Fully functionalized small-molecule probes for integrated phenotypic screening and target identification

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Supporting Information

A. Supplemental Tables and Figures

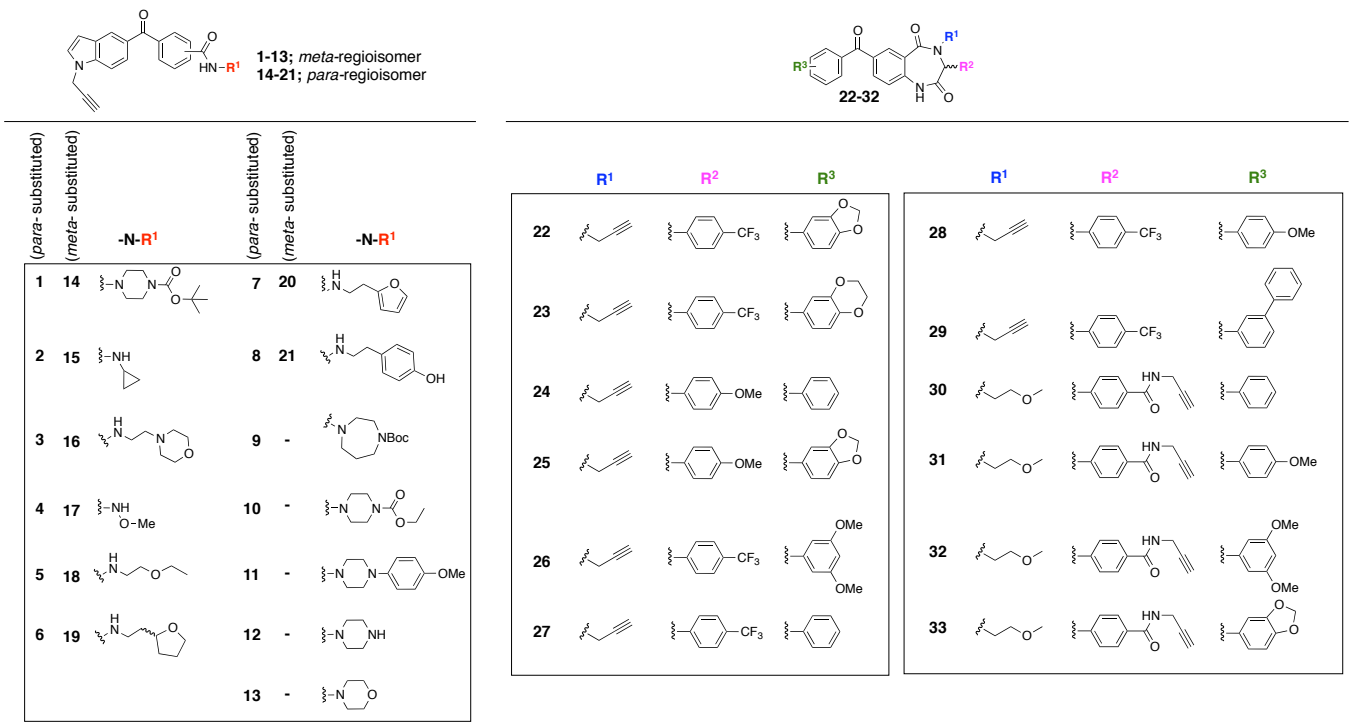


Figure S1. Structures of fully functionalized library members.

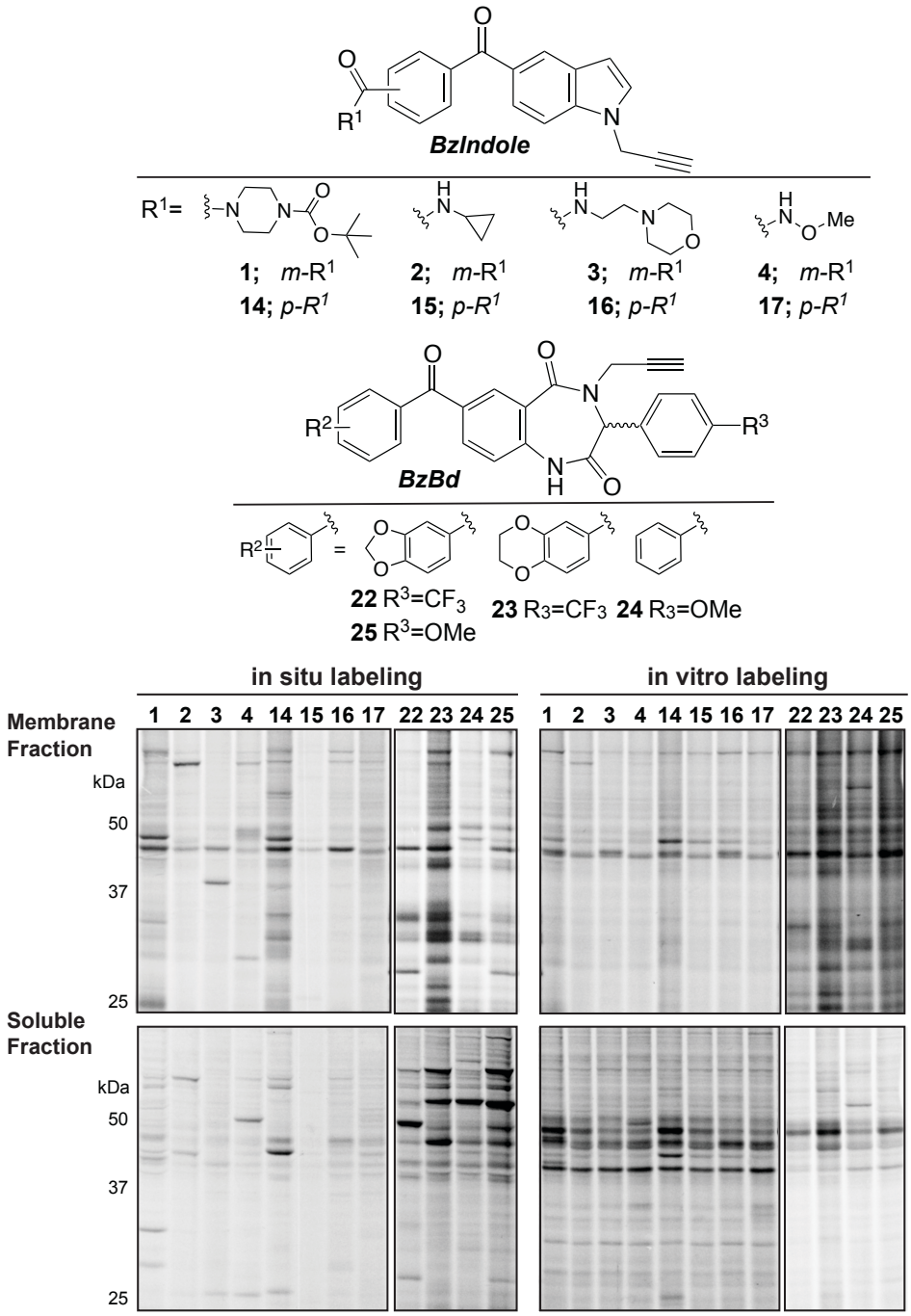
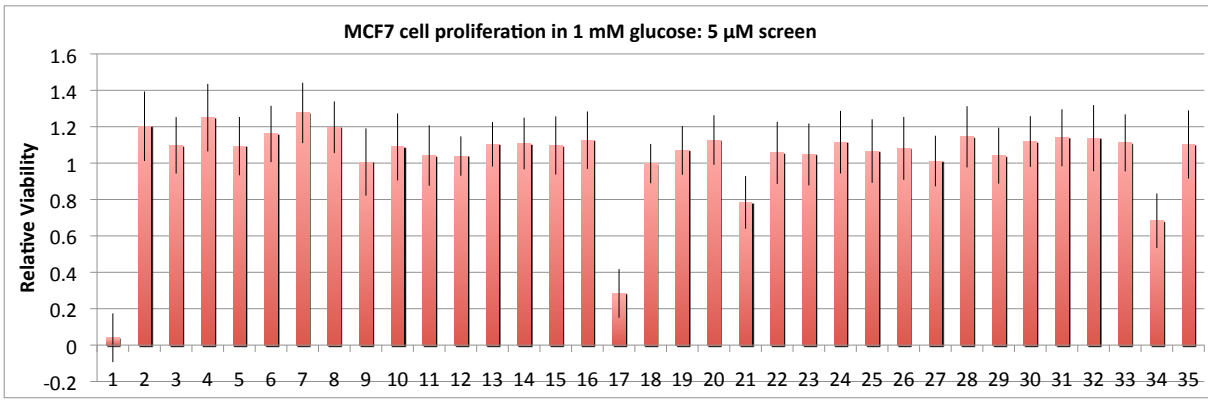
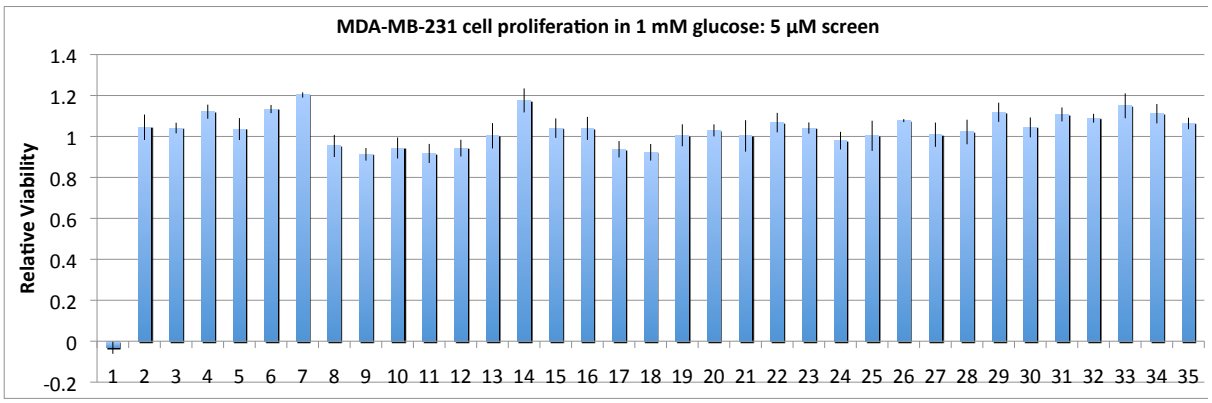


Figure S2. Comparison of *in situ* and *in vitro* protein labeling profiles for representative members of the fully functionalized compound library. Library members (10 μM each) were added to MDA-MB-231 proteomes (1 mg/mL protein) for 5 min or cells for 40 min before photocrosslinking, click chemistry conjugation to an azide-rhodamine tag, SDS-PAGE, and in-gel fluorescence scanning (fluorescent gels shown in gray scale). Note that the *in situ* gel panels are reproduced from **Figure 1B** of the manuscript to facilitate direct comparison to the *in vitro* profiles.





