Supporting Information for manuscript entitled,

# '2-Amino-5-arylbenzoxazole derivatives as potent inhibitors of fatty acid amide hydrolase (FAAH)'

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**Contents**:

Electronic Supplementary Material (ESI) for Medicinal Chemistry Communications


## General:

All reagents were purchased from commercial sources and used without further purification. All nmr spectra were recorded on a Brüker NMR Spectrometer and are quoted in ppm relative to a tetramethylsilane internal standard, or by referencing on the chemical shift of the deuterated solvent. Purity of final compounds was ascertained by high-performance liquid chromatography using at least two different methods and machines for analysis (HPLC system with conditions shown below and a prior run on an LCMS System using either a 6.5 min, 6 min or 2 min runtime).

**HPLC Conditions A**:

**Sample Preparation:**
Samples are dissolved in methanol at a concentration of 0.1 to 1.0 mg/ml.
**Injection:**
20ul by Autosampler. If this results in a peak greater than 2AUFS, a smaller injection volume was used.
**Pump Conditions:**
Flow rate: 2ml/min
Eluant A: 25mM Potassium Acetate, pH 4.0, in Water.
Eluant B: Acetonitrile
**Gradient:**

| Time | %B |
|---|---|
| | |
| 0.0 | 5 |
| 1.0 | 5 |
| 15.0 | 95 |
| 16.0 | 95 |
| 16.3 | 5 |
| 19.0 | 5 |

**Column:**
Manufacturer: Phenomenex (Phone (310) 212-0555)
Phase: Gemini C18 5um. 150 x 4.6mm + guard
Manufacturers P/N: 00F-4435-E0
**Detection:**
UV at 254nm, 2.0AUFS

**LCMS Systems**:
Vendor: Applied Biosystems
Instrument: API150EX
Features: Single Quadrapole spectrometer with Shimadzu front end
Software: Analyst (ABI)

- 6.5 or 6 minute run on 50mm column, or
- 2 minute run on 20mm cartridge column with 3μM packing
- Simultaneous MS and UV detection
- ELSD available (used for non-UV absorbing compounds)
- LCMS systems used for reaction monitoring and compound submission

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# ASSAYS

## Assay protocol for measurement of $IC_{50}$ values

HEK293-TRex cells (Invitrogen) stably transfected with hFAAH or rFAAH in the pCDNA5-Tet-off vector were used for microsomal enzyme preparation. For homogenization, the cell pellets are thawed on ice at room temperature and resuspended in homogenization buffer (50 mM HEPES (pH 7.4), 1 mM EDTA, 1 μM Pepstatin A, 100 μM Leupeptin, 0.1 mg/mL aprotinin). Cell suspensions are then homogenized on ice using the Polytron 1200C at setting 6 for three 30-second intervals with 30-second rests. The suspension is centrifuged at 1000g for 10 minutes at 4°C and the supernatant is collected and further centrifuged at 24,000 rpm for 30 minutes at 4°C using an ultracentrifuge. Pellets are resuspended by adding in cold microsomal buffer (50 mM HEPES (pH 7.4) and 1 mM EDTA) and sheared through a 23-gauge needle five times, keeping the suspension on ice. Protein concentrations are determined using the BCA assay and aliquoted preparations are stored at -80°C until needed. Compound potency against hFAAH or rFAAH is determined using an enzymatic assay with a fluorescence readout. Briefly, experiments were carried out in a 96-well plate format (Corning Costar, # 3370) with a total well volume of 160 μL with components added in the following order: assay buffer (50 mM HEPES (pH 7.4), 1 mM EDTA, 1.4 mg/mL BSA), compound solutions (7 different concentrations per compound in duplicate), microsomal enzyme preparation (10 μg per well) and substrate [AA-AMC (arachadonyl 7-amino 4-methyl coumarin amide), 2 μM]. After a brief shaking, a kinetic read of the plate is obtained using a Tecan Safire II in kinetic mode for 275 cycles with excitation and emission wavelengths of 355 and 460 nm, respectively. Raw data obtained between 2,000 and

5,000 seconds is then processed and analyzed using Assay Explorer and GraphPad Prism. URB-597 (**3**) was used as a control and displayed an average $IC_{50}$ of 3 nM under these conditions.

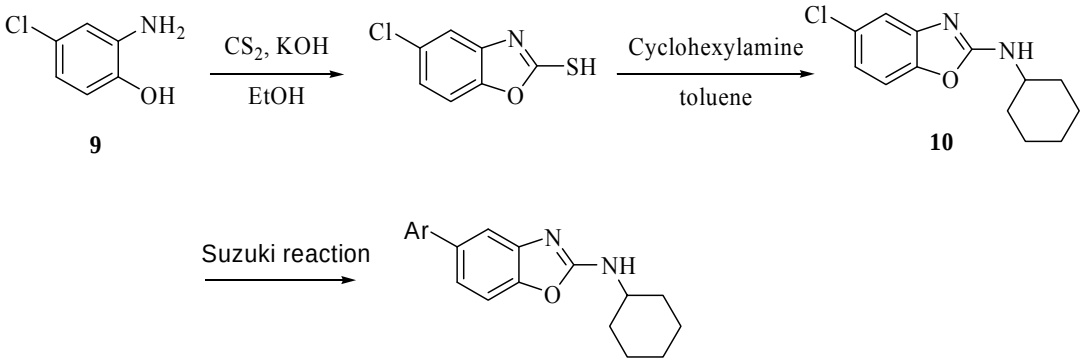
**Notes on biochemical experiments in Figure 3**:

Biochemical experiments shown in Figure 3 were conducted using a microsomal preparation of full length hFAAH expressed in mammalian cells coupled with a radioactive enzymatic assay measuring the hydrolysis of $^{3}$H-anandamide. Incubation time between enzyme and inhibitor was lengthened from 0h to 3h (assay duration: 15 minutes from substrate addition to reaction quenching).

