purity, RT 3.08 min. ES-MS (*m/z*): 536 $(M + H)^+$.

2-Ethoxy-*N*-phenyl-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5f) in 41% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 10.34 (1H, s), 7.63-7.57 (5H, m), 7.52 (1H, ddd, *J* = 8.6 Hz, 7.4 Hz, 1.1 Hz), 7.32-7.28 (3H, m), 7.24 (1H, dd, *J* = 7.4 Hz, 1.1 Hz), 7.18 (1H, t, *J* = 8.0 Hz), 7.07 (1H, t, *J* = 7.4 Hz), 6.87 (2H, d, *J* = 8.6 Hz), 6.78 (2H, d, *J* = 8.6 Hz), 5.33 (2H, s), 4.59 (2H, q, *J* = 6.9 Hz), 1.39 (3H, t, *J* = 6.9 Hz). HPLC: 97% purity, RT 2.82 min. ES-MS (*m/z*): 516 $(M + H)^+$.

2-Ethoxy-*N*-methyl-*N*-phenyl-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5g): prepared from tritylcandesartan **5** and *N*-methylaniline according to the synthesis of compound **5h** in 13% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 7.61 (2H, d, *J* = 8.5 Hz), 7.56-7.50 (3H, m), 7.27 (2H, brm), 7.12-7.09 (5H, m), 6.95 (2H, m), 6.70 (1H, brm), 6.51 (1H, brm), 5.38 (2H, s), 4.64 (2H, q, *J* = 6.8 Hz), 1.45 (3H, t, *J* = 6.8 Hz). HPLC: >99% purity, RT 2.73 min. ES-MS (*m/z*): 530 $(M + H)^+$.

***N*-(2-Chlorophenyl)-2-ethoxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5h)**

To an ice cooled (0℃) solution of commercially available tritylcandesartan **5** (100 mg, 0.15 mmol) in pyridine (15 mL) was slowly dropped thionyl chloride (3 mL). After being stirred at 0℃ for 2.5 h under Ar atmosphere, the reaction mixture was evaporated under reduced pressure. The residue was dissolved in CH_2Cl_2 (2 mL), then Et_3N (61 μL, 0.48 mmol) and 2-chloroaniline (23 μL, 0.22 mmol) were added. The mixture was stirred at room temperature overnight, aqueous NH_4Cl solution was added. The mixture was extracted with CH_2Cl_2 and the organic phase was evaporated. The residue was dissolved in EtOH (2mL) solution of boric acid (14 mg, 0.22 mmol). After being refluxed overnight, the reaction mixture was evaporated. Aqueous NH_4Cl solution was added to the residue and the mixture was extracted with CH_2Cl_2. Evaporation and purification by flash column chromatography (CH_2Cl_2 : MeOH = 100 : 2 to 100 : 3) afforded the title compound (25 mg, 0.045 mmol) in 31% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 10.15 (1H, s), 7.64-7.62 (3H, m), 7.54 (1H, d, *J* = 7.4 Hz), 7.51 (1H, d, *J* = 7.4 Hz), 7.44-7.42 (2H, m), 7.35 (1H, t, *J* = 7.4 Hz), 7.28 (1H, d, *J* = 7.4 Hz), 7.24 (2H, m), 6.99 (2H, d, *J* = 8.6 Hz), 6.96 (2H, d, *J* = 8.6 Hz), 5.46 (2H, s), 4.58 (2H, q, *J* = 6.9 Hz), 1.39 (3H, t, *J* = 6.9 Hz). HPLC: 97% purity, RT 2.85 min. ES-MS (*m/z*): 552 (^{37}ClM + H)$^+$, 550 (^{35}ClM + H)$^+$.

The following compounds (**5i-5j**) were synthesized from tritylcandesartan **5** and the corresponding amine in the same manner of compound **5b**.

***N*-(3-Chlorophenyl)-2-ethoxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5i)** in 40% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 10.44 (1H, s), 7.75 (1H, s), 7.63-7.59 (3H, m), 7.56-7.51 (2H, m), 7.32 (1H, t, *J* = 8.6 Hz), 7.27 (1H, d, *J* = 8.6 Hz), 7.24 (1H, d, *J* = 8.6 Hz), 7.19 (1H, t, *J* = 8.0 Hz), 7.13 (1H, dd, *J* = 8.0 Hz, 1.1 Hz), 6.84 (2H, d, *J* = 8.0 Hz), 6.78 (2H, d, *J* = 8.0 Hz), 5.34 (2H, s), 4.62 (2H, q, *J* = 6.9 Hz), 1.42 (3H, t, *J* = 6.9

Hz). HPLC: 97% purity, RT 3.12 min. ES-MS (*m/z*): 552 (^{37}ClM + H)$^+$, 550 (^{35}ClM + H)$^+$.

2-Ethoxy-*N*-(3-methoxyphenyl)-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5j) in 29% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 7.63-7.58 (3H, m), 7.52 (1H, t, *J* = 8.0 Hz), 7.30 (1H, d, *J* = 8.0 Hz), 7.24-7.16 (4H, m), 6.87 (2H, d, *J* = 8.0 Hz), 6.78 (2H, d, *J* = 8.0 Hz), 6.87 (1H, d, *J* = 8.0 Hz), 5.33 (2H, s), 4.59 (2H, q, *J* = 6.9 Hz), 1.41 (3H, t, *J* = 6.9 Hz). HPLC: >99% purity, RT 2.70 min. ES-MS (*m/z*): 546 (M + H)$^+$.

***N*-(4-Chlorophenyl)-2-ethoxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5k)**: prepared from tritylcandesartan **5** and 4-chloroaniline according to the synthesis of compound **5h** in 26% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 10.45 (1H, s), 7.66-7.59 (3H, m), 7.59 (2H, d, *J* = 8.0 Hz), 7.53 (1H, t, *J* = 7.4 Hz), 7.36 (2H, d, *J* = 8.0 Hz), 7.27 (1H, d, *J* = 8.0 Hz), 7.24 (1H, d, *J* = 7.4 Hz), 7.19 (1H, t, *J* = 8.0 Hz), 6.85 (2H, d, *J* = 8.0 Hz), 6.79 (2H, d, *J* = 8.0 Hz), 5.33 (2H, s), 4.60 (2H, q, *J* = 6.9 Hz), 1.41 (3H, t, *J* = 6.9 Hz). HPLC: >99% purity, RT 3.12 min. ES-MS (*m/z*): 552 (^{37}ClM + H)$^+$, 550 (^{35}ClM + H)$^+$.

***N*-Benzyl-2-ethoxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5l)**: prepared from tritylcandesartan **5** and benzylamine according to the synthesis of compound **5b** in 35% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.94 (1H, t, *J* = 5.7 Hz), 7.68-7.64 (2H, m), 7.57-7.54 (2H, m), 7.46 (1H, d, *J* = 7.4 Hz), 7.29-7.21 (6H, m), 7.14 (1H, t, *J* = 8.0 Hz), 6.94 (2H, d, *J* = 8.6 Hz), 6.90 (2H, d, *J* = 8.6 Hz), 5.40 (2H, s), 4.57 (2H, q, *J* = 7.4 Hz), 4.36 (2H, d, *J* = 5.7 Hz), 1.38 (3H, t, *J* = 7.4 Hz). HPLC: >99% purity, RT 2.87 min. ES-MS (*m/z*): 530 (M + H)$^+$.

The following compounds (**5m-5w**) were synthesized from tritylcandesartan **5** and the corresponding amine in the same manner of compound **5b**.

***N*-(2-Chlorobenzyl)-2-ethoxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5m)** in 57% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.95 (1H, t, *J* = 5.7 Hz), 7.69-7.64 (2H, m), 7.59-7.55 (2H, m), 7.54 (1H, d, *J* = 7.4 Hz), 7.48 (1H, d, *J* = 8.0 Hz), 7.43 (1H, dd, *J* = 8.0 Hz, 1.1 Hz), 7.33 (1H, dd, *J* = 8.0 Hz, 1.1 Hz), 7.29 (1H, d, *J* = 8.0 Hz), 7.27 (2H, dd, *J* = 8.0 Hz, 1.7Hz), 7.21 (1H, dd, *J* = 8.0 Hz, 1.1 Hz), 7.17 (1H, m), 6.98 (2H, d, *J* = 8.6 Hz), 6.90 (2H, d, *J* = 8.6 Hz), 5.40 (2H, s), 4.57 (2H, q, *J* = 6.9 Hz), 4.41 (2H, d, *J* = 5.7 Hz), 1.38 (3H, t, *J* = 6.9 Hz). HPLC: >99% purity, RT 2.97 min. ES-MS (*m/z*): 566 (^{37}ClM + H)$^+$, 564 (^{35}ClM + H)$^+$.