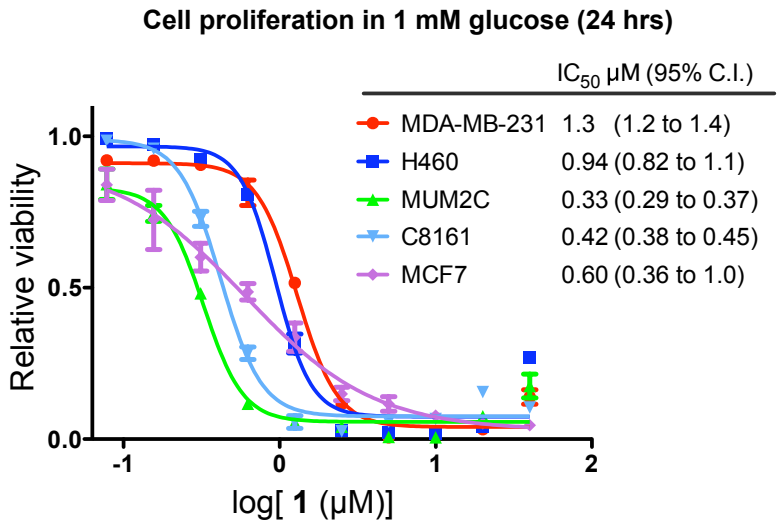


Figure S3. Screen of compound library for inhibitors of cancer cell proliferation under low glucose conditions (MDA-MB-231 cells or MCF7 cells; 1 mM glucose, RPMI media) and dose response for **1** in multiple human cancer cell lines under low glucose conditions. Cell viability was determined after 24 hr using the WST-1 assay (Clonetech) following the manufacture's instructions.

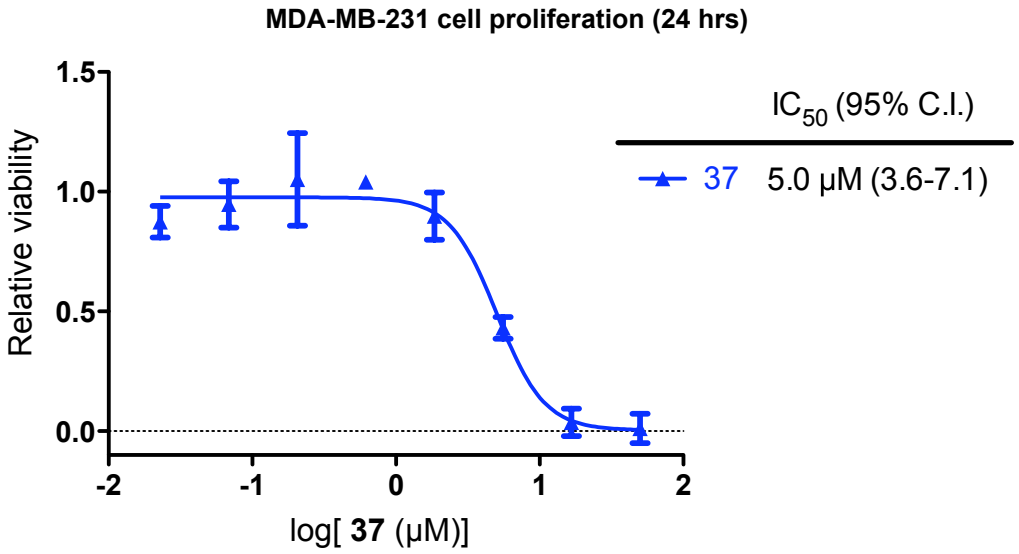


Figure S4. Concentration dependent inhibition of MDA-MB-231 cell proliferation by the nonclickable analog **37** under low glucose conditions. Cell viability was determined after 24 hrs using the WST-1 assay (Clonetech) following the manufacture's instructions.

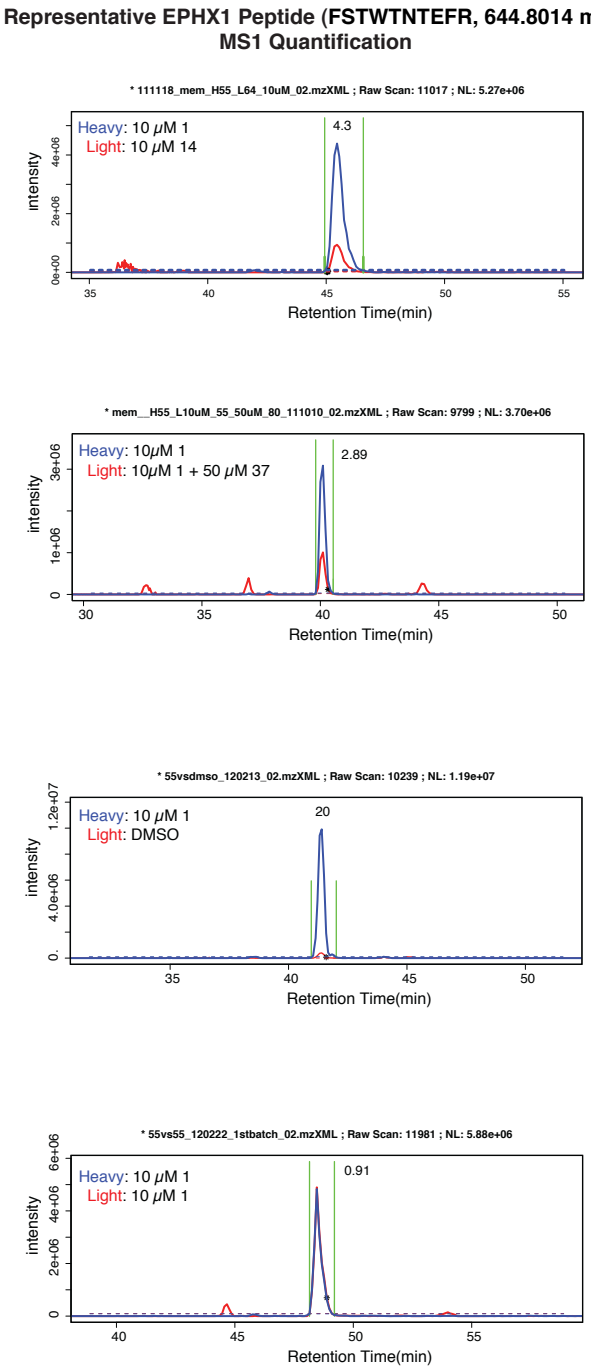


Figure S5. Extracted MS1 chromatographs for representative peptides from EPHX1. Ratio values (heavy/light) are given above the peak. The conditions for each proteomic experiment are shown as insets in the upper left corner of each trace.

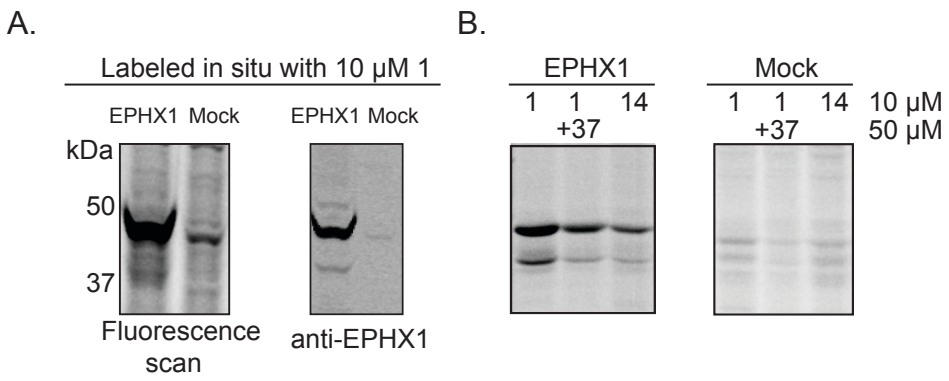


Figure S6. Confirmation that EPHX1 is a specific target of probe **1**. (A) HEK293T cells were transfected with a human EPHX1 cDNA or mock-transfected with empty vector and then labeled *in situ* with 10 μM **1**, harvested, and conjugated to a rhodamine-azide reporter tag under click chemistry conditions. Probe–protein conjugates were visualized by SDS-PAGE and in-gel fluorescence scanning (left panel). EPHX1 overexpression was confirmed by western blotting with anti-EPHX1 antibodies (right panel). (B) Comparison of *in situ* labeling of EPHX1-overexpressing cells and mock-transfected cells with: 1) probe **1**, 2) probe **1** + 5X **37**, and 3) control probe **14**. Recombinantly expressed EPHX1 protein migrated as two protein bands between 40-50 kDa that were detected by both probe **1** labeling and western blotting. Note that EPHX1 was labeled more strongly by probe **1** than probe **14** and this labeling was competed by 5X **37**.