## Synthesis of intermediates detailed in Schemes 1-5

## and General Procedures to Final Compounds:

**Scheme 1**:




**5-Chlorobenzo[*d*]oxazole-2-thiol**:

To a stirred solution of 2-amino-4-chlorophenol **9** (20.0 g, 139.3 mmol) in EtOH (500 mL) was added KOH (18.36 g, 327.3 mmol) followed by $CS_2$ (126.7 mL, 1318 mmol) and refluxed for 4 h. Volatiles were removed and the residue was diluted with water (100 mL) and acidified with 2 M HCl (50 mL) and extracted with $CH_2Cl_2$ (2 × 250 mL). The organic phase was dried over anhydrous $Na_2SO_4$ and concentrated under reduced pressure to give 23.5 g (91%) of the title compound as brown solid. $^1$H NMR (400 MHz, MeOD) δ 7.35-7.33 (m, 1H), 7.25-7.22 (m, 2H), 3.31-3.30 (m, 1H); MS: ESI *m/z* 186 [M + H]$^+$.

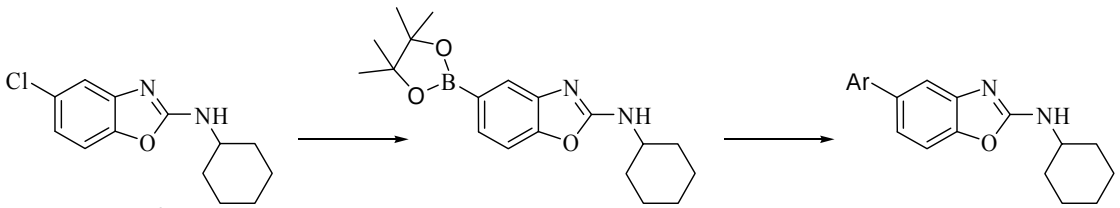
**Preparation of 5-chloro-*N*-cyclohexylbenzo[*d*]oxazol-2-amine 10**:

To the suspension of 5-Chlorobenzo[*d*]oxazole-2-thiol (9.40 g, 50.64 mmol) in toluene (120 mL) was added cyclohexylamine (11.5 mL, 101 mmol) and refluxed for 48 h. The solvent was evaporated and residue was purified by silica gel column chromatography (gradient: hexanes to 30% ethyl acetate in hexanes) to afford the title compound (7.00 g, 74%) as a light orange solid. $^1$H NMR (400 MHz, $CDCl_3$) δ 7.31 (s, 1H), 7.13 (d, *J* = 12

Hz, 1H), 6.99-6.97 (m, 1H), 5.08 (bs, 1H), 3.78-3.70 (m, 1H), 2.14-2.11 (m, 2H), 1.81-1.76(m, 2H), 1.65-1.57 (m, 2H), 1.49-1.31 (m, 2H), 1.29-1.28 (m, 2H); MS: ESI *m/z* 251 $[M + H]^+$.

The intermediate **10** was used in **General Procedure A** for synthesis of final compounds

**Scheme 2**:



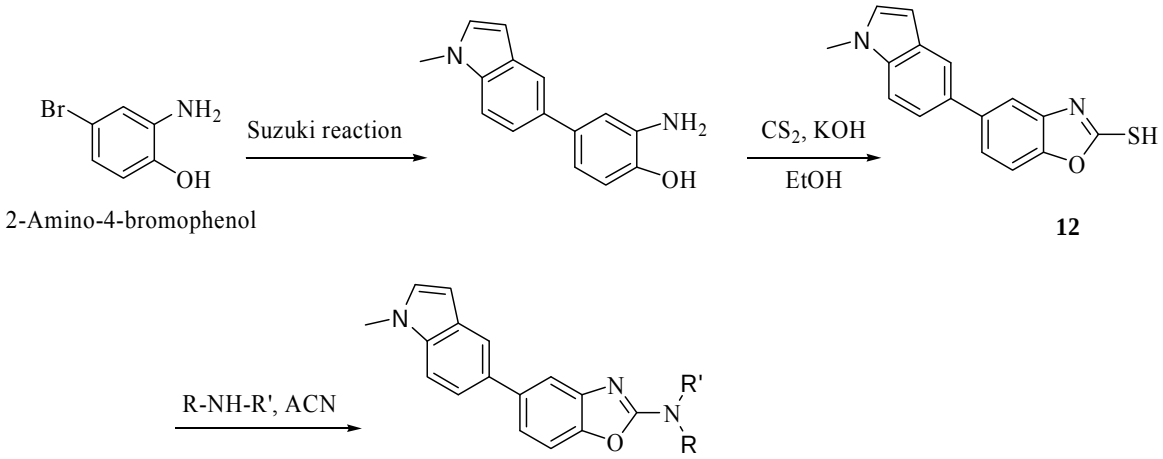

**Preparation of *N*-cyclohexyl-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)benzo[*d*]oxazol-2-amine 11:**

A 100 mL flask was charged with bis(dibenzylideneacetone)palladium(0) (0.325 g, 0.565 mmol) and tricyclohexylphosphine (0.400 g, 1.43 mmol), sealed, evacuated and purged with nitrogen. The mixture was charged with 1,4-dioxane (43 mL) and stirred at room temperature for 30 minutes to obtain a clear deep red solution. A mixture of bis(pinacolato)diboron (2.02 g, 7.98 mmol), 5-chloro-*N*-cyclohexylbenzo[*d*]oxazol-2-amine (2.00 g, 7.98 mmol), and potassium acetate (1.17 g, 12.0 mmol) were added and the mixture was heated at 80 °C for 65 hours. The reaction was quenched with water (1 mL) then filtered through celite and the filter cake was washed with EtOAc (2 x 75 mL) and water (75 mL). Volatiles were removed *in vacuo* and the residue was dissolved in EtOAc (150 mL) and washed with water (50 mL) followed by brine (50 mL), dried ($Na_2SO_4$), filtered and evaporated. The product was purified by silica gel chromatography