2-Ethoxy-*N*-(3-fluorobenzyl)-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5n) in 71% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.95 (1H, t, *J* = 6.3 Hz), 7.66-7.62 (2H, m), 7.56-7.53 (2H, m), 7.44 (1H, d, *J* = 7.4 Hz), 7.30 (1H, m), 7.21 (1H, d, *J* = 7.4 Hz), 7.15 (1H, d, *J* = 8.0 Hz), 7.14-7.11 (2H, m), 7.04 (1H, m), 6.92 (2H, d, *J* = 8.0 Hz), 6.88 (2H, d, *J* = 8.0 Hz), 5.37 (2H, s), 4.56 (2H, q, *J* = 6.9 Hz), 4.34 (2H, d, *J* = 6.3 Hz), 1.36 (3H, t, *J* = 6.9 Hz). HPLC: >99% purity, RT 2.87 min. ES-MS (*m/z*): 549 (M + H)$^+$.

***N*-(3-Chlorobenzyl)-2-ethoxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5o)** in 43% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.96 (1H, t, *J* = 6.3 Hz), 7.68-7.64 (2H, m), 7.58-7.54 (2H, m), 7.45 (1H, d, *J* = 8.0 Hz), 7.39 (1H, s), 7.29-7.25 (3H, m), 7.21 (1H, d, *J* = 8.0 Hz), 7.16 (1H, t, *J* = 8.0 Hz), 6.92 (2H, d, *J* = 8.6 Hz), 6.88 (2H, d, *J* = 8.6 Hz), 5.37 (2H, s), 4.56 (2H, q, *J* = 6.9 Hz), 4.33 (2H, d, *J* = 6.3 Hz), 1.36 (3H, t, *J* = 6.9 Hz). HPLC: 98% purity, RT 3.00 min. ES-MS (*m/z*): 566 (^{37}ClM + H)$^+$, 564 (^{35}ClM + H)$^+$.

2-Ethoxy-*N*-(3-methylbenzyl)-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5p) in 74% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.89 (1H, t, *J* = 6.3 Hz), 7.67-7.63 (2H, m), 7.55-7.53 (2H, m), 7.44 (1H, d, *J* = 7.4 Hz), 7.19 (1H, d, *J* = 8.0 Hz), 7.15-7.12 (3H, m), 7.06 (1H, d, *J* = 8.0 Hz), 7.01 (1H, d, *J* = 7.4 Hz), 6.91 (4H, m), 5.37 (2H, s), 4.55 (2H, q, *J* = 6.9 Hz), 4.30 (2H, d, *J* = 6.3 Hz), 2.21 (3H, s), 1.37 (3H, t, *J* = 6.9 Hz). HPLC:

>99% purity, RT 2.95 min. ES-MS (*m/z*): 544 $(M + H)^+$.

2-Ethoxy-*N*-(3-methoxybenzyl)-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5q) in 19% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.88 (1H, brs), 7.59-7.47 (4H, m), 7.37-7.36 (2H, m), 7.17-7.13 (3H, m), 7.05-6.78 (6H, m), 5.37 (2H, s), 4.58 (2H, q, J = 7.4 Hz), 4.32 (2H, brs), 3.69 (3H, s), 1.23 (2H, t, J = 7.4 Hz). HPLC: >99% purity, RT 2.80 min. ES-MS (*m/z*): 560 $(M + H)^+$.

***N*-(4-Chlorobenzyl)-2-ethoxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5r)** in 42% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.93 (1H, t, J = 6.3 Hz), 7.69-7.64 (2H, m), 7,58-7.56 (2H, m), 7.46 (1H, d, J = 8.0 Hz), 7.32 (2H, d, J = 8.6 Hz), 7.28 (2H, d, J = 8.6 Hz), 7.21 (1H, d, J = 8.0 Hz), 7.15 (1H, t, J = 8.0 Hz), 6.92 (2H, d, J = 8.6 Hz), 6.85 (2H, d, J = 8.6 Hz), 5.37 (2H, s), 4.54 (2H, q, J = 6.9 Hz), 4.30 (2H, d, J = 6.3 Hz), 1.36 (3H, t, J = 6.9 Hz). HPLC: 97% purity, RT 3.02 min. ES-MS (*m/z*): 566 (^{37}ClM + H)$^+$, 564 (^{35}ClM + H)$^+$.

2-Ethoxy-*N*-(1-naphthylmethyl)-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (5s) in 14% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 9.02 (1H, t, J = 5.7 Hz), 8.19 (1H, m), 7.93 (1H, m), 7.82 (1H, d, J = 8.6 Hz), 7.66-7.63 (2H, m), 7.56-7.51 (4H, m), 7.48 (1H, d, J = 8.0 Hz), 7.41-7.37 (2H, m), 7.19 (1H, d, J = 7.4 Hz), 7.11 (1H, d, J = 7.4 Hz), 6.84 (4H, m), 5.37 (2H, s), 4.80 (2H, q, J = 5.7 Hz), 4.55 (2H, d, J = 6.9 Hz), 1.35 (3H, t, J = 6.9 Hz). HPLC: 98% purity, RT 2.95 min. ES-MS (*m/z*): 580 $(M + H)^+$.

2-Ethoxy-*N*-(2-naphthylmethyl)-1-{2'-[5-(1*H*-tetrazolyl)]-4-biphenylylmethyl}-7-(1*H*)-benzimidazolecarboxamide (5t) in 85% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 9.03 (1H, t, J = 6.2 Hz), 7.86-7.81 (4H, m), 7.64-7.61 (2H, m), 7.56-7.53 (2H, m), 7.47-7.44 (3H, m), 7.33 (1H, d, J = 8.5 Hz), 7.26 (1H, d, J = 7.9 Hz), 7.15 (1H, t, J = 7.9 Hz), 6.88-6.81 (4H, m), 5.40 (2H, s), 4.57 (2H, q, J = 7.4 Hz), 4.52 (2H, d, J = 6.2 Hz), 1.37 (3H, t, J = 7.4 Hz). HPLC: >99% purity, RT 2.95 min. ES-MS (*m/z*): 580 $(M + H)^+$.

(*dl*)-2-Ethoxy-*N*-(1-phenethyl)-1-{2'-[5-(1*H*-tetrazolyl)]-4-biphenylylmethyl}-7-(1*H*)-benzimidazolecarboxamide (5u) in 60% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.93 (1H, d, J = 8.5 Hz), 7.66-7.62 (2H, m), 7.56-7.53 (2H, m), 7.41 (1H, d, J = 7.9 Hz), 7.36 (2H, d, J = 7.4 Hz), 7.28-7.25 (2H, m), 7.20-7.12 (3H, m), 6.90-6.86 (4H, m), 5.37 (1H, d, J = 16 Hz), 5.32 (1H, d, J = 16 Hz), 5.06 (1H, m), 4.56 (2H, q, J = 6.8 Hz), 1.38 (3H, t, J = 6.8 Hz), 1.27 (3H, d, J = 7.4 Hz). HPLC: 99% purity, RT 2.85 min. ES-MS (*m/z*): 544 $(M + H)^+$.

2-Ethoxy-*N*-(2-phenyl-2-propyl)-1-{2'-[5-(1*H*-tetrazolyl)]-4-biphenylylmethyl}-7-(1*H*)-benzimidazolecarboxamide (5v) in 36% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.72 (1H, s), 7.67-7.62 (2H, m), 7.56-7.54 (2H, m), 7.49 (1H, d, J = 7.9 Hz), 7.34-7.31 (3H, m), 7.19-7.11 (4H, m), 7.01 (2H, d, J = 8.5 Hz), 6.91 (2H, d, J = 7.9 Hz), 5.30 (2H, s), 4.49 (2H, q, J = 7.4 Hz), 1.45 (6H, s), 1.30 (3H, t, J = 7.4 Hz). HPLC: 97% purity, RT 3.00 min. ES-MS (*m/z*): 558 $(M + H)^+$.