				1 vrs **14** experiment					**1** vrs **1** + 5X **37** experiment					**1** vrs **DMSO** experiment		**1** vrs **1** experiment
MW (kDa)	IPI	description	symbol	Rep 1	Rep 2	Rep 3	**AVE**	**sem**	Rep 1	Rep 2	Rep 3	**AVE**	**sem**	Rep 1	Rep 2	Rep 1
53	P07099	EPHX1 Epoxide hydrolase 1	EPHX1	4.33	4.54	6.7	**5.19**	**0.76**	1.66	2.79	2.37	**2.27**	**0.33**	20	20	0.89
35.7	P03886	MT-ND1 NADH-ubiquinone oxidoreductase chain 1	MT-ND1	2.25	3.21	3.55	**3.00**	**0.39**	1.82	3.2	2.13	**2.38**	**0.42**	20	20	0.74
46.5/32.2	Q9HDC9	APMAP Adipocyte plasma membrane-associated protein	APMAP	1.69	1.91	1.85	**1.82**	**0.07**	1.49	2.5	2.73	**2.24**	**0.38**	16.5	20	0.82
47.1	Q8NBX0	SCCPDH Probable saccharopine dehydrogenase	SCCPDH	1.37	1.6	1.94	**1.64**	**0.17**	4.76	6.17	4.96	**5.30**	**0.44**	20	20	0.86
61.6	Q96S52	PIGS GPI transamidase component PIG-S	PIGS	1.2	*0*	1.55	**1.38**	0.47	*0*	2.42	2.12	2.27	0.76	20	0	0.94
42.3/37.1	Q9H4I3	TRABD TraB domain-containing protein	TRABD	1.1	1.1	1.71	**1.30**	**0.20**	1.38	3.24	2.39	**2.34**	**0.54**	20	20	0.83
21.1	Q9NRV9	HEBP1 Heme-binding protein 1	HEBP1	0.85	*0*	1.03	**0.94**	**0.32**	*0*	2.12	2.47	**2.30**	**0.77**	20	20	0.81

Table S1. Chemoproteomic assignment of selective targets of **1**. Targets that showed > 2-fold higher signal in proteomic experiments with **1** versus **14** and **1** versus **1** + 5X **37** are highlighted. See **Table S2** for a complete list of protein targets identified in proteomic experiments. To be considered specific protein targets of compound **1**, proteins also needed to possess 1) ratios that were > 10 in **1**-treated versus DMSO-treated experiments, and 2) > 2 quantifiable peptides per replicate. The values in each replicate (Rep) experiment correspond to median values for all of the peptide ratios identified for each protein in that experiment. Note that, for the **1** versus DMSO data set, a value of 20 was given as an upper limit ratio for peptides that appeared as singletons (i.e., peptides that were only found in proteomes from compound **1**-treated but not DMSO-treated cells)

B. Chemoproteomic and Assay Experimental Protocols

Materials.

Chemical reagents were obtained from Sigma-Aldrich or ThermoFisher unless otherwise noted. Cell culture media and supplements wree obtained through CellGro or Omega Scientific. Crosslinking was performed with a Spectrolinker XL-1000 (Spectroline).

Cell culture.

Cell lines were obtained from ATCC and maintained in DMEM (H460), or RPMI (C8161, MUM2C) media supplemented with 10% fetal bovine serum and 2 mM L-glutamine in 5% CO_2 incubator at 37 ºC. MDA-MB-231 cells were also routinely maintained in L15 in an incubator at 37 ºC. For comparison of normal and low glucose conditions, glucose was supplemented to glucose-free media. For probe treatments, all solutions were made using 10 mM or 50 mM stock DMSO solutions.

Photocrosslinking and gel-based analysis.

For *in situ* labeling, MDA-MB-231 cells were plated in 10 cm dishes and grown to 90% confluency, washed with PBS, and treated with 10 µM compound (in 5 mL serum free media). After 40 min at 37 ºC, cells were placed at 4 ºC for 5 min and subsequently irradiated with 365 nm light for 5 min (ca. 12 J/cm^2) at 4 ºC. Cells were then harvested by washing 2x with cold PBS (pH 7.5) and then scraped into cold PBS. Cell pellets were isolated by centrifugation (1400 x g, 3 min), resuspended in PBS, sonicated, centrifuged (100,000 x g, 45 min) to provide the soluble (supernatant) and membrane (pellet) fractions. Protein concentrations were determined using the BCA protein assay (Bio-Rad) using a microplate reader. For conjugation to rhodamine-azide, 50 µg of lysate was treated 6 µL of a master mix solution: 3µL of 1.7 mM Tris[(1-benzyl-1H-1,2,3-triazol-4-yl)methyl]amine in 4:1 DMSO:*t*-BuOH, 1 µL of 50 mM $CuSO_4$ in H_2O, 1 µL 50 mM TCEP in PBS, 1µL of 1.25 mM rhodamine-azide in DMSO. After 1.5 hours at room temperature, samples were mixed with 2X SDS loading buffer (without boiling) and 25 µg was separated using SDS-PAGE. Imaging was achieved using in-gel fluorescent scanning on a Hitachi FMBIO-II flatbed fluorescence scanner.

For *in vitro* labeling, MDA-MB-231 cells were harvested, lysed, and centrifuged to produce soluble and membrane fractions as described above. Protein concentrations were obtained and each fraction was adjusted to 1mg/ml. In a 96-well plate, 50 µL of the membrane or soluble proteome was treated with probe (10 µM), mixed, and allowed to stand for 5 min at rt. The samples were cooled to 4 ºC and crosslinked for 5 min at 4 ºC. Conjugation to rhodamine-azide, SDS-PAGE, and imaging was carried out as described above with the exception that 12.5 µg was loaded onto the gel.

WST-1 cell viability assay.

10,000 cells were added to each well of a 96-well plate in 100 µL media (Glucose-free media supplemented with 8mM glucose or 1mM glucose). After 15 hrs, compound in 100 µL media was added. Cells were grown for an additional 24hrs, whereupon each well was aspirated. 100 µL of media (with 8mM glucose) containing 10% premixed WST-1 reagent (Clonetech) was added and incubated at 37 ºC for 45 min. Absorbance at 440 nm was read using a microplate reader. Relative cell viability was determined by subtracting background signal (2 µM staurosporine treated cells) and normalizing to background subtracted control cells treated with DMSO. Reported relative cell viability values are the average of three replicates. Error bars indicate the standard error of the mean. IC_{50} values were determined from dose response curves from three replicates at each inhibitor concentration fitted using Prism software (Graphpad).

Photocrosslinking and protein profiling using mass spectrometry.

We performed quantitative proteomics experiments using SILAC (stable isotope labeling by amino acids in cell culture) methods[1] as follows. MDA-MB-231 cells were passaged six times in DMEM minus L-Lysine and L-Arginine (Thermo) supplemented with 10% dialyzed FBS (Gemini) and 50 mg / L [$^{13}C_6$,$^{15}N_4$] L- Arginine-HCl and 50 mg / L [$^{13}C_6$,$^{15}N_2$] L-Lysine-HCl (Aldrich) or L-Arginine-HCl and L-Lysine-HCl (Sigma) and cell aliquots were frozen and replaced periodically. Heavy and light MDA-MB-231 cells were plated in 15 cm dishes and grown to 100% confluency, washed with PBS, and treated with 10 µM compound (in 10 mL serum free media). For competition experiments (**1** vrs **1**+5x**37** experiment), heavy cells were treated with 10 µM **1** and light cells were treated with 10 µM **1** and 50 µM 1ane (**37**). For **1** and **14** comparison (**1** vrs **14** experiment), heavy cells were treated with 10 µM **1** and light cells were treated with 10 µM **14**. For control experiments, heavy and light cells were treated with 10 µM **1** (**1** vrs **1** experiment) or heavy cells were treated with 10 µM **1** and light cells were treated with DMSO (**1** vrs DMSO experiment). Cells were incubated with compound(s) at 37 ºC for 40 min and harvested as described above to yield heavy and light lysate (200 µL per sample). 1mg of heavy and 1mg light lysate were combined and centrifuged (100,000 x g, 45 min) to provide the soluble (supernatant) and membrane (pellet) fractions. Each fraction was adjusted to 1mg/mL and click chemistry reagents were added to achieve the following final concentrations: 20 µM $CuSO_4$, 2 µM TBTA, 20 µM TCEP, and 100 µM biotin-azide. After mixing for 1.5 hr, the reaction was diluted to 2 mL with PBS. 2mL MeOH and 0.5 mL $CHCl_3$ were added, shaken, and centrifuged at 4000x g for 10min at room temperature yielding a protein solid between the aqueous and organic layers. The top and bottom liquid phases were aspirated, leaving the protein solid. The protein solid was washed 3 x 2 mL 1:1 MeOH:$CHCl_3$.. 2 mL MeOH was added and the solution was sonicated to yield a cloudy mixture. 0.5 mL $CHCl_3$ was added and the solution was centrifuged (13,000 x g,

5 min), aspirated, and the pellet was solubilized in 6 M urea in PBS (450 µL final volume). Membrane fractions were supplemented with 1% SDS. The solution was treated with 10 mM TCEP (neutralized to pH 7) for 30 minutes at 37 °C and followed with treatment with 20 mM iodoacetamide for 30 minutes at room temperature. The sample was diluted 10x in PBS and SDS was added to a final concentration of 0.2 %. Streptavidin beads (Thermo) (100 µL slurry) were added and rotated for 2 hours at rt. Beads were washed with 3 x 10mL PBS/0.5% SDS and 3 x 10mL PBS and transferred to screw-top eppendorf tubes and resuspended in 2 M urea/PBS supplemented with 1 mM calcium chloride and sequence grade porcine trypsin (Promega) for overnight digestion at 37°C. The eluant was collected the following day and acidified with 5% formic acid and stored at - 20 ºC.