(0 to 30% EtOAc in hexanes) to obtain the title compound **11** (1.44 g, 53%) as a solid; *m/z* = 343.1 $(M + H)^+$. $^1H$ NMR (400 MHz; $CDCl_3$) δ 7.79 (s, 1H), 7.52 (dd, *J* = 8.0, 1.0 Hz, 1H), 7.23 (d, *J* = 7.9 Hz, 1H), 4.95 (br. d, *J* = 6.5 Hz, 1H), 3.82-3.69 (m, 1H), 2.17-2.07 (m, 2H), 1.82-1.72 (m, 2H), 1.69-1.60 (m, 1H), 1.51-1.16 (m, 5H), 1.34 (s, 12 H).

The intermediate **11** was used in **General Procedure B** for synthesis of final compounds

**Scheme 3**:




**Preparation 2-Amino-4-(1-methyl-1H-indol-5-yl)-phenol**:

In a 250 mL flask, 2-Amino-4-bromo-phenol (3.22 g, 17.1 mmol), 1-methyl-1*H*-indol-5-ylboronic acid (4.5 g, 26 mmol), Potassium carbonate (9.48 g, 68.6 mmol) and Bis(triphenylphosphine)palladium(II) chloride (602 mg, 0.857 mmol) were added and the flask was degassed using vacuum/$N_2$. The reaction was dissolved in degassed 1,4-Dioxane (30 mL, 400 mmol) and the flask was filed with $N_2$ using vacuum (x2). The reaction mixture was heated for 16 hr at 110°C. The reaction was concentrated *in vacuo*, neutralized to pH 6 with $NaH_2PO_4$ and concentrated in vacuo. The residue was purified

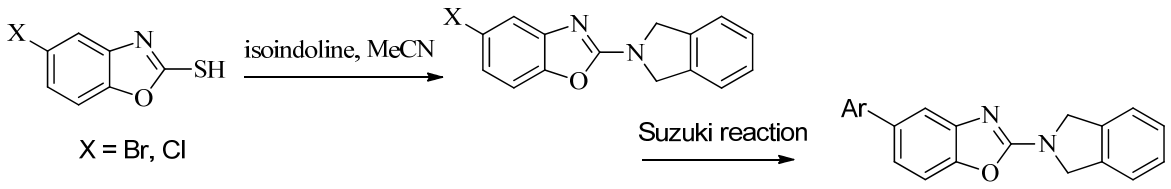
by column chromatography (Hex:EtOAc, 20-60%) to yield the title product (1.07 g, 26 %) as a brown solid. MS: ESI *m/z* 239 [M + H].

**Preparation of 5-(1-Methyl-1*H*-indol-5-yl)-benzoxazole-2-thiol 12**:

To a solution of 2-amino-4-(1-methyl-1H-indol-5-yl)-phenol (700 mg, 3 mmol) in ethanol (10 mL, 200 mmol), Potassium hydroxide (412.0 mg, 7.344 mmol) was added. The reaction mixture was soluble after 2 min and became dark brown. Then, carbon disulfide (1.590 mL, 26.44 mmol) was added and the reaction mixture was heated to reflux for 6h. The reaction mixture was concentrated in vacuo and the residue was dissolved in water and acidified with 12 M of hydrogen chloride in water (0.7344 mL, 8.813 mmol). The precipitate formed was filtered and washed with water to yield the title product (750 mg, 91%) as a grey solid that was used without further purification into the next step. MS: ESI *m/z* 281 [M + H].

The intermediate **12** was used in **General Procedure C** for synthesis of final compounds

**Scheme 4**:

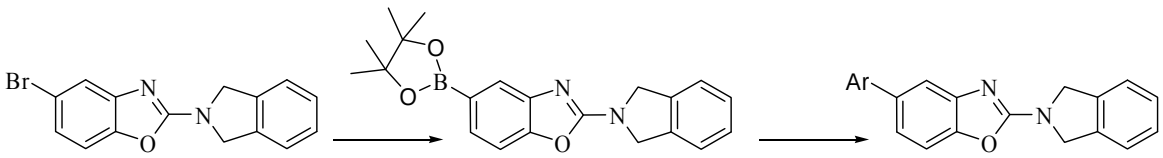



**5-Bromo-2-(1,3-dihydro-isoindol-2-yl)-benzoxazole**

A microwave vial was charged with 5-bromo-benzooxazole-2-thiol (400 mg, 1.74 mmol), isoindoline (590 µL, 5.2 mmol), and acetonitrile (5.0 mL). The mixture was purged with nitrogen then heated in a microwave at 190 °C for 30 min (LCMS indicates product, a

thioamide ($H_2S$ reacted with ACN and the resulting amide reacted with isoindoline) and a debrominated version of the desired product). After allowing to cool, the crude product was presorbed directly onto silica gel (6 g) and the mixture was purified by column chromatography on silica gel (40g Isco cartridge) using hexane to DCM as eluent to give a solid (494 mg; contains about 85% desired product, 10% thioamide and 5% debromination by product). This material was presorbed onto silica gel (4g) and purified by column chromatography (40g Isco cartridge) using hexanes to DCM as eluent to give the product (394 mg, 72%) as a solid. $^1$H NMR (400 MHz, $CDCl_3$) 7.54 (br. d, $J$ = 1.0 Hz, 1H), 7.36 (s, 4H), 7.18 (d, $J$ = 8.4 Hz, 1H), 7.16 (dd, $J$ = 1.7 Hz, J = 8.4 Hz, 1H), 5.02 (s, 4H). LC/MS: 1.47 min on 2 min run; ESI *m/z* 315.3 [M+H].