2-Ethoxy-*N*-(2-phenethyl)-1-{2'-[5-(1*H*-tetrazolyl)]-4-biphenylylmethyl}-7-(1*H*)-benzimidazole carboxamide (5w) in 54% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.41 (1H, t, J = 5.2 Hz), 7.63-7.51 (4H, m), 7.40 (1H, d, J = 8.0 Hz), 7.28-7.25 (2H, m), 7.19-7.18 (3H, m), 7.10 (1H, t, J = 8.0 Hz), 7.05 (1H, d, J = 8.0 Hz), 6.96 (2H, d, J = 8.0 Hz), 6.92 (2H, d, J = 8.0 Hz), 5.38 (2H, s), 4.56 (2H, q, J = 6.9 Hz), 3.32 (2H, brs), 2.66 (2H, t, J =7.9 Hz), 1.36 (3H, t, J = 6.9 Hz). HPLC: >99% purity, RT 2.88 min. ES-MS (*m/z*): 544 $(M + H)^+$.

7-Amino-2-ethoxy-1-[2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazole (6)

To an ice cooled solution of commercially available tritylcandesartan (2.50 g, 3.66 mmol) in toluene (25 mL) and triethylamine (1.52 mL, 10.9 mmol) was added diphenylphosphoryl

azide (1.18 mL, 5.48 mmol). After being stirred at 0℃ for 2.5 h under Ar atomosphere, the reaction mixture was refluxed for 4 h. H_2O was added therein and the mixture was refluxed again overnight. After the reaction mixture was cooled to room temperature, brine was added therein, followed by extraction with CH_2Cl_2. The organic phase was dried over sodium sulfate and evaporated under reduced pressure. The residue was purified by silica gel column chromatography (hexane : AcOEt = 2 : 1 to 1 : 1) to afford the title compound (883 mg, 1.35 mmol) as pale yellow solid in 37% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 7.76 (1H, d, J = 7.4 Hz), 7.58 (1H, t, J = 7.4 Hz), 7.51 (1H, t, J = 7.4 Hz), 7.42 (1H, d, J = 7.9 Hz), 7.37-7.29 (10H, m), 7.06-7.05 (2H, m), 7.00 (2H, d, J = 7.9 Hz), 6.86-6.85 (5H, m), 6.80 (1H, t, J = 7.9 Hz), 6.75 (1H, d, J = 7.9 Hz), 6.40 (1H, d, J = 7.4 Hz), 5.33 (2H, s), 4.73 (2H, brs), 4.42 (2H, q, J = 6.8 Hz), 1.27 (3H, t, J = 6.8 Hz). ES-MS (*m/z*): 654 $(M + H)^+$.

_N_-{2-Ethoxy-1-[2'-(5-(1_H_)-tetrazolyl)-4-biphenylylmethyl]-7-(1_H_)-benzimidazolyl}benzamide (7a)

To a solution of compound **6** (80 mg, 0.12 mmol) in CH_2CH_2 (1 mL) were added benzoyl chloride (17 μL, 0.15 mmol) and triethylamine (34 μL, 0.24 mmol). After being stirred overnight at room temperature, sat. $NaHCO_3$aq. was added therein. The mixture was extracted with CH_2Cl_2, followed by evaporation. The residue was dissolved in EtOH (2 mL) and boric acid (11 mg, 0.18 mmol). The mixture was refluxed overnight, evaporated, suspended in brine, followed by extraction with CH_2Cl_2. The residue was purified by silica gel column chromatography (CH_2Cl_2 : MeOH = 100 : 3 to 100 : 4) to afford the title compound (22 mg, 0.04 mmol) as pale brown solid in 33% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 10.21 (1H, s), 7.82 (2H, d, J = 7.4 Hz), 7.65-7.48 (6H, m), 7.43-7.38 (2H, m), 7.12 (1H, t, J = 7.9 Hz), 6.93-6.90 (3H, m), 6.85 (2H, d, J = 7.9 Hz), 5.17 (2H, s), 4.55 (2H, q, J = 7.4 Hz), 1.36 (3H, t, J = 7.4 Hz). HPLC: 98% purity, RT 2.47 min. ES-MS (*m/z*): 516 $(M + H)^+$.

The following compounds (**7b**-**7e**) were synthesized from compound **6** and the corresponding acyl chloride in the same manner of compound **7a**.

_N_-{2-Ethoxy-1-[2'-(5-(1_H_)-tetrazolyl)-4-biphenylylmethyl]-7-(1_H_)-benzimidazolyl}-2-chloro-benzamide (7b) in 56% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 10.34 (1H, s), 7.66-7.62 (2H, m), 7.56-7.53 (2H, m), 7.50-7.47 (2H, m), 7.42-7.39 (2H, m), 7.14 (1H, t, J = 7.9 Hz), 7.10 (1H, dd, J = 7.9 Hz, 1.7 Hz), 7.04-7.00 (5H, m), 5.36 (2H, s), 4.52 (2H, q, J = 6.8 Hz), 1.34 (3H, t, J = 6.8 Hz). HPLC: >99% purity, RT 2.58 min. ES-MS (*m/z*): 552 (^{37}ClM + H)$^+$, 550 (^{35}ClM + H)$^+$.

_N_-{2-Ethoxy-1-[2'-(5-(1_H_)-tetrazolyl)-4-biphenylylmethyl]-7-(1_H_)-benzimidazolyl}-3-chloro-benzamide (7c) in 61% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 7.79-7.75 (2H, m), 7.66-7.62 (3H, m), 7.56-7.51 (2H, m), 7.41 (2H, d, J = 7.9 Hz), 7.13 (1H, t, J = 7.9 Hz), 6.94-6.89 (3H, m), 6.82 (2H, d, J = 7.9 Hz), 5.21 (2H, s), 4.57 (2H, q, J = 6.8 Hz), 1.38 (3H, t, J = 6.8 Hz). HPLC: 99% purity, RT 2.67 min. ES-MS (*m/z*): 552 (^{37}ClM + H)$^+$, 550 (^{35}ClM + H)$^+$.

_N_-{2-Ethoxy-1-[2'-(5-(1_H_)-tetrazolyl)-4-biphenylylmethyl]-7-(1_H_)-benzimidazolyl}-4-chloro-benzamide (7d) in 59% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 10.26 (1H, s), 7.80 (2H, d, J = 8.5 Hz), 7.62-7.60 (2H, m), 7.57-7.51 (3H, m), 7.41-7.38 (2H, m), 7.11 (1H, t, J = 7.9 Hz), 6.91-6.90 (3H, m), 6.81 (2H, d, J = 7.4 Hz), 5.16 (2H, s), 4.54 (2H, q, J = 6.8 Hz), 1.36 (3H, t, J = 6.8 Hz). HPLC: 99% purity, RT 2.72 min. ES-MS (*m/z*): 552 (^{37}ClM + H)$^+$, 550 (^{35}ClM + H)$^+$.

_N_-{2-Ethoxy-1-[2'-(5-(1_H_)-tetrazolyl)-4-biphenylylmethyl]-7-(1_H_)-benzimidazolyl}-2-phenyl-acetamide (7e) in 51% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 9.90 (1H, s), 7.67-7.63 (2H, m), 7.55 (1H, t, J = 7.4 Hz), 7.47 (1H, d, J = 7.9 Hz), 7.31 (1H, d, J = 7.9 Hz), 7.27-7.18 (5H, m), 7.05 (1H, t, J = 7.9 Hz), 6.97 (2H, d, J = 7.9 Hz), 6.84-6.80 (3H, m), 5.08 (2H, s), 4.51 (2H, q, J = 6.8 Hz), 3.54 (2H, s), 1.33 (3H, t, J = 6.8 Hz). HPLC: 95% purity, RT 2.85 min. ES-MS (*m/z*): 530 $(M + H)^+$.