Mass Spectrometry.

Mass spectrometry was performed using a Thermo Orbitrap Velos mass spectrometer. Peptides were eluted using a 5-step MudPIT protocol (using 0%, 25%, 50%, 80%, and 100% salt bumps of 500 mM aqueous ammonium acetate, each step followed by an increasing gradient of aqueous acetonitrile/0.1% formic acid) and data were collected in data-dependent acquisition mode with dynamic exclusion turned on (20 s, repeat of 1). Specifically, one full MS (MS1) scan (400-1800 m/z) was followed by 30 MS2 scans of the most abundant ions. The MS2 spectra data were extracted from the raw file using RAW Xtractor (version 1.9.1; publicly available at http://fields.scripps.edu). ProLuCID searches allowed for variable oxidation of methionine (+15.9949), static modification of cysteine residues (+57.0215 due to alkylation), and no enzyme specificity. Each data set was independently searched with light and heavy params files; for the light search, all other amino acids were left at default masses; for the heavy search, static modifications on lysine (8.0142) and arginine (10.0082) were specified. The precursor ion mass tolerance was set to 50 ppm and the fragment ion mass tolerance was left at the default assignment of 0. The data was searched using a human reverse-concatenated non-redundant (gene-centric) FASTA database. The resulting MS2 spectra matches were assembled into protein identifications and filtered using DTASelect (version 2.0.47) with the --modstat --mass and --trypstat options, which applies different statistical models for the analysis of high resolution masses, peptide digestion state, and methionine oxidation states, respectively. Redundant peptide identifications common between multiple proteins were allowed, but the database was restricted to a single consensus splice variant. SILAC ratios were quantified using CIMAGE software.[2] The program was updated to identify cases where complete inhibition could not be quantified based on light/heavy peak pairs due the absence of a MS1 signal from either the heavy or light sample. In order to identify these cases, all single MS1 chromatographic peaks (from either the light or the heavy sample) were identified within a retention time window. Next, these peaks were aligned with the corresponding sequence ProLuCID /DTASelect identification and the charge state and monoisotopic mass were validated using the “envelope correlation score” filter[2]. Finally, the candidate peak was cross-checked to ensure there was no corresponding (heavy or light) peak co-eluting around the same retention time window.

Only after all these conditions are met, the peptide was assigned as the case of complete inhibition with an artificial threshold ratio of 20. The described **1** versus **14** and **1** versus **1**+5x**37** experiments were performed in triplicate. The "**1** versus DMSO" experiment was performed in duplicate, and one replicate of the "**1** versus **1**" experiment was performed. Reported ratios are the mean of all the unique, quantified peptides per protein. Proteins were included in the analysis only those targets showed 1) > 2-fold reduction in signal in proteomic experiments with 5X competitor (compound **37**); and 2) > 2-fold higher signal in proteomic experiments with **1** compared to **14**. See **Table S2** for a complete list of protein targets identified in proteomic experiments. Only high-confidence protein signals were considered for this analysis, which required that they possess 1) ratios that were > 10 in 1-treated versus DMSO-treated experiments, and 2) > 2 quantifiable peptides per replicate.

EPHX1 overexpression and labeling

HEK293T cells were grown to 70% confluency in 10 cm dishes in DMEM (supplemented with 10% FCS). Cells were transfected using Fugene HD and 5 µg vector containing human EPHX1 (OpenBiosciences Cat. # MHS1010-98051239) or empty vector control ("mock") using the manufactures protocols. After 24 hrs, the cells were trypsanized and replated in 6-well dishes. *in situ* labeling, cell harvesting, and conjugation to rhodamine-azide were peformed as described above. SDS-PAGE was performed on 4-20% tris-glycine gels and followed by in-gel fluorescence scanning. Western blotting was performed using anti-EPHX1 (1:1000 genetex Cat. # GTX109360).

Complex 1 NADH oxidase activity assay.

Bovine heart submitochondrial particles were prepared by the method of Yagi and Hatefi[3] using sonication medium containing 0.25 mM sucrose, 1mM succinate, 1.5 mM ATP, 10 mM $MgCl_2$, 10 mM $MnCl_2$, and 10 mM Tris-HCl (pH 7.4) and stored in buffer containing 25 mM sucrose and 10 mM Tris-HCl (pH 7.4) at -80 ºC. The NADH oxidase activity was measured using a Tecan platereader. To a 96-well plate, 100 µL reaction medium (0.25 mM sucrose, 10 mM Tris-HCl (pH 7.4), 200 µM NADH) was added. Submitochondrial particles were added to make a final concentration of 15 µg/mL. After 5 min, compound was added and the reaction was monitored for absorbance at 340 nm every 30 s for 15 min. For each compound dose, the initial rate of NADH oxidation was measured relative to a rotenone (1µM) treated sample. IC_{50} values were determined from dose response curves from three replicates at each inhibitor concentration fitted using Prism software (Graphpad).

C. Synthetic Materials and Methods

Reagents were obtained from Combi-blocks (www.combi-blocks.com), (Aldrich Chemical (www.sigma-aldrich.com), or Acros Organics (www.fishersci.com) and used without further purification. Optima grade solvents were obtained from Fisher Scientific (www.fishersci.com), degassed with Ar, and purified on a solvent drying system unless otherwise indicated. TLC was performed on 0.25 mm E. Merck silica gel 60 F254 plates and visualized under UV light (254 nm) or by staining with potassium permanganate ($KMnO_4$), cerium ammonium molybdenate (CAM), phosphomolybdic acid (PMA). Silica flash chromatography was performed on E. Merck 230–400 mesh silica gel 60. NMR spectra were recorded on Bruker DRX-600 equipped with a cryoprobe, DRX-500, or Bruker AMX-400 instruments. Chemical shifts are expressed in ppm relative to TMS (^1H, 0 ppm) or solvent signals: $CDCl_3$ (^{13}C, 77.16 ppm) (MeOD-d_4 (^{13}C, 49.00) coupling constants are expressed in Hz. Mass spectra were obtained on an Agilent ESI-TOF (high resolution) or Agilent MSD 1100 (low resolution) machines.

SAFETY NOTE: SEVERAL REACTIONS DESCRIBED BELOW INVOLVE THE USE OF CARBON MONOXIDE, A TOXIC GAS THAT IS COLORLESS AND ODERLESS. HANDLING OF THIS GAS SHOULD BE PERFORMED IN A PROPERLY FUNCTIONING CHEMICAL FUMEHOOD AND BE DONE WITH CARE.

Synthesis of 5-benzoyl indoles (BzIndoles)

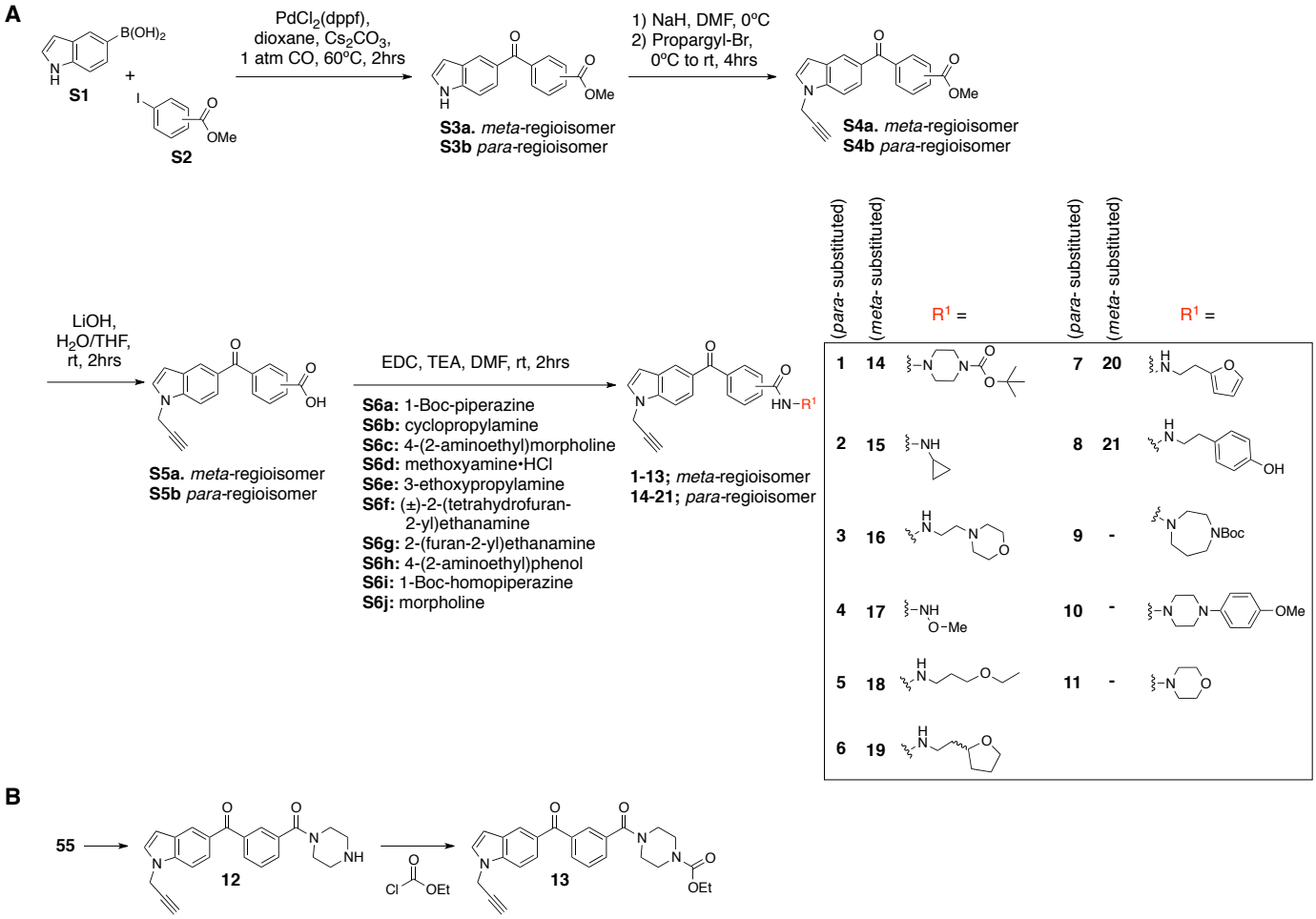
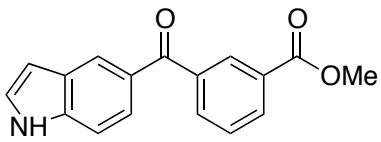
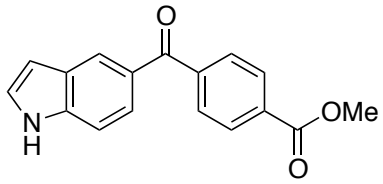


Figure S6. Synthesis of 7-benzoyl-indoles (BzIndoles) 1-21. A. Preparation of BzIndoles **1-11** and **14-21**. B. Preparation of BzIndoles **12** and **13**. (EDC = 1-Ethyl-3-(3-dimethylamino-propyl)carbodiimide, TEA = triethylamine, dppf = 1,1'-Bis(diphenylphosphino)ferrocene, DMF = *N*,*N*-dimethylformamide).