The above intermediate was used in **General Procedure D** for synthesis of final compounds



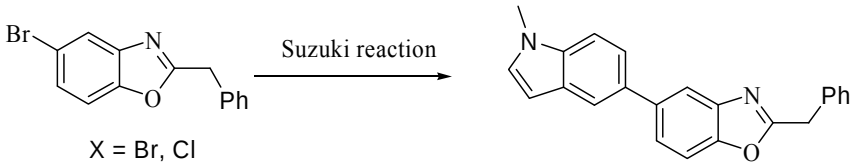

**2-(1,3-Dihydro-isoindol-2-yl)-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-benzoxazole**

To a mixture of 5-bromo-2-(1,3-dihydro-isoindol-2-yl)-benzoxazole (200 mg, 0.63 mmol), bis(pinacolato)diboron (242 mg, 0.953 mmol), KOAc (311 mg, 3.17 mmol), and [1,1'-bis(diphenylphosphino)ferrocene]dichloropalladium(II),complex with DCM (1:1) (51.8 mg, 0.0634 mmol) under nitrogen was added N,N-dimethylformamide (1.3 mL). The mixture was heated under nitrogen at 80 °C overnight. LC/MS at 15 hours indicates complete reaction (about 4:1 boronic ester to boronic acid, but no SM). The mixture was

diluted with EtOAc (20 mL) and washed with brine (20 mL). The aqueous was back extracted with EtOAc (5 mL) and the combined organics washed with brine (2 x 20 mL), dried ($Na_2SO_4$), filtered and concentrated under vacuum. The crude product was presorbed onto silica gel (1.3g) and purified by column chromatography on silica gel (12g Isco cartridge) using hexane to EtOAc as eluent (product elutes with about 20-25% EtOAc) to obtain the desired product (206 mg, 90%) as a solid. $^1$HNMR (400 MHz; $CDCl_3$) 7.87 (br s, 1H), 7.54 (d, *J* = 8.0 Hz, 1H), 7.35 (br s, 4H), 7.31 (d, *J* = 8.0 Hz, 1H), 5.03 (s, 4H), 1.39 (s, 12 H). LC/MS: 1.53 min on 2 min run; ESI *m/z* 363.2 [M+H].

The above intermediate was used in **General Procedure E** for synthesis of final compounds

**Scheme 5**:




**2-Benzyl-5-(1-methyl-1*H*-indol-5-yl)-benzoxazole**

Prepared using General Procedure A below from 2-benzyl-5-bromobenzoxazole (commercial). After workup, the mixture was dissolved in DMSO and purified by HPLC to give the title compound (17 mg, 58%) as a solid. $^1$H-NMR (400 MHz; $d_6$-DMSO) δ 7.91 (d, *J* = 1.3 Hz, 1H), 7.83 (d, *J* = 1.0 Hz, 1H), 7.70 (dd, *J* = 8.5, 0.3 Hz, 1H), 7.64 (dd, *J* = 8.4, 1.8 Hz, 1H), 7.54 (d, *J* = 8.5 Hz, 1H), 7.77 (dd, *J* = 8.5, 1.7 Hz, 1H), 7.41-7.35 (m, 5H), 7.31-7.27 (m, 1H), 6.45 (dd, *J* = 3.0, 0.6 Hz, 1H), 4.36 (s, 2H), 3.81 (s, 3H); LC/MS: 3.91 min; ESI *m/z* 339.3 [M+H]; Purity = 99.9% by HPLC Conditions A

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## General Procedures for Synthesis of Final Compounds

**General procedure A**:

In a 5mL microwave vial, 5-chloro-*N*-cyclohexylbenzo[*d*]oxazol-2-amine (20 mg, 0.08 mmol), the appropriate boronic acid or boronic ester acd (0.12 mmol), $Cs_2CO_3$ (100 mg, 0.3 mmol), $Bu_4NI$ (4 mg, 0.01 mmol) and dihydrogen dichlorobis(di-*tert*-butyl phosphino-*k*P)palladate(II) (1 mg, 0.002 mmol) were dissolved in $H_2O$ (0.2 mL) and DMF (1 mL). The reaction mixture was heated under microwave irradiation at 150 °C for 20 mins. After allowing to cool to room temperature, the mixture was purified directly by preparative high-performance liquid chromatography [acetonitrile:water buffered to pH 10 with eluent used in a gradient system] to give the title compound as a solid.

**General Procedure B**:

To a 5 mL microwave vial was added appropriate halide or pseudohalide (0.06 mmol) and POPd (1.5 mg, 0.003 mmol). A solution of *N*-cyclohexyl-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)benzo[*d*]oxazol-2-amine **11** (10 mg, 0.03 mmol) and tetra-*N*-butylammonium iodide (1.1 mg, 0.003 mmol) in *N*,*N*-dimethylformamide (600 μL), followed by a solution of cesium carbonate (38 mg, 0.12 mmol) in water (150 μL). The system was sealed and purged with nitrogen then irradiated in the microwave for 20 min at 150 °C. LC/MS indicates complete reaction. The product was precipitated out of solution with water (about 2 mL) and buffered to near neutral pH with 1 M $NaH_2PO_4$ (200 μL). The solution was filtered and the residue was dissolved in DMSO (400 μL) and filtered. The vial was rinsed with DMSO (2 x 200 μL) and purified by reverse phase HPLC (acetonitrile:water gradient buffered with 10 mM diethyl amine in aqueous to pH 10).