2-(Methoxycarbonyl)-6-nitrobenzoic acid

To an ice cooled solution of 3-nitrophtalic acid (3.0 g, 14 mmol) in MeOH (100 mL) was slowly dropped thionyl chloride (1.6 mL, 21 mmol). After being refluxed overnight under Ar atmosphere, the reaction mixture was evaporated under reduced pressure. The residue was dissolved in AcOEt and the mixture was extracted with sat. $NaHCO_3$*aq*. The water phase was acidified with concd. HCl and extracted with AcOEt. The organic phase was dried over sodium sulfate and evaporated under reduced pressure to afford the title compound (2.9 g, 13 mmol) as colourless solid in 90% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.32 (1H, d, J = 8.0 Hz, 8.21 (1H, d, J = 8.0 Hz), 7.82 (1H, t, J = 8.0 Hz), 3.85 (3H, s).

Methyl 2-amino-3-nitrobenzoate

To a solution of 2-(methoxycarbonyl)-6-nitrobenzoic acid (4.51 g, 20.0 mmol) in THF (54 mL) and Et_3N (11.2 mL, 80.2 mmol) was dropped diphenylphosphoryl azide (11.3 mL, 52.1 mmol). After being stirred at room temperature overnight under Ar atmosphere, the reaction mixture was refluxed for 1.5 h. H_2O (25 mL) was added to reaction mixture. After being further refluxed for 42 h under Ar atmosphere, the reaction mixture was evaporated under reduced pressure. The residue was suspended in H_2O and the suspension was extracted with AcOEt. The organic phase was dried over sodium sulfate and evaporated under reduced pressure. The residue was purified by silica gel column chromatography (hexane : AcOEt = 8 : 1 to 7 : 1) to afford the title compound (3.25 g, 16.6 mmol) as yellow solid in 83% yield. ^{1}H NMR (500MHz, $CDCl_3$) δ: 8.52 (1H, brs), 8.38 (1H, d, J = 8.0 Hz), 8.24 (1H, d, J = 8.0 Hz), 6.66 (1H, t, J = 8.0 Hz), 3.92 (3H, s). ES-MS (*m/z*): 197 $(M + H)^+$.

Methyl 2-trifluoroacetylamino-3-nitrobenzoate

To an ice cooled solution of methyl 2-amino-3-nitrobenzoate (3.26 g, 16.6 mmol) in THF (150 mL) and Et_3N (7.70 mL, 54.8 mmol) was added TFAA (7.70 mL, 54.8 mmol). After being stirred at room temperature overnight under Ar atmosphere, H_2O was added to the reaction mixture. The resulting precipitate was filtered and washed with hexane to afford the title compound (4.53 g, 15.5 mmol) as colourless solid in 93% yield. ^{1}H NMR (500MHz,$CDCl_3$) δ: 11.36 (1H, brs), 8.32 (1H, d, J = 8.0 Hz), 8.20 (1H, d, J = 8.0 Hz), 7.50 (1H, t, J = 8.0 Hz), 4.02 (3H, s). ES-MS (*m/z*): 293 $(M + H)^+$.

Methyl 2-(4-biphenylylmethyl)amino-3-nitrobenzoate

To a solution of methyl 2-trifluoroacetylamino-3-nitrobenzoate (203 mg, 0.695 mmol) in CH_3CN (12 mL) was added K_2CO_3 (115 mg, 0.832 mmol) and 4-biphenylylmethylbromide (190 mg, 0.769 mmol). After being refluxed for 7.5 h under Ar atmosphere, the reaction mixture was filtered and the filtrate was evaporated under reduced pressure. The residue was suspended in brine and was extracted with CH_2Cl_2. The organic phase was dried over sodium sulfate and evaporated. The residue was dissolved in MeOH (11 mL) and K_2CO_3 (117 mg, 0.847 mmol) was added therein. After being stirred at room temperature for 18 h under Ar atmosphere, the reaction mixture was filtered and the filtrate was evaporated under reduced pressure to afford the title compound (201 mg, 0.556 mmol) in 80% yield. ^{1}H NMR (500MHz, $CDCl_3$) δ: 8.83 (1H, brs), 8.11 (1H, d, J = 8.0 Hz), 8.01 (1H, d, J = 8.6 Hz), 7.59-7.56 (4H, m), 7.44 (2H, t, J = 8.0 Hz), 7.37-7.33 (3H, m), 6.72 (1H, t, J = 7.4 Hz), 4.19 (2H, d, J = 5.2 Hz), 3.89 (3H, s).

Methyl 1-(4-biphenylyl)methyl-2-ethoxy-7-(1*H*)-benzimidazolecarboxylate

To a cooling (10℃) solution of methyl 2-(4-biphenylylmethyl)amino-3-nitrobenzoate (190 mg, 0.527 mmol) in AcOH (12 mL) was added iron powder (296 mg, 5.30 mmol). After being stirred at 85℃ for 2 h under Ar atmosphere, the reaction mixture was filtered through Celite and the filtrate was evaporated under reduced pressure. The residue was dissolved in AcOH and tetraethyl orthocarbonate (0.882 mL, 4.22 mmol) was added threin. After being stirred at room temperature for 1 h under Ar atmosphere, the reaction mixture was evaporated under reduced pressure. The residue was suspended in sat. $NaHCO_3$*aq*., followed by extraction with

CH_2Cl_2. The organic phase was dried over sodium sulfate and evaporated under reduced pressure. The residue was purified by silica gel column chromatography (hexane : AcOEt = 7 : 1) to afford the title compound (166 mg, 0.430 mmol) as colourless solid in 81% yield. ^{1}H NMR (500MHz, $CDCl_3$) δ: 7.74 (1H, d, *J* = 8.0 Hz), 7.55-7.51 (3H, m), 7.45 (2H, d, *J* = 8.6 Hz), 7.40 (2H, t, *J* = 7.4 Hz), 7.32 (1H, m), 7.17 (1H, t, *J* = 8.0 Hz), 7.04 (2H, d, *J* = 8.0 Hz), 5.66 (2H, s), 4.67 (2H, q, *J* = 6.9 Hz), 3.75 (3H, s), 1.49 (3H, t, *J* = 6.9 Hz). ES-MS (*m/z*): 387 $(M + H)^+$.

1-(4-Biphenylyl)methyl-2-ethoxy-7-(1*H*)-benzimidazolecarboxylic acid
To a solution of methyl 1-(4-biphenylyl)methyl-2-ethoxy-7-(1*H*)-benzimidazolecarboxylate (150 mg, 0.389 mmol) in EtOH (12 mL) was added 4 mol/L NaOH*aq.* (1.1 mL) and H_2O (4 mL). After being stirred at room temperature overnight, the reaction mixture was evaporated under reduced pressure. The residue was dissolved in water, followed by washing with CH_2Cl_2. The water phase was acidified with 5 mol/L HCl*aq*. The resulting precipitate was filtered to afford the title compound (104 mg, 0.279 mmol) as colourless solid in 72% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 7.66 (1H, dd, *J* = 7.4 Hz, 1.1 Hz), 7.58 (2H, d, *J* = 7.4 Hz), 7.55 (2H, d, *J* = 8.6 Hz), 7.50 (1H, dd, *J* = 8.0 Hz, 1.1 Hz), 7.41 (2H, t, *J* = 7.4 Hz), 7.33 (1H, m), 7.16 (1H, t, *J* = 8.0 Hz), 7.03 (2H, d, *J* = 8.6 Hz), 5.65 (2H, s), 4.60 (2H, q, *J* = 6.9 Hz), 1.40 (3H, t, *J* = 6.9 Hz). ES-MS (*m/z*): 373 $(M + H)^+$.