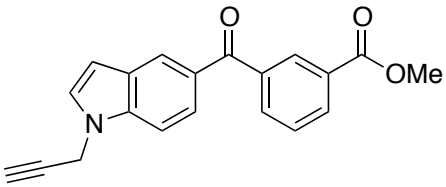


methyl 3-(1*H*-indole-5-carbonyl)benzoate (S3a). A solution of methyl 3-iodobenzoate (620 mg, 2.3 mmol, 1.0 equiv), 5-indolylboronic acid (460 mg, 2.5 mmol, 1.1 equiv), [1,1'-Bis(diphenylphosphino)ferrocene]dichloropalladium(II) (84 mg, 0.12 mmol, 0.05 equiv), Cs_2CO_3 (2.2 g, 6.9 mmol,3.0 equiv) in 1,4-dioxane (10 mL, degassed with CO) was flushed with CO for 5 min and heated to

60 °C under 1 atm CO. After 8 hrs, the solution was poured into EtOAc and the organic layer was washed with 1 N NaOH and sat aq NaCl, dried with Na_2SO_4 and concentrated under reduced pressure. Purification by silica gel chromatography (5:1→4:1 hexanes/EtOAc) yielded **S3a** (372 mg, 1.33 mmol, 58%). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ 9.32 (s, 1H), 8.54 (s, 1H), 8.30 (d, *J* = 7.8 Hz, 1H), 8.17 (s, 1H), 8.05 (d, *J* = 7.6 Hz, 1H), 7.79 (d, *J* = 8.6 Hz, 1H), 7.60 (t, *J* = 7.7 Hz, 1H), 7.44 (d, *J* = 8.6 Hz, 1H), 7.28 (t, *J* = 2.5 Hz, 1H), 6.65 (s, 1H), 4.01-3.98 (m, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ 196.94, 166.64, 139.33, 138.65, 134.09, 132.53, 130.78, 130.06, 128.77, 128.46, 127.24, 126.33, 125.34, 123.85, 111.45, 103.99, 52.40. **HRMS** *m/z*: cacld for $C_{17}H_{13}NO_3$ [M + H]$^+$ 280.0968, found: 280.0979.

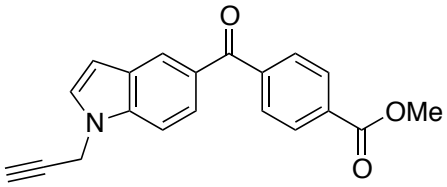


methyl 4-(1*H*-indole-5-carbonyl)benzoate (S3b). Prepared as described for **S3a** using methyl 4-iodobenzoate (1 g, 3.8 mmol, 1.0 equiv), 5-indolylboronic acid (672 mg, 4.2 mmol, 1.1 equiv) [1,1'-Bis(diphenylphosphino)ferrocene]dichloro-palladium(II) (83 mg, 0.11 mmol, 0.03 equiv), Cs_2CO_3 (3.7 g, 11.3 mmol, 3.0 equiv) in 1,4-dioxane (20 mL, degassed with CO) yielding **S3b** (603 mg, 2.16 mmol, 57%). **^{1}H-NMR** (500 MHz, $CDCl_3$): δ 8.78 (br s, 1H), 8.15 (d, *J* = 8.1 Hz, 2H), 8.11 (d, *J* = 0.8 Hz, 1H), 7.84 (d, *J* = 8.2 Hz, 2H), 7.78 (dd, *J* = 8.6, 1.6 Hz, 1H), 7.46 (d, *J* = 8.6 Hz, 1H), 7.29 (s, 1H), 6.64 (s, 1H), 3.97 (s, 3H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.73, 166.69, 143.08, 138.62, 132.63, 129.74, 129.49, 129.29, 127.36, 126.07, 125.60, 124.23, 111.34, 104.51, 52.57.**HRMS** *m/z*: cacld for $C_{17}H_{13}NO_3$ [M + H]$^+$ 280.0968, found: 280.0979.

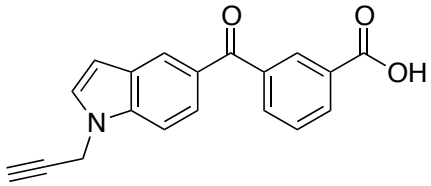


methyl 3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzoate (S4a). A solution of **S3a** (372 mg, 1.3 mmol, 1.0 equiv) in DMF (5 mL) was treated with Cs_2CO_3 (845 mg, 2.6 mmol, 2.0 equiv). After 5 min at rt, propargyl bromide (170 µL 80% wt in PhMe, 1.5 mmol, 1.2 equiv) was added. After an additional 2 hrs at rt, the solution was poured into EtOAc and the organic layer was washed with 1 N HCl and sat aq NaCl, dried with Na_2SO_4 and concentrated under reduced pressure. Purification by silica gel chromatography (3:1 hexanes/EtOAc)

yielded **S4a** (330 mg, 1.04 mmol, 80%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.43 (d, *J* = 1.5 Hz, 1H), 8.25-8.23 (m, 1H), 8.09 (d, *J* = 1.0 Hz, 1H), 7.99 (dt, *J* = 7.6, 1.3 Hz, 1H), 7.82 (dd, *J* = 8.6, 1.5 Hz, 1H), 7.58 (t, *J* = 7.7 Hz, 1H), 7.49 (d, *J* = 8.6 Hz, 1H), 7.30 (d, *J* = 3.2 Hz, 2H), 6.64 (d, *J* = 2.9 Hz, 1H), 4.93 (d, *J* = 2.5 Hz, 2H), 3.92 (s, 3H), 2.45 (t, *J* = 2.5 Hz, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 197.02, 167.36, 140.18, 139.06, 134.87, 133.38, 131.66, 131.03, 130.19, 129.86, 129.29, 129.10, 126.38, 124.98, 110.31, 104.91, 77.88, 75.04, 53.21, 36.99. **ESI-MS** *m/z* (pos): cacld for $C_{20}H_{15}NO_3$ $[M + H]^+$ 318.1, found: 318.1.

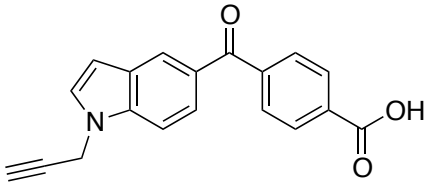


methyl 4-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzoate (S4b). Prepared as described for **S4a** using **S3b** (134 mg, 0.48 mmol, 1.0 equiv) in DMF (5 mL) was treated with Cs_2CO_3 (325 mg, 2.6 mmol, 2.0 equiv), and propargyl bromide (100 µL 80% wt in PhMe, 0.85 mmol, 1.8 equiv) yielding **S4b** (130 mg, 0.41 mmol, 85%). **^{1}H-NMR** (600 MHz, CDCl3): δ 8.15 (d, *J* = 8.3 Hz, 2H), 8.09 (s, 1H), 7.83 (d, *J* = 7.6 Hz, 3H), 7.48 (d, *J* = 8.6 Hz, 1H), 7.31 (s, 1H), 6.63 (d, *J* = 0.6 Hz, 1H), 4.93 (s, 2H), 3.97 (s, 3H), 2.46 (s, 1H). **^{13}C-NMR** (126 MHz, $CDCl_3$): δ 196.74, 166.91, 143.30, 138.66, 132.95, 130.01, 129.78, 129.65, 129.47, 128.63, 126.05, 124.47, 109.86, 104.44, 77.39, 74.61, 52.82, 36.54. **ESI-MS** *m/z* (pos): cacld for $C_{20}H_{15}NO_3$ $[M + H]^+$ 318.1, found: 318.1.**ESI-MS** *m/z* (pos): cacld for $C_{20}H_{15}NO_3$ $[M + H]^+$ 318.1, found: 318.1.