**General Procedure C**:

A microwave vial was charged with 5-(1-methyl-1*H*-indol-5-yl)-benzoxazole-2-thiol (20 mg, 0.07 mmol), isoindoline (26 mg, 0.21 mmol), and acetonitrile (0.2 mL). The mixture was heated in the microwave at 190 °C for 1 hr. The crude mixture was dissolved in DMSO, filtered and purified by preparative HPLC to yield the title product

**General procedure D:**

A microwave vial was charged with 5-Bromo-2-(1,3-dihydro-isoindol-2-yl)-benzoxazole (15 mg, 0.05 mmol), the boronic acid or boronic ester (0.1 mmol), POPd (2.4 mg, 0.005 mmol), tetra-*N*-butylammonium iodide (2.1 mg, 0.006 mmol) and $Cs_2CO_3$ (62 mg, 0.2 mmol). *N*,*N*-Dimethylformamide (0.7 mL) and water (0.18 mL) was added and the mixture was placed under an atmosphere of nitrogen (air was evacuated x 2). The mixture was heated in the microwave at 150 °C for 20 min. The crude mixture was dissolved in DMSO, filtered and purified by preparative HPLC to yield the title product.

**General Procedure E**:

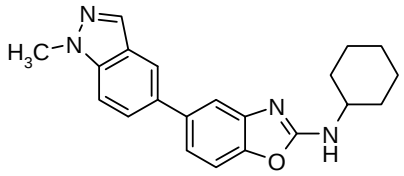
To a small microwave vial was added the appropriate halide (0.044 mmol) and POPd (1.1 mg, 0.0022 mmol). A solution of 2-(1,3-dihydro-isoindol-2-yl)-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-benzoxazole (8.0 mg, 0.022 mmol) and tetra-*N*-butylammonium iodide (0.82 mg, 0.0022 mmol) in *N*,N-dimethylformamide (600 μL) followed by a solution of $Cs_2CO_3$ (29 mg, 0.088 mmol) in Water (150 μL). The system was sealed and purged with nitrogen then heated in the microwave for 20 min at 150 °C. LC/MS indicates complete reaction. The reaction was diluted with EtOAc (5 mL) and washed with brine (5 mL). The aqueous layer was back extracted with EtOAc (about 2 mL) and the combined organics were washed with brine (2 x 5 mL), dried ($Na_2SO_4$), filtered and

evaporated. The crude product was dissolved in DMSO (450 μL) and purified by reverse phase HPLC (acetonitrile in water gradient buffered to pH10 with 10 mM $Et_2NH$ in water) to obtain a the desired product

## Synthesis of key compounds detailed in Tables 1-3

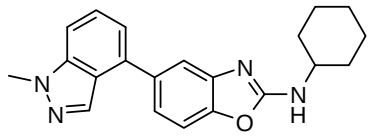
### Preparation of key compounds 23 and 24 in Table 1:

**_N_-cyclohexyl-5-(1-methyl-1_H_-indol-5-yl)benzo[_d_]oxazol-2-amine 23**



The compound was prepared using general procedure A. After completion of the reaction, the mixture was diluted with EtOAc (5 mL) and washed with brine (5 mL). The aqueous layer was back-extracted with EtOAc (2 mL) and the combined organics were washed with brine (2 x 5 mL), dried ($Na_2SO_4$), filtered and evaporated. The crude product was dissolved in DMSO (450 μL) and purified by reverse-phase HPLC (40-95% acetonitrile:water gradient buffered with 10 mM diethylamine in the aqueous to pH 10). The combined pure fractions were concentrated under vacuum to afford the title compound as a solid (13.5 mg, 65%). LC/MS: 3.69 min on 6.5 min run; *m/z* = 346.9 $(M + H)^+$. $^1$H NMR (400 MHz; $d_6$-DMSO) δ 8.07 (s, 1H), 7.96 (app t, *J* = 1.2 Hz, 1H), 7.92 (d, *J* = 7.8 Hz, 1H), 7.71-7.68 (m, 2H), 7.52 (d, *J* = 1.7 Hz, 1H), 7.39 (d, *J* = 8.2 Hz, 1H), 7.26 (dd, *J* = 8.2, 1.8 Hz, 1H), 4.07 (s, 3H), 3.64-3.50 (m, 1H), 2.05-1.92 (m, 2H), 1.81-1.68 (m, 2H), 1.65-1.55 (m, 1H), 1.40-1.25 (m, 4H), 1.25-1.10 (m, 1H); Purity = 99.3% by HPLC Conditions A

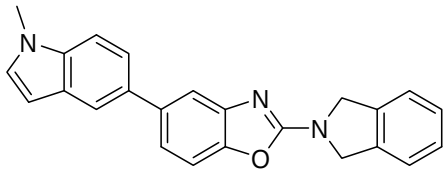
**_N_-cyclohexyl-5-(1-methyl-1_H_-indol-4-yl)benzo[_d_]oxazol-2-amine 24**



The compound was prepared using general procedure B. *m/z* = 347.4 $(M + H)^+$; Purity = 99.1% by HPLC Conditions A.