***N*-Benzyl-1-(4-biphenylyl)methyl-2-ethoxy-7-(1*H*)-benzimidazolecarboxamide (12a)**
To a solution of 1-(4-biphenylyl)methyl-2-ethoxy-7-(1*H*)-benzimidazolecarboxylic acid (58 mg, 0.15 mmol) in CH_2Cl_2 (6 mL) was added Hünig base (41 µL, 0.23 mmol), HBTU (87 mg, 0.23 mmol) and benzylamine (19 µL, 0.17 mmol). After being stirred at room temperature overnight under Ar atmosphere, H_2O was added. The mixture was extracted with CH_2Cl_2 and the organic phase was dried over sodium sulphate, followed by evaporation. The precipitate was purified by recrystalization from MeOH to afford the title compound (53 mg, 0.11 mmol) as colourless solid in 74% yield. ^{1}H NMR (500MHz, $CDCl_3$) δ: 7.66 (1H, m), 7.53 (2H, d, *J* = 7.4 Hz), 7.46 (2H, d, *J* = 8.6 Hz), 7.43 (2H, t, *J* = 7.4 Hz), 7.35 (1H, t, *J* = 7.4 Hz), 7.19-7.18 (3H, m), 7.13-7.07 (6H, m), 5.83 (1H, t, *J* = 5.7 Hz), 5.61 (2H, s), 4.69 (2H, q, *J* = 6.9 Hz), 4.38 (2H, d, *J* = 5.7 Hz), 1.50 (3H, t, *J* = 6.9 Hz). HPLC: >99% purity, RT 3.47 min. ES-MS (*m/z*): 462 $(M + H)^+$.

Methyl 2-(4-iodobenzyl)amino-3-nitrobenzoate: prepared from methyl 2-trifluoroacetylamino-3-nitrobenzoate and 4-iodobenzyl bromide according to the synthesis of methyl 2-(4-biphenylylmethyl)amino-3-nitrobenzoate as yellow solid in 87% yield. ^{1}H NMR (500MHz,$CDCl_3$) δ: 8.77 (1H, brs), 8.10 (1H, d, *J* = 7.4 Hz), 7.94 (1H, d, *J* = 8.0 Hz), 7.65 (2H, d, *J* = 8.0 Hz), 7.03 (2H, d, *J* = 8.0 Hz), 6.72 (1H, t, *J* = 8.0 Hz), 4.09 (2H, d, *J* = 4.6 Hz), 3.88 (3H, s). ES-MS (*m/z*): 413 $(M + H)^+$.

Methyl 2-ethoxy-1-(4-iodobenzyl)-7-(1*H*)-benzimidazolecarboxylate: prepared from methyl 2-(4-iodobenzyl)amino-3-nitrobenzoate according to the synthesis of methyl 1-(4-biphenylyl)methyl-2-ethoxy-7-(1*H*)-benzimidazolecarboxylate as colourless solid in 75% yield. ^{1}H NMR (500MHz, $CDCl_3$) δ: 10.68 (1H, s), 8.01 (1H, d, *J* = 8.0 Hz), 7.92 (1H, d, *J* = 7.4 Hz), 7.88 (2H, d, *J* = 8.6 Hz), 7.82 (2H, d, *J* = 8.6 Hz), 7.33 (1H, t, *J* = 8.0 Hz), 4.03 (3H, s). ES-MS (*m/z*): 379 $(M + H)^+$.

2-Ethoxy-1-(4-iodobenzyl)-7-(1*H*)-benzimidazolecarboxylic acid: prepared from methyl 2-ethoxy-1-(4-iodobenzyl)-7-(1*H*)-benzimidazolecarboxylate according to the synthesis of 1-(4-biphenylyl)methyl-2-ethoxy-7-(1*H*)-benzimidazolecarboxylic acid as colourless solid in quantitative yield. ^{1}H NMR (500MHz, CD_3OD) δ: 7.63 (1H, dd, *J* = 8.0 Hz, 1.1 Hz), 7.60-7.57 (3H, m), 7.19 (1H, t, *J* = 8.0 Hz), 6.79 (2H, d, *J* = 8.6 Hz), 5.63 (2H, s), 4.60 (2H, q, *J* = 6.9 Hz), 1.46 (3H, t, *J* = 6.9 Hz). ES-MS (*m/z*): 423 $(M + H)^+$.

***N*-Benzyl-2-ethoxy-1-(4-iodobenzyl)-7-(1*H*)-benzimidazolecarboxamide**: prepared from 2-ethoxy-1-(4-iodobenzyl)-7-(1*H*)-benzimidazolecarboxylic acid according to the synthesis of

compound **12a** as colourless solid in 95% yield. ^{1}H NMR (500MHz, $CDCl_3$) δ: 7.66-7.64 (1H, m), 7.53 (2H, d, *J* = 8.0 Hz), 7.36-7.29 (3H, m), 7.14-7.10 (4H, m), 6.74 (2H, d, *J* = 8.0 Hz), 5.88 (1H, t, *J* = 5.2 Hz), 5.50 (2H, s), 4.66 (2H, q, *J* = 6.9 Hz), 4.38 (2H, d, *J* = 5.2 Hz), 1.48 (3H, t, *J* = 6.9 Hz). ES-MS (*m/z*): 512 $(M + H)^+$.

***N*-Benzyl-2-ethoxy-1-(2'-fluoro-4-biphenylylmethyl)-7-(1*H*)-benzimidazolecarboxamide (12b)**
To a solution of *N*-benzyl-2-ethoxy-1-(4-iodobenzyl)-7-(1*H*)-benzimidazolecarboxamide (51 mg, 0.10 mmol) in DMF (1 mL) and 1,4-dioxane (5 mL) was added $Pd(OAc)_2$ (9 mg, 0.04 mmol), Sphos (33 mg, 0.080 mmol), K_3PO_4 (85 mg, 0.40 mmol) and *o*-fluorophenylboronic acid (30 mg, 0.21 mmol). After being stirred at 85℃ for 8 h under Ar atmosphere, the reaction mixture was evaporated under reduced pressure. The residue was suspended in H_2O and was extracted with CH_2Cl_2. The organic phase was dried over sodium sulfate and evaporated under reduced pressure. The residue was purified by silica gel column chromatography (hexane : AcOEt = 3 : 2), followed by recrystallization from AcOEt/hexane to afford the title compound (13 mg, 0.027 mmol) as colourless solid in 27% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.98 (1H, t, *J* = 5.7 Hz), 7.56 (1H, d, *J* = 8.0 Hz), 7.45-7.38 (4H, m), 7.30-7.18 (8H, m), 7.14 (1H, t, *J* = 8.0 Hz), 7.03 (2H, d, *J* = 8.0 Hz), 5.47 (2H, s), 4.60 (2H, q, *J* = 6.9 Hz), 4.38 (2H, d, *J* = 5.7 Hz), 1.40 (3H, t, *J* = 6.9 Hz). HPLC: 97% purity, RT 3.53 min. ES-MS (*m/z*): 480 $(M + H)^+$.

***N*-Benzyl-2-ethoxy-1-(2'-trifluoromethyl-4-biphenylylmethyl)-7-(1*H*)-benzimidazolecarboxamide (12c)**: prepared from *N*-benzyl-2-ethoxy-1-(4-iodobenzyl)-7-(1*H*)-benzimidazolecarboxamide and *o*-trifluoromethylphenylboronic acid at 100℃ by using Xphos instead of Sphos according to the synthesis of **12b** as colourless solid in 54% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.98 (1H, t, *J* = 6.3 Hz), 7.79 (1H, d, *J* = 7.4 Hz), 7.68 (1H, d, *J* = 7.4 Hz), 7.60-7.55 (2H, m), 7.30-7.14 (10H, m), 7.01 (2H, d, *J* = 8.0 Hz), 5.47 (2H, s), 4.59 (2H, q, *J* = 6.9 Hz), 4.36 (2H, d, *J* = 6.3 Hz), 1.37 (3H, t, *J* = 6.9 Hz). HPLC: 99% purity, RT 3.72 min. ES-MS (*m/z*): 530 $(M + H)^+$.