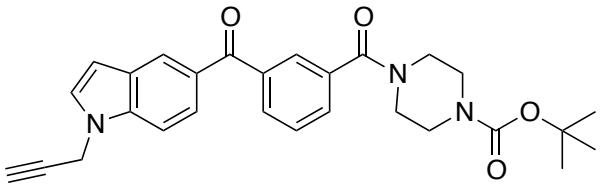
3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzoic acid (S5a). A solution of **S4a** (330 mg, 1.04 mmol, 1.0 equiv) in 1:1:2 (H_2O:MeOH:THF, 5 mL) was treated with LiOH (63 mg, 1.5 mmol, 1.5equiv). After 4hrs, the solution was poured into EtOAc and the organic layer was washed with 1 N HCl, dried with Na_2SO_4 and concentrated under reduced pressure yielding **S5a** (214 mg, 0.70 mmol, 70%). **^{1}H-NMR** (600 MHz, MeOD-d_4): δ 8.37 (s, 1H), 8.25 (d, *J* = 7.8 Hz, 1H), 8.04 (d, *J* = 1.3 Hz, 1H), 7.95 (d, *J* = 7.6 Hz, 1H), 7.72 (dd, *J* = 8.6, 1.4 Hz, 1H), 7.63 (t, *J* = 7.7 Hz, 1H), 7.59 (d, *J* = 8.7 Hz, 1H), 7.41 (d, *J* = 3.2 Hz, 1H), 6.62 (d, *J* = 3.2 Hz, 1H), 5.05 (d, *J* = 2.5 Hz, 2H), 2.86 (t, *J* = 2.4 Hz, 1H). **^{13}C-NMR** (151 MHz, MeOD-d_4): δ 198.29, 168.91, 140.53, 139.89, 134.92, 133.83, 132.23, 131.81, 130.89, 130.05, 129.77, 129.64, 126.33, 124.69, 110.90, 104.68,

78.81, 74.92, 36.53. **ESI-MS** *m/z* (pos): cacld for $C_{20}H_{15}NO_3$ [M + H]$^+$ 318.1, found: 318.1. **ESI-MS** *m/z* (pos): cacld for $C_{19}H_{13}NO_3$ [M + H]$^+$ 304.1, found: (neg) 304.1. [M - H]$^-$ 302.1, found: 302.1.



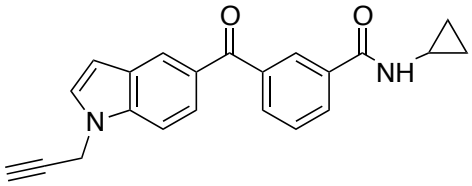
4-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzoic acid (S5b). Prepared as described for **S5a** using **S4b** (130 mg, 0.41 mmol) and LiOH (43 mg, 1.0 mmol) and yielded **S5b** (90 mg, 0.30 mmol, 72%) **^{1}H-NMR** (600 MHz, MeOD-d_4): δ 8.17 (d, *J* = 8.3 Hz, 2H), 8.07 (s, 1H), 7.82 (d, *J* = 8.4 Hz, 2H), 7.76 (dd, *J* = 8.6, 1.1 Hz, 1H), 7.62 (d, *J* = 8.7 Hz, 1H), 7.43 (d, *J* = 3.0 Hz, 1H), 6.64 (s, 1H), 5.07 (s, 2H), 2.87 (s, 1H). **^{13}C-NMR** (151 MHz, MeOD-d_4): δ 198.63, 169.04, 144.14, 139.97, 134.73, 130.96, 130.59, 130.56, 129.97, 129.77, 126.56, 124.67, 110.94, 104.74, 78.80, 74.91, 36.54. **ESI-MS** *m/z* (pos): cacld for $C_{19}H_{13}NO_3$ [M + H]$^+$ 304.1, found: 304.1; (neg) [M - H]$^-$ 302.1, found: 302.1.

General procedure A: Coupling of acids S5a-b to amines S6a-j. A solution of carboxcylic acid **S5a** or **S5b** (10mg, 0.033 mmol, 1equiv) and amine **S6a-j** (0.066 mmol, 2.0 equiv) in DMF (500 uL) and triethylamine (50 µL) was treated with EDC (7.5 mg, 0.040 mmol, 1.2 equiv) at rt. After 2hrs, the reaction was poured into sat aq NaCl and extracted with EtOAc and the organic layer was washed with sat aq NaCl, dried with Na_2SO_4 and concentrated under reduced pressure. Purification by silica gel chromatography (150:1→50:1 CH_2Cl_2/MeOH) yielded amides **1**–**11** and **14**–**21**.

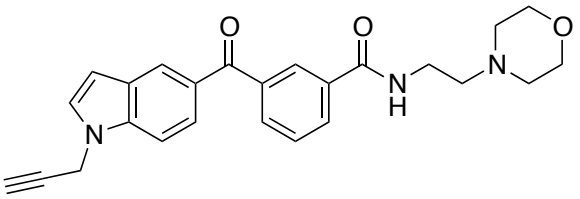


***tert*-butyl 4-(3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzoyl)piperazine-1-carb-oxylate (1).** Prepared using general procedure A from **S5a** and 1-(*tert*-butylcarbonyl-piperazine) **S6a** and yielded **1** (5.2 mg, 0.025 mmol, 75%). **^{1}H-NMR** (600 MHz, CDCl3): δ 8.10 (d, *J* = 1.4 Hz, 1H), 7.87 (d, *J* = 7.6 Hz, 1H), 7.82-7.78 (m, 2H), 7.64-7.63 (m, 1H), 7.57 (t, *J* = 7.6 Hz, 1H), 7.49 (d, *J* = 8.7 Hz, 1H), 7.31 (d, *J* = 3.2 Hz, 1H), 6.64 (d, *J* = 3.2 Hz, 1H), 4.94 (s, 2H), 3.75-3.71 (m, 2H), 3.51-3.40 (m, 6H), 2.46 (s, 1H), 1.46 (s, 9H).**^{13}C-NMR** (101 MHz,

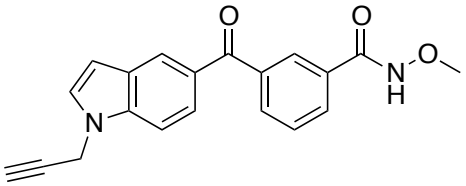
$CDCl_3$): δ 196.16, 169.82, 154.55, 139.35, 138.26, 135.51, 131.25, 130.33, 129.27, 129.17, 128.80, 128.28, 128.17, 125.53, 124.11, 109.55, 104.05, 80.44, 77.10, 74.30, 36.19, 28.42. **HRMS** *m/z*: cacld for $C_{28}H_{29}N_3O_4$ $[M + H]^+$ 472.2231, found: 472.2232.



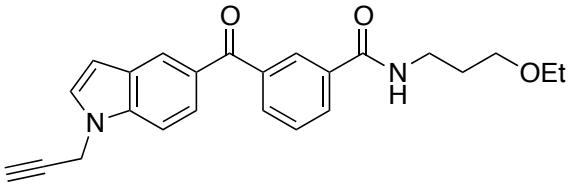
***N*-cyclopropyl-3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzamide (2).** Prepared using general procedure A from **S5a** and cyclopropylamine **S6b** and yielded **2** (10 mg, 0.029 mmol, 89%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.08-8.07 (m, 2H), 8.05-8.04 (m, 1H), 7.88 (dd, *J* = 7.6, 1.3 Hz, 1H), 7.80 (dd, *J* = 8.6, 1.6 Hz, 1H), 7.55 (t, *J* = 7.7 Hz, 1H), 7.49 (d, *J* = 8.6 Hz, 1H), 7.31 (d, *J* = 3.3 Hz, 1H), 6.64 (d, *J* = 3.2 Hz, 1H), 6.41 (s, 1H), 4.93 (d, *J* = 2.5 Hz, 2H), 2.92 (dt, *J* = 7.1, 3.5 Hz, 1H), 2.46 (t, *J* = 2.5 Hz, 1H), 0.89-0.85 (m, 2H), 0.65-0.62 (m, 2H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.35, 168.13, 139.13, 138.18, 134.56, 132.77, 130.61, 129.27, 129.05, 128.52, 128.23, 127.29, 125.50, 124.08, 109.45, 104.03, 77.1(obsc), 74.19, 36.13, 23.23, 6.75. **HRMS** *m/z*: cacld for $C_{22}H_{18}N_2O_2$ $[M + H]^+$ 343.1141, found: 343.1440.



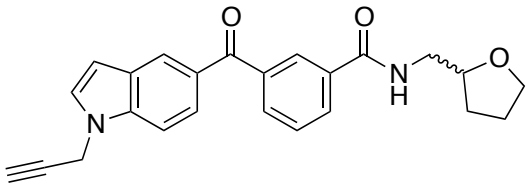
***N*-(2-morpholinoethyl)-3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzamide (3).** Prepared using general procedure A and from **S5a** 4-(2-aminoethyl)morpholine **S6c** and yielded **3** (10.5 mg, 0.025 mmol, 77%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.11 (d, *J* = 18.0 Hz, 2H), 8.06-8.05 (m, 1H), 7.92 (d, *J* = 7.2 Hz, 1H), 7.81 (s, 1H), 7.58 (s, 1H), 7.49 (d, *J* = 8.4 Hz, 1H), 7.31 (d, *J* = 0.8 Hz, 1H), 6.85 (s, 1H), 6.63 (s, 1H), 4.93 (s, 2H), 3.64 (s, 4H), 3.55 (s, 2H), 2.59 (s, 2H), 2.46 (s, 4H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 197.25, 167.57, 140.08, 139.04, 135.58, 133.51, 131.43, 130.19, 129.94, 129.46, 129.09, 128.51, 126.35, 124.93, 110.33, 104.86, 77.1(obsc), 75.07, 67.76, 57.67, 54.16, 37.00. **HRMS** *m/z*: cacld for $C_{25}H_{25}N_3O_3$ $[M + H]^+$ 416.1969, found: 416.1970.