**Preparation of key compound 40 (RN-9605) in Table 2**

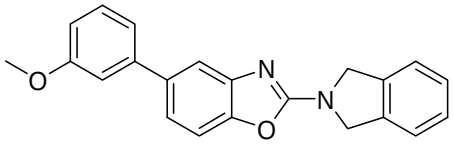
**2-(1,3-Dihydro-isoindol-2-yl)-5-(1-methyl-1*H*-indol-5-yl)-benzoxazole 40**



A microwave vial was charged with 5-(1-methyl-1*H*-indol-5-yl)-benzoxazole-2-thiol (20 mg, 0.07 mmol), isoindoline (26 mg, 0.21 mmol), and acetonitrile (0.2 mL). The mixture was heated in the microwave at 190 °C for 1 hr. The crude mixture was dissolved in DMSO, filtered and purified by preparative HPLC to yield the title product (4 mg) as a solid. LC-MS: 3.92 min; ESI *m/z* 366.3 [M+H]. $^1$H-NMR (400 MHZ; $d_6$-DMSO) δ 7.80 (d, *J* = 1.2 Hz, 1H), 7.58 (d, *J* = 1.5 Hz, 1H), 7.50 (d, *J* = 8.3 Hz, 2H), 7.47-7.43 (m, 3H), 7.38-7.35 (m, 3H), 7.31 (dd, *J* = 8.3, 1.8 Hz, 1H), 6.47 (dd, *J* = 3.0, 0.6 Hz, 1H), 4.97 (s, 4H), 3.82 (s, 3H); Purity = 94.9% by HPLC Conditions A

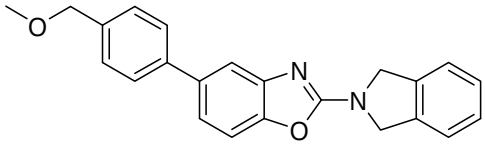
## Preparation of compounds in Table 3

### 2-(1,3-Dihydro-isoindol-2-yl)-5-(3-methoxy-phenyl)-benzoxazole 44



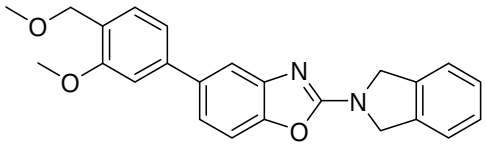
The compound was synthesized using General procedure D using 3-methoxyphenyl boronic acid (7.7 mg, 0.05 mmol) to yield the title product (7.2 mg, 82%) as a solid. $^1$H-NMR (400 MHz; $d_6$-DMSO) δ 7.61 (d, $J$ = 1.7 Hz, 1H), 7.53 (d, $J$ = 8.3 Hz, 1H), 7.48-7.42 (m, 2H), 7.40-7.34 (m, 3H), 7.33 (dd, $J$ = 1.9 Hz, $J$ = 8.3 Hz, 1H), 7.23 (d with fine str., $J$ = 6.8 Hz, 1H), 7.19 (app t, $J$ = 2.0 Hz, 1H), 6.92 (ddd, $J$ = 0.7 Hz, $J$ = 2.5 Hz, $J$ = 8.2 Hz, 1H), 4.97 (s, 4H), 3.83 (s, 3H). LC/MS: 3.90 min on 6.5 min run; ESI *m/z* 343.1 [M+H]; Purity = 99.4% by HPLC Conditions A

### 2-(1,3-Dihydro-isoindol-2-yl)-5-(4-methoxymethyl-phenyl)-benzoxazole 45



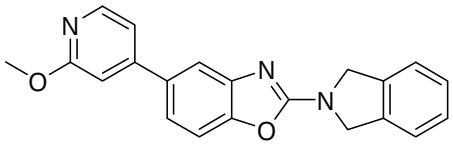
The compound was synthesized using General Procedure E to give the desired product (6.08 mg, 67%) as a solid. $^1$H NMR (400 MHz; $d_6$-DMSO) 7.66 (d with fine str, $J$ = 8.2 Hz, 2H), 7.60 (d, $J$ = 1.7 Hz, 1H), 7.53 (d, $J$ = 8.3 Hz, 1H), 7.48-7.42 (m, 2H), 7.40 (d, $J$ = 8.3 Hz, 2H), 7.38-7.34 (m, 2H), 7.32 (dd, $J$ = 1.9 Hz, $J$ = 8.3 Hz, 1H), 4.97 (s, 4H), 4.45 (s, 2H), 3.32 (s, 3H). LC/MS: 3.85 min on 6.5 min run; ESI *m/z* 357.1 [M+H]; Purity = 99.2% by HPLC Conditions A

**2-(1,3-Dihydro-isoindol-2-yl)-5-(3-methoxy-4-methoxymethyl-phenyl)-benzoxazole 46**



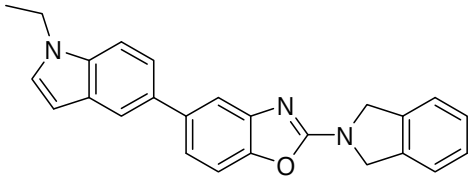
To a small microwave vial was added 4-bromo-2-methoxy-1-methoxymethylbenzene (100 mg, 0.044 mmol) and POPd (1.1 mg, 0.0022 mmol). A solution of 2-(1,3-dihydro-isoindol-2-yl)-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-benzoxazole (8.0 mg, 0.022 mmol) and tetra-*N*-butylammonium iodide (0.82 mg, 0.0022 mmol) in *N*,N-dimethylformamide (600 μL) followed by a solution of $Cs_2CO_3$ (29 mg, 0.088 mmol) in Water (150 μL). The system was sealed and purged with nitrogen then heated in the microwave for 20 min at 150 °C. LC/MS indicates complete reaction. The reaction was diluted with EtOAc (5 mL) and washed with brine (5 mL). The aqueous layer was back extracted with EtOAc (about 2 mL) and the combined organics were washed with brine (2 x 5 mL), dried ($Na_2SO_4$), filtered and evaporated. The crude product was dissolved in DMSO (450 μL) and purified by reverse phase HPLC (acetonitrile in water gradient buffered to pH10 with 10 mM $Et_2NH$ in water) to obtain a the desired product (1.14 mg, 13%) as a solid. $^1$H NMR (400 MHz; $d_4$-MeOH) 7.55 (d, $J$ = 1.7 Hz, 1H), 7.47-7.40 (m, 3H), 7.40-7.33 (m, 4H), 7.21-7.17 (m, 2H), 5.02 (s, 4H), 4.53 (s, 2H), 3.93 (s, 3H), 3.41 (s, 3 H). LC/MS: 3.88 min on 6.5 min run; ESI *m/z* 387.2 [M+H]; Purity = 98.1% by HPLC Conditions A