***N*-Benzyl-2-ethoxy-1-(2'-formyl-4-biphenylylmethyl)-7-(1*H*)-benzimidazolecarboxamide (12d)**: prepared from *N*-benzyl-2-ethoxy-1-(4-iodobenzyl)-7-(1*H*)-benzimidazolecarboxamide and *o*-formylphenylboronic acid according to the synthesis of **12b** as colourless solid in 25% yield. ^{1}H NMR (500MHz, $CDCl_3$) δ: 9.86 (1H, s), 7.99 (1H, dd, *J* = 8.0 Hz, 1.1 Hz), 7.66 (1H, dd, *J* = 8.0 Hz, 1.1 Hz), 7.60 (1H, ddd, *J* = 8.6 Hz, 7.4 Hz, 1.1 Hz), 7.49 (1H, t, *J* = 8.0 Hz), 7.34 (1H, d, *J* = 8.0 Hz), 7.25-7.11 (11H, m), 6.19 (1H, t, *J* = 5.2 Hz), 5.63 (2H, s), 4.69 (2H, q, *J* = 6.9 Hz), 4.44 (2H, d, *J* = 5.2 Hz), 1.50 (3H, t, *J* = 6.9 Hz). HPLC: 98% purity, RT 3.32 min. ES-MS (*m/z*): 490 $(M + H)^+$.

***N*-Benzyl-2-ethoxy-1-(2'-hydroxy-4-biphenylylmethyl)-7-(1*H*)-benzimidazolecarboxamide (12e)**: prepared from *N*-benzyl-2-ethoxy-1-(4-iodobenzyl)-7-(1*H*)-benzimidazolecarboxamide and *o*-hydroxyphenylboronic acid according to the synthesis of **12b** as colourless solid in 32% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 9.49 (1H, s), 9.00 (1H, t, *J* = 5.7 Hz), 7.55 (1H, d, *J* = 8.0 Hz), 7.38 (2H, d, *J* = 8.6 Hz), 7.26-7.25 (4H, m), 7.22-7.11 (5H, m), 6.97 (2H, d, *J* = 8.6 Hz), 6.90 (1H, d, *J* = 8.0 Hz), 6.83 (1H, t, *J* = 8.0 Hz), 5.55 (2H, s), 4.60 (2H, q, *J* = 6.9 Hz), 4.41 (2H, d, *J* = 5.7 Hz), 1.42 (3H, t, *J* = 6.9 Hz). HPLC: 98% purity, RT 3.15 min. ES-MS (*m/z*): 478 $(M + H)^+$.

***N*-Benzyl-1-(2'-carboxy-4-biphenylylmethyl)-2-ethoxy-7-(1*H*)-benzimidazolecarboxamide (12f)**: prepared from *N*-benzyl-2-ethoxy-1-(4-iodobenzyl)-7-(1*H*)-benzimidazolecarboxamide and *o*-carboxyphenylboronic acid by using Xphos instead of Sphos according to the synthesis of **12b** as colourless solid in 17% yield. ^{1}H NMR (500MHz, $CDCl_3$) δ: 7.81 (1H, d, *J* = 7.4 Hz), 7.56-7.52 (2H,m), 7.42 (1H, ddd, *J* = 8.6 Hz, 7.4 Hz, 1.1 Hz), 7.32 (1H, d, *J* = 7.4 Hz), 7.23-7.17 (7H, m), 6.97-6.95 (3H, m), 6.86 (1H, t, *J* = 8.0 Hz), 6.19 (1H, brs), 5.53 (2H, s), 4.51 (2H, q, *J* = 6.9 Hz), 4.33 (2H, d, *J* = 5.7 Hz), 1.40 (3H, t, *J* = 6.9 Hz). HPLC: >99% purity, RT 3.32 min.

ES-MS (*m/z*): 506 $(M + H)^+$.

***N*-Benzyl-1-(3'-cyano-4-biphenylylmethyl)-2-ethoxy-7-(1*H*)-benzimidazolecarboxamide**: prepared from *N*-benzyl-2-ethoxy-1-(4-iodobenzyl)-7-(1*H*)-benzimidazolecarboxamide and *m*-cyanophenylboronic acid according to the synthesis of **12b** as colourless solid in 81% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.94 (1H, t, J = 5.7 Hz), 8.06 (1H, s), 7.94 (1H, d, J = 8.0 Hz), 7.80 (1H, d, J = 8.0 Hz), 7.64 (1H, t, J = 8.0 Hz), 7.59-7.55 (3H, m), 7.24-7.18 (6H, m), 7.14 (1H, t, J = 8.0 Hz), 7.04 (2H, d, J = 8.6 Hz), 5.47 (2H, s), 4.59 (2H, q, J = 6.9 Hz), 4.38 (2H, d, J = 5.7 Hz), 1.40 (3H, t, J = 6.9 Hz). ES-MS (*m/z*): 487 $(M + H)^+$.

***N*-Benzyl-2-ethoxy-1-[3'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (12g)**
To a solution of **19h** (43 mg, 0.089 mmol) in DMF (5 mL) was added tri-*n*-butyltin chloride (120 μL, 0.44 mmol) and NaN_3 (29 mg, 0.45 mmol). After being stirred at 130℃ for 48 h under Ar atmosphere, H_2O was added. The mixture was extracted with CH_2Cl_2 and the organic phase was dried over sodium sulphate, followed by evaporation under reduced pressure. The residue was purified by silica gel column chromatography (CH_2Cl_2 : MeOH = 100 : 5), recrystallization from CH_2Cl_2/hexane and reslurry from acetone to afford the title compound (12 mg, 0.023 mmol) as colourless solid in 26% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.96 (1H, t, J = 6.3 Hz), 8.25 (1H, s), 8.00 (1H, d, J = 8.0 Hz), 7.77 (1H, d, J = 8.0 Hz), 7.65 (1H, t, J = 8.0 Hz), 7.58-7.55 (3H, m), 7.25-7.06 (9H, m), 5.48 (2H, s), 4.61 (2H, q, J = 6.9 Hz), 4.40 (2H, d, J = 6.3 Hz), 1.41 (3H, t, J = 6.9 Hz). HPLC: >99% purity, RT 2.83 min. ES-MS (*m/z*): 530 $(M + H)^+$.

***N*-Benzyl-1-[4'-cyano-4-biphenylylmethyl]-2-ethoxy-7-(1*H*)-benzimidazolecarboxamide**: prepared from *N*-benzyl-2-ethoxy-1-(4-iodobenzyl)-7-(1*H*)-benzimidazolecarboxamide and *p*-cyanophenylboronic acid at 100℃ by using Xphos instead of Sphos according to the synthesis of **12b** as colourless solid in 68% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.94 (1H, t, J = 6.3 Hz), 7.89 (2H, d, J = 8.6 Hz), 7.79 (2H, d, J = 8.6 Hz), 7.59-7.55 (3H, m), 7.23-7.18 (6H, m), 7.14 (1H, t, J = 8.0 Hz), 7.05 (2H, d, J = 8.0 Hz), 5.48 (2H, s), 4.59 (2H, q, J = 6.9 Hz), 4.37 (2H, d, J = 6.3 Hz), 1.39 (3H, t, J = 6.9 Hz). ES-MS (*m/z*): 487 $(M + H)^+$.

***N*-Benzyl-2-ethoxy-1-[4'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (12h)**: prepared from *N*-benzyl-1-[4'-cyano-4-biphenylylmethyl]-2-ethoxy-7-(1*H*)-benzimidazolecarboxamide according to the synthesis of **12g** as colourless solid in 22% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.96 (1H, t, J = 6.3 Hz), 8.10 (2H, d, J = 8.0 Hz), 7.83 (2H, d, J = 8.0 Hz), 7.60 (2H, d, J = 8.0 Hz), 7.56 (1H, d, J = 8.0 Hz), 7.24-7.19 (6H, m), 7.14 (1H, t, J = 8.0 Hz), 7.06 (2H, d, J = 8.0 Hz), 5.48 (2H, s), 4.60 (2H, q, J = 7.4 Hz), 4.40 (2H, d, J = 6.3 Hz), 1.41 (3H, t, J = 7.4 Hz). HPLC: 97% purity, RT 2.80 min. ES-MS (*m/z*): 530 $(M + H)^+$.