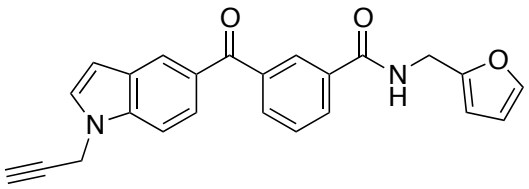
***N*-methoxy-3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzamide (4).** Prepared using general procedure A from **S5a** and methoxyamine•HCl **S6d** and yielded **4** (9.7 mg, 0.029 mmol, 88%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 9.08 (br, 1H), 8.10 (s, 1H), 8.07 (d, *J* = 1.4 Hz, 1H), 8.02 (d, *J* = 7.9 Hz, 1H), 7.92 (d, *J* = 7.7 Hz, 1H), 7.79 (dd, *J* = 8.6, 1.6 Hz, 1H), 7.57 (t, *J* = 7.7 Hz, 1H), 7.49 (d, *J* = 8.7 Hz, 1H), 7.31 (d, *J* = 3.3 Hz, 1H), 6.64 (d, *J* = 3.2 Hz, 1H), 4.93 (d, *J* = 2.5 Hz, 2H), 3.89 (s, 3H), 2.46 (t, *J* = 2.5 Hz, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.17, 165.70, 139.31, 138.23, 133.30, 132.04, 130.68, 129.66, 129.11, 128.72, 128.25, 127.65, 125.54, 124.07, 109.50, 104.05, 77.01, 74.21, 64.73, 36.14. **HRMS** *m/z*: cacld for $C_{20}H_{16}N_2O_3$ $[M + H]^+$ 333.1234, found: 333.1232.



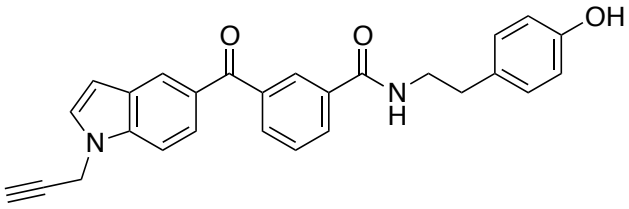
***N*-(3-ethoxypropyl)-3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzamide (5).** Prepared using general procedure A from **S5a** and 3-ethoxypropylamine **S6a** and yielded **5** (9.4 mg, 0.024 mmol, 73%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.13 (s, 1H), 8.09 (d, *J* = 1.2 Hz, 1H), 8.05 (d, *J* = 7.8 Hz, 1H), 7.89 (d, *J* = 7.6 Hz, 1H), 7.81 (dd, *J* = 8.6, 1.5 Hz, 1H), 7.56 (t, *J* = 7.7 Hz, 1H), 7.48 (d, *J* = 8.6 Hz, 1H), 7.30 (d, *J* = 3.2 Hz, 1H), 7.23 (br s, 1H), 6.64 (d, *J* = 3.2 Hz, 1H), 4.93 (d, *J* = 2.5 Hz, 2H), 3.60 (m, 5H), 3.45 (q, *J* = 7.0 Hz, 2H), 2.45 (t, *J* = 2.5 Hz, 1H), 1.89 (m, 2H), 1.13 (t, *J* = 7.0 Hz, 3H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.38, 166.29, 139.24, 138.16, 134.87, 132.48, 130.34, 129.42, 128.95, 128.42, 128.23, 127.52, 125.46, 124.08, 109.39, 104.03, 77.1 (obsc), 74.14, 70.52, 66.65, 39.67, 36.11, 28.73, 15.20. **HRMS** *m/z*: cacld for $C_{24}H_{24}N_2O_3$ $[M + H]^+$ 389.1860, found: 389.1858.



3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)-*N*-((tetrahydrofuran-2-yl)methyl)benzamide (6). Prepared using general procedure A from **S5a** and (±)-2-(tetrahydrofuran-2-yl)ethanamine **S6f** and yielded **6** (9.6 mg, 0.025 mmol, 75%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.16 (s, 1H), 8.10 (d, *J* = 1.0 Hz, 1H), 8.05-8.03 (m, 1H), 7.90 (dd, *J* = 7.6, 1.2 Hz, 1H), 7.82 (dd, *J* = 8.6, 1.6 Hz, 1H), 7.56 (t, *J* = 7.7 Hz, 1H), 7.49 (d, *J* = 8.6 Hz, 1H), 7.31 (d, *J* = 3.3 Hz, 1H), 6.64 (d, *J* = 3.2 Hz, 1H), 6.59 (br s, 1H), 4.94 (d, *J* = 2.5 Hz, 2H), 4.07 (qd, *J* = 7.2, 3.3 Hz, 1H), 3.89-3.85 (m, 1H), 3.81-3.74 (m, 2H), 3.36 (m, 1H), 2.46 (t, *J* = 2.5 Hz, 1H), 2.05-2.00 (m, 1H), 1.91 (quintet, *J* = 7.2 Hz, 2H), 1.64-1.60 (m, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.31, 166.84, 139.28, 138.19, 134.68, 132.67, 130.42, 129.34, 128.99, 128.48, 128.24, 127.73, 125.51, 124.10, 109.43, 104.04, 77.62, 77.1 (obsc), 74.17, 68.15, 43.72, 36.12, 28.69, 25.90. **HRMS** *m/z*: cacld for $C_{24}H_{22}N_2O_3$ $[M + H]^+$ 387.1703, found: 387.1702.

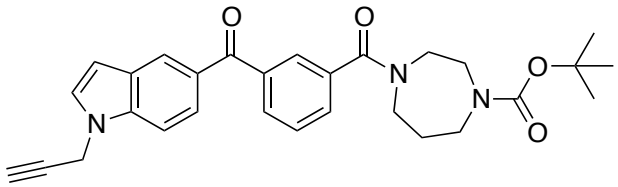


***N*-(furan-2-ylmethyl)-3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzamide (7).** Prepared using general procedure A from **S5a** and 2-(furan-2yl)ethanamine **S6g** and yielded **7** (11.1 mg, 0.029 mmol, 88%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.15 (t, *J* = 1.5 Hz, 1H), 8.09 (d, *J* = 1.3 Hz, 1H), 8.07 (dt, *J* = 7.8, 1.4 Hz, 1H), 7.90 (dt, *J* = 7.6, 1.3 Hz, 1H), 7.80 (dd, *J* = 8.6, 1.6 Hz, 1H), 7.56 (t, *J* = 7.7 Hz, 1H), 7.48 (d, *J* = 8.6 Hz, 1H), 7.37 (d, *J* = 1.0 Hz, 1H), 7.31 (d, *J* = 3.2 Hz, 1H), 6.64 (d, *J* = 3.2 Hz, 1H), 6.55 (br s, 1H), 6.34-6.30 (m, 2H), 4.93 (d, *J* = 2.5 Hz, 2H), 4.65 (d, *J* = 5.5 Hz, 2H), 2.46 (t, *J* = 2.5 Hz, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.26, 166.41, 150.84, 142.41, 139.26, 138.19, 134.31, 132.89, 130.62, 129.27, 129.03, 128.54, 128.23, 127.57, 125.50, 124.08, 110.52, 109.45, 107.91, 104.04, 77.1(obsc), 74.18, 37.08, 36.12. **HRMS** *m/z*: cacld for $C_{24}H_{18}N_2O_3$ $[M + H]^+$ 383.1390, found: 383.1391.

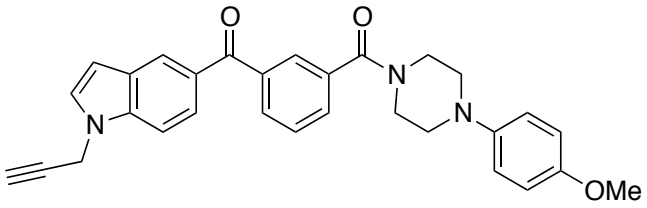


***N*-(4-hydroxyphenethyl)-3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzamide (8).** Prepared using general procedure A from **S5a** and 4-(2-aminoethyl)phenol **S6h** and yielded **8** (6.0 mg, 0.014 mmol, 43%). **^{1}H-**

NMR (600 MHz, $CDCl_3$): δ 8.08 (d, *J* = 1.3 Hz, 1H), 8.01-7.99 (m, 2H), 7.90 (d, *J* = 7.6 Hz, 1H), 7.80-7.78 (m, 1H), 7.55 (t, *J* = 7.7 Hz, 1H), 7.50 (d, *J* = 8.7 Hz, 1H), 7.32 (d, *J* = 3.2 Hz, 1H), 7.05 (d, *J* = 8.3 Hz, 2H), 6.71 (d, *J* = 8.4 Hz, 2H), 6.65 (d, *J* = 3.2 Hz, 1H), 6.21 (br t, *J* = 5.2 Hz, 1H), 5.37 (br s, 1H), 4.95 (d, *J* = 2.5 Hz, 2H), 3.67 (q, *J* = 6.4 Hz, 2H), 2.85 (t, *J* = 6.9 Hz, 2H), 2.46 (t, *J* = 2.5 Hz, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.39, 166.78, 154.50, 139.13, 138.19, 134.73, 132.71, 130.58, 130.52, 129.88, 129.31, 129.08, 128.64, 128.23, 127.46, 125.49, 124.11, 115.59, 109.45, 104.04, 77.06, 74.21, 41.41, 36.14, 34.74. **HRMS** *m/z*: cacld for $C_{27}H_{22}N_2O_3$ $[M + H]^+$ 423.1703, found: 423.1704.