**2-(1,3-Dihydro-isoindol-2-yl)-5-(2-methoxy-pyridin-4-yl)-benzoxazole 47**



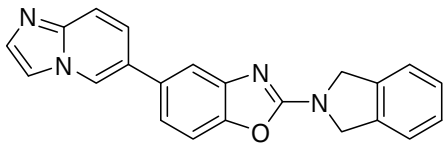
The compound was synthesized using General Procedure E to give the desired product (2.4 mg, 32%) as a solid. $^1$H NMR (400 MHz; $d_6$-DMSO) 8.21 (d, $J$ = 5.4 Hz, 1H), 7.75 (d, $J$ = 1.8 Hz, 1H), 7.58 (d, $J$ = 8.3 Hz, 1H), 7.48-7.42 (m, 3H), 7.40-7.32 (m, 3H), 7.12 (d, $J$ = 1.0 Hz, 1H), 4.98 (s, 4H), 3.90 (s, 3H). LC/MS: 3.59 min on 6.5 min run; ESI m/z 344.3 [M+H]; Purity = 99.0% by HPLC Conditions A

**2-(1,3-Dihydro-isoindol-2-yl)-5-(1-ethyl-1*H*-indol-5-yl)-benzoxazole 48**



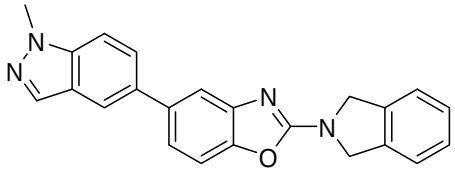
The compound was synthesized using General Procedure E to give the desired product (1.9 mg, 22%) as a solid. $^1$H NMR (400 MHz; $d_6$-DMSO) 7.80 (d, $J$ = 1.3 Hz, 1H), 7.58 (d, $J$ = 1.7 Hz, 1H), 7.55 (d, $J$ = 8.6 Hz, 1H), 7.51 (d, $J$ = 8.3 Hz, 1H), 7.48-7.40 (m, 4H), 7.40-7.34 (m, 2H), 7.31 (dd, $J$ = 1.8 Hz, $J$ = 8.3 Hz, 1H), 6.48 (dd, $J$ = 0.6 Hz, $J$ = 3.1 Hz, 1H), 4.98 (s, 4H), 4.24 (q, $J$ = 7.2 Hz, 2H), 1.38 (t, $J$ = 7.2 Hz, 3H). LC/MS: 4.08 min on 6.5 min run; ESI *m/z* 380.0 [M+H]; Purity = 96.7% by HPLC Conditions A

**2-(1,3-Dihydro-isoindol-2-yl)-5-imidazo[1,2-*a*]pyridin-6-yl-benzoxazole 49**



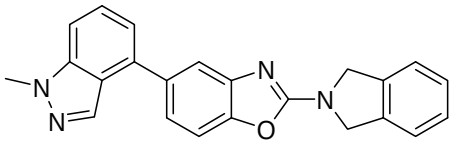
A microwave vial was charged with 5-Bromo-2-(1,3-dihydro-isoindol-2-yl)-benzoxazole (15 mg, 0.05 mmol), imidazo[1,2-a]pyridin-6-ylboronic acid (15 mg, 0.1 mmol), POPd (2.4 mg, 0.005 mmol), tetra-*N*-butylammonium iodide (2.1 mg, 0.006 mmol) and $Cs_2CO_3$ (62 mg, 0.2 mmol). *N*,*N*-Dimethylformamide (0.7 mL) and water (0.18 mL) was added and the mixture was placed under an atmosphere of nitrogen (air was evacuated x 2). The mixture was heated in the microwave at 150 °C for 20 min. The crude mixture was dissolved in DMSO, filtered and purified by preparative HPLC to yield the title product (7 mg) as a solid. This product was repurified by preparative HPLC to yield the title product (2.5 mg) as a solid. $^1$H-NMR (300 MHz; $d_6$-DMSO) δ 8.90 (s, 1H), 8.54 (s, 1H), 7.90 (s, 1H), 7.66 (d, *J* = 1.2 Hz, 1H), 7.64 (d, *J* = 8.4 Hz, 1H), 7.61-7.57 (m, 3H), 7.46-7.44 (m, 2H), 7.38-7.34 (m, 2H), 4.98 (s, 4H). LC-MS: 2.03 min; ESI *m/z* 353.1 [M+H]; Purity = 94.7% by HPLC Conditions A