3-Nitro-2-[2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]aminobenzoic acid
To a solution of methyl 2-trifluoroacetylamino-3-nitrobenzoate (2.25 g, 7.70 mmol) in CH_3CN (250 mL) was added K_2CO_3 (1.28 g, 9.25 mmol) and commercially available 2'-(1-trityl-5-(1*H*)-tetrazolyl)biphenylylmethyl bromide (4.87 g, 8.47 mmol). After being refluxed overnight under Ar atmosphere, the reaction mixture was filtered and the filtrate was evaporated under reduced pressure. The residue was suspended in brine, followed by extraction with CH_2Cl_2. The organic phase was dried over sodium sulfate and evaporated under reduced pressure. The residue was dissolved in THF (150 mL) and 4 mol/L NaOH*aq.* (39 mL) and H_2O (50 mL) was added therein. After being stirred at room temperature for 20 h, the reaction mixture was evaporated under reduced pressure. 5 mol/L HCl*aq* (31.2 mL) was added and the resulting precipitate was filtered to afford the title compound.

***N*-Benzyl-3-nitro-2-{2'-[1-trityl-5-(1*H*-tetrazolyl)]-4-biphenylylmethylamino}benzamide**: prepared from 3-nitro-2-[2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]aminobenzoic acid

according to the synthesis of compound **12a** as yellow solid in 72% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 9.17 (1H, t, J = 6.3 Hz), 8.37 (1H, t, J = 5.2 Hz), 8.06 (1H, dd, J = 8.6 Hz, 1.7 Hz), 7.89 (1H, dd, J = 7.4 Hz, 1.1 Hz), 7.70 (1H, dd, J = 7.4 Hz, 1.1 Hz), 7.63-7.52 (2H, m), 7.45 (1H, d, J = 8.6 Hz), 7.32-7.25 (13H, m), 7.18 (1H, t, J = 7.4 Hz), 7.03 (2H, d, J = 8.6 Hz), 7.00 (2H, d, J = 8.6 Hz), 6.84-6.81 (7H, m), 4.41 (2H, d, J = 6.3 Hz), 4.12 (2H, d, J = 5.2 Hz).

***N*-Benzyl-1-[2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide**

To an ice cooled solution of sodium hydrosulfite (972 mg, 5.58 mmol) in H_2O (12 mL) was added dropwise a solution of *N*-benzyl-3-nitro-2-{2'-[1-trityl-5-(1*H*-tetrazolyl)]-4-biphenylylmethylamino}benzamide (261 mg, 0.35 mmol) in THF (10 mL) and EtOH (5 mL). After being stirred at room temperature for 2.5 h under Ar atmosphere, sat. $NaHCO_3$*aq.* was added, followed by extraction with CH_2Cl_2. The organic phase was dried over sodium sulfate and evaporated under reduced pressure. The residue was dissolved in $CH(OMe)_3$ (14 mL) and *p*-TsOH (7 mg, 0.035 mmol), followed by stirring at room temperature overnight under Ar atmosphere. sat. $NaHCO_3$*aq.* was added to the reaction mixture and the mixture was extracted with CH_2Cl_2. The organic phase was dried over sodium sulfate and evaporated under reduced pressure. The residue was purified by silica gel column chromatography (hexane : AcOEt = 1 : 3) to afford the title compound (145 mg, 0.20 mmol) in 57% yield. ^{1}H NMR (500MHz, $CDCl_3$) δ: 8.02 (1H, s), 7.81 (1H, dd, J = 8.0 Hz, 1.1 Hz), 7.77 (1H, dd, J = 8.0 Hz, 1.1 Hz), 7.49 (1H, ddd, J = 8.0 Hz, 7.4 Hz, 1.1 Hz), 7.43 (1H, ddd, J = 8.0 Hz, 7.4 Hz, 1.1 Hz), 7.36 (1H, dd, J = 7.4 Hz, 1.1 Hz), 7.33-7.30 (3H, m), 7.23-7.13 (11H, m), 6.98 (1H, t, J = 8.0 Hz), 6.94-6.87 (9H, m), 6.67 (2H, d, J = 8.0 Hz), 6.44 (1H, t, J = 6.3 Hz), 5.62 (2H, s), 4.37 (2H, d, J = 6.3 Hz). ES-MS (*m/z*): 728 $(M + H)^+$.

***N*-Benzyl-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (15a)**

To a solution of *N*-benzyl-1-[2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (75 mg, 0.10 mmol) in EtOH (3 mL) was added boric acid (3.4 mg, 0.055 mmol). After being refluxed for 7.5 h under Ar atmosphere, the reaction mixture was evaporated under reduced pressure. The residue was purified by recrystalization from MeOH to afford the title compound (21 mg, 0.042 mmol) as colourless solid in 41% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.94 (1H, t, J = 6.3 Hz), 8.50 (1H, s), 7.80 (1H, dd, J = 8.0 Hz, 1.1 Hz), 7.67-7.64 (2H, m), 7.55 (1H, m), 7.44 (1H, d, J = 8.0 Hz), 7.37 (1H, d, J = 8.0 Hz), 7.31-7.22 (6H, m), 6.94 (2H, d, J = 8.0 Hz), 6.90 (2H, d, J = 8.0 Hz), 5.64 (2H, s), 4.36 (2H, d, J = 6.3 Hz). HPLC: >99% purity, RT 2.28 min. ES-MS (*m/z*): 486 $(M + H)^+$.

***N*-Benzyl-2-hydroxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (15b)**

To a solution of compound **5l** (200 mg, 0.38 mmol) in 1,4-dioxane (6 mL) was added concd. HCl (1.5 mL). After being stirred at room temperature overnight under Ar atmosphere, the reaction mixture was evaporated under reduced pressure. Addition of H_2O, followed by filtration of the precipitate and reslurry from H_2O afforded the title compound (341 mg, 0.68 mmol) in quantitative yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 11.28 (1H, s), 8.78 (1H, t, J = 6.3 Hz), 7.67-7.63 (2H, m), 7.54 (1H, t, J = 7.4 Hz), 7.46 (1H, d, J = 7.4 Hz), 7.25-7.20 (5H, m), 7.11 (1H, dd, J = 7.4 Hz, 1.7 Hz), 7.05-7.01 (2H, m), 6.93 (4H, m), 5.17 (2H, s), 4.28 (2H, d, J = 6.3 Hz). HPLC: >99% purity, RT 2.47 min. ES-MS (*m/z*): 502 $(M + H)^+$.

***N*-Benzyl-2-methoxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (15c)**: prepared from *N*-benzyl-3-nitro-2-{2'-[1-trityl-5-(1*H*-tetrazolyl)]-4-biphenylyl-methylamino}benzamide and tetramethoxy orthocarbonate via *N*-benzyl-2-methoxy-1-[2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide according to the synthesis of **15a** as colourless solid in 15% yield (in 2 steps). ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.93 (1H, t, J = 6.3 Hz), 7.64 (1H, d, J = 8.0 Hz), 7.63 (1H, d, J = 8.0 Hz), 7.56 (1H, dd, J = 8.0 Hz, 1.1 Hz), 7.54 (1H, d, J = 8.0 Hz), 7.44 (1H, d, J = 8.0 Hz),

7.25-7.18 (6H, m), 7.14 (1H, t, *J* = 8.0 Hz), 6.93 (2H, d, *J* = 8.0 Hz), 6.87 (2H, d, *J* = 8.0 Hz), 5.40 (2H, s), 4.33 (2H, d, *J* = 6.3 Hz), 4.14 (3H, s). HPLC: 95% purity, RT 2.70 min. ES-MS (*m/z*): 516 $(M + H)^+$.

***N*-Benzyl-2-propoxy-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazole-carboxamide (15d)**: prepared from *N*-benzyl-3-nitro-2-{2'-[1-trityl-5-(1*H*-tetrazolyl)]-4-biphenylyl-methylamino}benzamide and tetrapropoxy orthocarbonate via *N*-benzyl-2-propoxy-1-2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarbox-amide according to the synthesis of **15a** as colourless solid in 28% yield (in 2 steps). ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.95 (1H, t, *J* = 5.7 Hz), 7.65 (1H, d, *J* = 7.4 Hz), 7.63 (1H, d, *J* = 7.4 Hz), 7.56-7.53 (2H, m), 7.44 (1H, d, *J* = 8.0 Hz), 7.28-7.20 (6H, m), 7.13 (1H, t, *J* = 7.4 Hz), 6.93 (2H, d, *J* = 8.6 Hz), 6.89 (2H, d, *J* = 8.6 Hz), 5.39 (2H, s), 4.46 (2H, t, *J* = 6.9 Hz), 4.36 (2H, d, *J* = 5.7 Hz), 1.76 (2H, m), 0.91 (3H, t, *J* = 6.9 Hz). HPLC: 98% purity, RT 2.98 min. ES-MS (*m/z*): 544 $(M + H)^+$.