**2-(1,3-Dihydro-isoindol-2-yl)-5-(1-methyl-1*H*-indazol-5-yl)-benzoxazole 50**



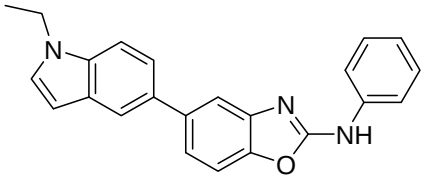
Prepared by Method E. After completion of the reaction, the mixture was dissolved in DMSO and purified by HPLC to give the title compound (0.4 mg, 2%) as a solid. LC-MS: 3.61 min on a 6.5 min runtime; ESI *m/z* 367.3 [M+H]; $^1$H-NMR (400 MHZ; $d_6$-DMSO) δ 8.07 (s, 1H), 7.72-7.71 (m, 2H), 7.94 (br. s, 1H), 7.63 (d, *J* = 1.8 Hz, 1H), 7.54 (d, *J* = 8.3 Hz, 1H), 7.47-7.43 (m, 2H), 7.38-7.34 (m, 3H), 4.98 (s, 4H), 4.07 (s, 3H); Purity = 98.1% by HPLC Conditions A

**2-(1,3-Dihydro-isoindol-2-yl)-5-(1-methyl-1*H*-indazol-4-yl)-benzoxazole 51**



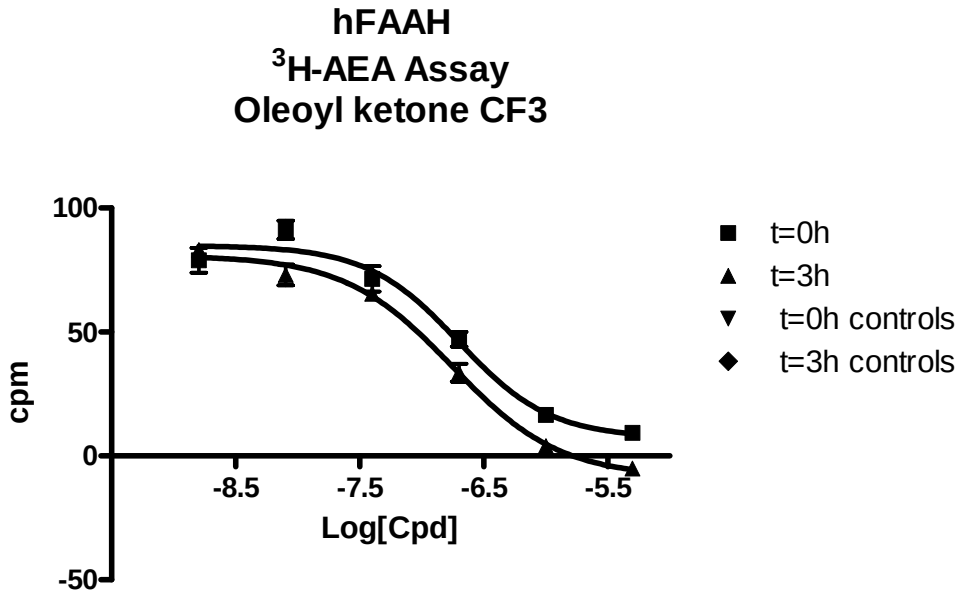
The compound was synthesized using General Procedure E to give the desired product (2.54 mg, 31%) as a solid. $^1$H NMR (400 MHz; $d_6$-DMSO) 8.12 (d, $J$ = 0.9 Hz, 1H), 7.66-7.59 (m, 3H), 7.52-7.43 (m, 3H), 7.41-7.34 (m, 3H), 7.27 (d, $J$ = 6.5 Hz, 1H), 4.99 (s, 4H), 4.10 (s, 3H). LC/MS: 3.67 min on 6.5 min run; ESI *m/z* 367.3 [M+H]; Purity = 99.1% by HPLC Conditions A.

**5-(1-Ethyl-1*H*-indol-5-yl)-*N*-phenylbenzo[*d*]oxazol-2-amine 52 (RN-7205)**



The compound was synthesized using Method B. After completion of the reaction and cooling, the product was precipitated from the mixture by slow addition of $H_2O$ (2 mL). The emerging solid was filtered and the filter cake purified by preparative HPLC [20-98% acetonitrile:water buffered to pH 10 with 10 mM diethylamine in water] to afford the title compound. H NMR (400 MHz; $d_6$-DMSO) 10.64 (br. s, 1H), 7.82 (d, $J$ = 1.5 Hz, 1H), 7.79 (d, $J$ = 7.7 Hz, 2H), 7.69 (d, $J$ = 1.5 Hz, 1H), 7.54 (app. t, $J$ = 8.6 Hz, 2H), 7.46 (dd, $J$ = 8.6, 1.7 Hz, 1H), 7.44-7.36 (m, 4H), 7.04 (t, $J$ = 7.4 Hz, 1H), 6.49 (d, $J$ = 2.8 Hz, 1H), 4.24 (q, $J$ = 7.2 Hz, 2H), 1.38 (t, $J$ = 7.2 Hz, 3H); ESI *m/z* 354.5 [M+H]; Purity = 99.8% by HPLC Conditions A.

## IC50 Shift Experiments with Oleyl trifluoromethyl ketone 6

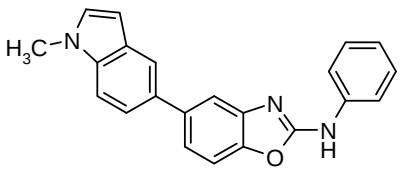



t=0: $IC_{50}$=198.2nM
t=3h: $IC_{50}$=176.2nM

Average $IC_{50}$ = 187 nM (literature $IC_{50}$ using a similar $^{3}H$ anandamide assay is 74nM)[1]

1. S. J. Wilson, T. W. Lovenberg and A. J. Barbier, *Anal. Biochem.* 2003, **318**, 270-275.

## Hepatocyte Study with Compound 25




Compound **25**

Rat hepatocyte stability with compound **25** was measured and shown to be >500 min (results from Renovis DMPK group).