***N*-Benzyl-2-mercapto-1-[2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benz-imidazolecarboxamide**: prepared from *N*-benzyl-3-nitro-2-{2'-[1-trityl-5-(1*H*-tetrazolyl)]-4-biphenylylmethylamino}- benzamide and TCDI by using THF instead of AcOH according to the synthesis of *N*-benzyl-1-[2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide in 30% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 13.13 (1H, s), 8.66 (1H, t, *J* = 5.3 Hz), 7.75 (1H, d, *J* = 7.4 Hz), 7.58 (1H, t, *J* = 7.4 Hz), 7.51 (1H, t, *J* = 7.4 Hz), 7.37-7.30 (11H, m), 7.25-7.16 (7H, m), 6.96-6.86 (10H, m), 5.77 (2H, s), 4.26 (2H, d, *J* = 4.9 Hz). ES-MS (*m/z*): 518 (M + H−trityl)$^+$.

***N*-Benzyl-2-mercapto-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazole-carboxamide (15e)**: prepared from *N*-benzyl-2-mercapto-1-[2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide according to the synthesis of **15a** as colourless solid in 74% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 13.10 (1H, s), 8.67 (1H, t, *J* = 5.9 Hz), 7.66-7.63 (2H, m), 7.54 (1H, t, *J* = 8.0 Hz), 7.45 (1H, d, *J* = 8.0 Hz), 7.35 (1H, dd, *J* = 8.6 Hz, 1.7 Hz), 7.24-7.20 (7H, m), 6.96 (4H, m), 5.75 (2H, s), 4.26 (2H, d, *J* = 5.9 Hz). HPLC: 99% purity, RT 2.55 min. ES-MS (*m/z*): 518 $(M + H)^+$.

***N*-Benzyl-2-ethylthio-1-[2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benz-imidazolecarboxamide**

To an ice cooled solution of *N*-benzyl-2-mercapto-1-[2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide (89 mg, 0.12 mmol) and K_2CO_3 (24 mg, 0.18 mmol) in DMF (10 mL) was added iodoethane (10 μL, 0.13 mmol). After being stirred at room temperature for 1 h under Ar atmosphere, the reaction mixture was evaporated under reduced pressure. The residue was suspended in H_2O, followed by extraction with CH_2Cl_2. The organic phase was dried over sodium sulfate and evaporated under reduced pressure to afford the title compound (97.7 mg, 0.117 mmol) as colourless solid in quantitative yield. ^{1}H NMR (500MHz, $CDCl_3$) δ: 7.76 (1H, d, *J* = 8.0 Hz), 7.70 (1H, d, *J* = 8.0 Hz), 7.49 (1H, ddd, *J* = 8.6 Hz, 7.4 Hz, 1.1 Hz), 7.43 (1H, ddd, *J* = 8.6 Hz, 7.4 Hz, 1.1 Hz), 7.36 (1H, d, *J* = 8.0 Hz), 7.32-7.29 (3H, m), 7.20-7.17 (9H, m), 7.11-7.10 (2H, m), 6.93 (2H, d, *J* = 8.0 Hz), 6.91-6.88 (7H, m), 6.78 (1H, d, *J* = 8.0 Hz), 6.71 (2H, d, *J* = 8.60 Hz), 6.27 (1H, t, *J* = 6.3 Hz), 5.62 (2H, s), 4.33 (2H, d, *J* = 6.3 Hz), 3.44 (2H, q, *J* = 7.4 Hz), 1.48 (3H, t, *J* = 7.4 Hz). ES-MS (*m/z*): 788 $(M + H)^+$.

***N*-Benzyl-2-ethylthio-1-[2'-(5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazole-carboxamide (15f)**: prepared from *N*-benzyl-2-ethylthio-1-[2'-(1-trityl-5-(1*H*)-tetrazolyl)-4-biphenylylmethyl]-7-(1*H*)-benzimidazolecarboxamide according to the synthesis of **15a** as colourless solid in 61% yield. ^{1}H NMR (500MHz, DMSO-d_6) δ: 8.95 (1H, t, *J* = 6.3 Hz), 7.72 (1H, dd, *J* = 8.0 Hz, 1.1 Hz), 7.68-7.64 (2H, m), 7.56 (1H, ddd, *J* = 8.6 Hz, 7.4 Hz, 1.1 Hz), 7.46 (1H, d, *J* = 8.0 Hz), 7.28 (1H, d, *J* = 8.0 Hz), 7.25-7.20 (6H, m), 6.98 (2H, d, *J* = 8.6 Hz), 6.86

(2H, d, *J* = 8.6 Hz), 5.44 (2H, s), 4.32 (2H, d, *J* = 6.3 Hz), 3.46 (2H, q, *J* = 7.4 Hz), 1.39 (3H, t, *J* = 7.4 Hz). HPLC: 96% purity, RT 2.82 min. ES-MS (*m/z*): 546 $(M + H)^+$.

2. Molecular modelling

The geometry optimization and electrostatic potential calculation of the derivatives were performed using Gaussian 03 with the B3LYP hybrid functional and the 6-31+G(d) basis set. The restrained electrostatic potential charge fitting of the electrostatic potentials of the optimized structures was carried out with the antechamber module in AMBER9. The crystal structure (ID: 2d0t) of IDO was obtained from PDB. All water molecules and buffer solutes were stripped from the coordinates of the crystal structure, and then hydrogen atoms were added. A grid of 37.5 Å x 37.5 Å x 37.5 Å with 0.375 Å spacing was calculated using AutoGrid. Automated ligand-flexible molecular docking calculations were performed and analyzed using Autodock 4.2 and AutoDockTools. One hundred docking runs for every compound were performed. Each docking calculation consisted of 25 million energy evaluations (long mode) using the Lamarckian genetic algorithm method. Maximum number of generations was set to 270,000. All the other parameters were set by default.

3. Biological evaluations

rhIDO inhibition

rhIDO activity was determined as follow. In brief, the standard reaction mixture (200 µL) contained 50 mM KPB (pH 6.5), 20 mM ascorbic acid (neutralized with NaOH and HCl), 100 µg/mL catalase, 10 µM methylene blue, 200 mM L-tryptophan, 5 nM rhIDO, and DMSO solution of the compound (4 µL). The reaction was carried out at 37°C for 60 min and stopped by the addition of 40 µL of 30% (w/v) CCl_3COOH. After heating at 50°C for 15 min, the reaction mixture was centrifuged at 15000*g* for 5 min. The supernatant (150 µL) was transferred into a well of a 96-well microplate and mixed with 150 µL of 2% (w/v) *p*-dimethylaminobenzaldehyde in acetic acid. The yellow pigment derived from kynurenine was measured at 480 nm using a SPECTRAmax M5SK microplate reader (Molecular Devices). IC_{50} values were calculated from dose-response curves obtained in triplicate experiments.

Kynurenine production in A431 cells

A431 cells (2.0×10^5 cells/mL) were seeded in a 96-well culture plate (100 µL/well) and grown overnight. Serial DMSO dilutions of compounds (10 µL) in a total volume of 200 µL culture medium including tryptophan and human IFN-γ (5 ng/mL final concentration) per well were added into wells containing the cells. After an additional 24 h of incubation, 200 µL/well of a mixed solution of 7% (v/v) aqueous CCl_3COOH and 2% (w/v) *p*-dimethylaminobenzaldehyde in acetic acid (2:5) were added into each well. The yellow colour derived from kynurenine was measured at 480 nm using a SPECTRAmax M5SK microplate reader (Molecular Devices). IC_{50} values were calculated from dose-response curves obtained in triplicate experiments.