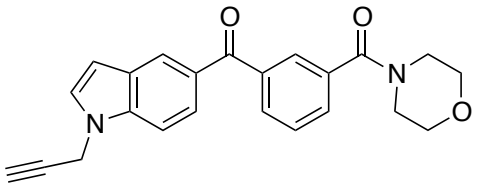


***tert*-butyl 4-(3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzoyl)-1,4-diazepane-1-carboxylate (9).** Prepared using general procedure A from **S5a** and 1-(*tert*-butylcorbonyl)homopiperazine **S6i** and yielded **9** (11.6 mg, 0.024 mmol, 73%). **^{1}H-NMR** (500 MHz, $CDCl_3$): δ 8.10 (s, 1H), 7.86-7.77 (m, 3H), 7.60-7.53 (m, 2H), 7.48 (d, *J* = 8.7 Hz, 1H), 7.30 (d, *J* = 3.2 Hz, 1H), 6.63 (d, *J* = 2.5 Hz, 1H), 4.94-4.91 (m, 2H), 3.79 (m, 1H), 3.68 (m, 1H), 3.61 (m, 1H), 3.50-3.40 (m, 5H), 2.45 (m, 1H), 1.99 (m, 1H), 1.69 (m, 1H), 1.50-1.33 (m, 9H). **^{13}C-NMR** (126 MHz, $CDCl_3$): δ 196.35, 171.09, 155.21, 143.96, 139.31, 138.32, 130.90, 129.39, 129.17, 129.11, 128.74, 128.36, 127.85, 125.62, 124.22, 109.57, 104.16, 80.06, 77.1(obsc), 74.30, 50.41, 48.97, 48.59, 47.32, 45.67, 36.25, 28.52. **HRMS** *m/z*: cacld for $C_{29}H_{31}N_3O_4$ $[M + H]^+$ 486.2387, found: 486.2389.

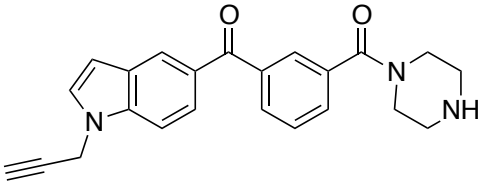


(4-(4-methoxyphenyl)piperazin-1-yl)(3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)phenyl)methanone (10). Prepared using general procedure A from **S5a** and 1-(4methoxyphenyl)piperazine **S6j** and yielded **10** (9.0 mg, 0.019 mmol, 58%). **^{1}H-NMR** (500 MHz, $CDCl_3$): δ 8.11-8.11 (m, 1H), 7.88 (dt, *J* = 7.7, 1.5 Hz, 1H), 7.82 (m, 2H), 7.67 (dt, *J* = 7.7, 1.5 Hz, 1H), 7.57 (t, *J* = 7.7 Hz, 1H), 7.49 (d, *J* = 8.7 Hz, 1H), 7.31 (d, *J* = 3.3 Hz, 1H), 6.89 (d, *J* = 9.1 Hz, 2H), 6.84 (d, *J* = 9.2 Hz, 2H), 6.64 (dd, *J* = 3.3, 0.8 Hz, 1H), 4.93 (d, *J* = 2.6 Hz, 2H), 3.94 (s, 2H), 3.77 (s, 3H), 3.59 (s, 2H), 3.17-2.99 (m, 4H), 2.45 (t, *J* = 2.6 Hz, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ

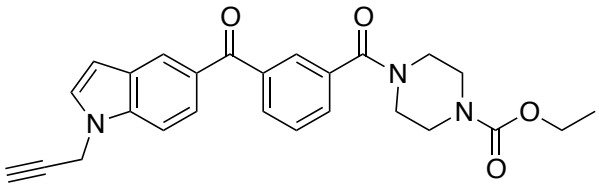
196.37, 171.10, 155.20, 139.39, 138.30, 136.45, 130.91, 130.02, 129.58, 129.35, 129.17, 128.75, 128.33, 127.85, 127.59, 125.63, 124.21, 109.58, 104.14, 80.06, 77.1(obsc), 74.30, 50.40, 48.59, 47.31, 45.66, 45.13, 36.24. **HRMS** *m/z*: cacld for $C_{30}H_{27}N_3O_3$ $[M + H]^+$ 478.2120, found: 478.2126.



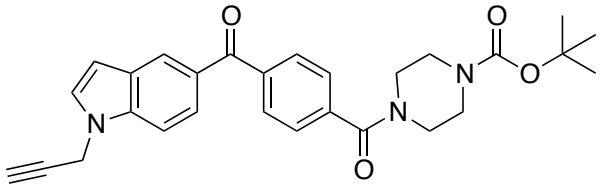
(3-(morpholine-4-carbonyl)phenyl)(1-(prop-2-yn-1-yl)-1*H*-indol-5-yl)methanone (11). Prepared using general procedure A from **S5a** and morpholine **S6k** and yielded **11** (9.6 mg, 0.016 mmol, 79%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.10 (d, *J* = 0.9 Hz, 1H), 7.87 (d, *J* = 7.7 Hz, 1H), 7.83-7.79 (m, 2H), 7.64 (d, *J* = 7.6 Hz, 1H), 7.56 (t, *J* = 7.6 Hz, 1H), 7.49 (d, *J* = 8.6 Hz, 1H), 7.31 (d, *J* = 3.2 Hz, 1H), 6.64 (d, *J* = 3.2 Hz, 1H), 4.94 (d, *J* = 2.5 Hz, 2H), 3.78-3.46 (m, 8H), 2.46 (t, *J* = 2.5 Hz, 1H). **^{13}C-NMR** (151 MHz, CDCl3): δ 196.33, 169.75, 139.43, 138.34, 135.46, 131.30, 130.45, 129.35, 129.21, 128.87, 128.35, 128.27, 125.68, 124.23, 109.60, 104.16, 77.1(obsc), 74.33, 66.97, 42.75, 36.28. **HRMS** *m/z*: cacld for $C_{23}H_{20}N_2O_3$ $[M + H]^+$ 373.1047, found: 373.1546.



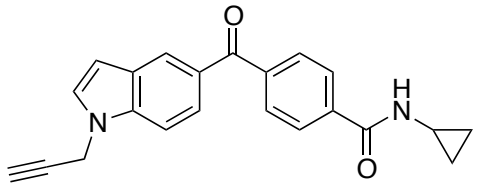
piperazin-1-yl(3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)phenyl)methanone (12). A solution of **1** (50 mg, 0.106 mmol, 1.0 equiv) in CH_2Cl_2 (2mL) was treated with trifluoroacetic acid (600 µL, added dropwise). After stirring for 15 min at rt the reaction was concentrated. Purification by silica gel chromatography (10:1 CH_2Cl_2/MeOH) yielded **12** (30 mg, 81 µmol, 76%) **^{1}H-NMR** (600 MHz, MeOD-d_4): δ 7.99 (d, *J* = 1.4 Hz, 1H), 7.79 (dt, *J* = 7.5, 1.5 Hz, 1H), 7.68-7.66 (m, 2H), 7.61-7.53 (m, 3H), 7.35 (d, *J* = 3.2 Hz, 1H), 6.56 (s, 1H), 5.00 (d, *J* = 2.5 Hz, 2H), 3.67 (br s, 2H), 3.41 (br s, 2H), 2.83-2.74 (m, 5H). **^{13}C-NMR** (151 MHz, MeOD-d_4): δ 197.64, 171.00, 140.10, 139.37, 136.25, 131.66, 130.84, 130.41, 129.51, 129.42, 128.64, 125.86, 124.12, 110.40, 104.11, 78.26, 74.36, 49.01, 39.85, 35.98, 30.22. **HRMS** *m/z*: cacld for $C_{23}H_{21}N_3O_2$ $[M + H]^+$ 372.1706, found: 372.1708.



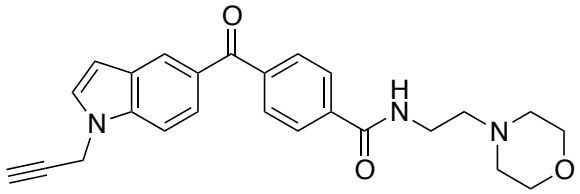
ethyl 4-(3-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzoyl)piperazine-1-carboxylate (13). A solution of **12** (10mg, 0.035 mmol, 1.0 equiv) and *N,N*-diisopropylethylamine (31 µL, 0.18 mmol, 5 equiv) in CH_2Cl_2 was cooled to 0 °C and treated with an ethylchloroformate solution (6.9 µL, 0.070 mmol, 2.0 equiv in 0.5 mL CH_2Cl_2). After warming to rt and stirring for 1 hr, the reaction was poured into sat aq NaCl and extracted with EtOAc and the organic layer was washed with sat aq NaCl, dried with Na_2SO_4 and concentrated under reduced pressure. Purification by silica gel chromatography (150:1→50:1 CH_2Cl_2/MeOH) yielded **13** (4.1 mg, 0.009 mmol, 50%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.10-8.10 (m, 1H), 7.88 (d, *J* = 7.6 Hz, 1H), 7.82-7.79 (m, 2H), 7.65-7.63 (m, 1H), 7.57 (t, *J* = 7.6 Hz, 1H), 7.49 (d, *J* = 8.6 Hz, 1H), 7.31 (d, *J* = 3.2 Hz, 1H), 6.64 (s, 1H), 4.16 (q, *J* = 7.1 Hz, 2H), 3.76-3.44 (m, 8H), 2.46 (t, *J* = 2.5 Hz, 1H), 1.27 (t, *J* = 7.1 Hz, 3H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.29, 169.96, 155.48, 139.47, 138.35, 135.49, 131.41, 130.43, 129.35, 129.23, 128.93, 128.37, 128.26, 125.67, 124.24, 109.62, 104.17, 77.0(obsc), 74.36, 61.93, 47.63, 43.50, 36.30, 14.78. **HRMS** *m/z*: cacld for $C_{26}H_{25}N_3O_4$ $[M + H]^+$ 444.1010, found: 444.1019.



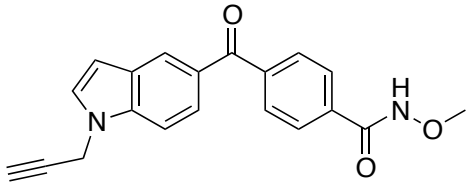
***tert*-butyl 4-(4-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzoyl)piperazine-1-carboxylate (14).** Prepared using general procedure A from **S5b** and 1-(*tert*-butylcarbonyl-piperazine) **S6a** and yielded **14** (10.4 mg, 0.022 mmol, 67%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.10 (s, 1H), 7.83 (d, *J* = 6.1 Hz, 3H), 7.49 (dd, *J* = 14.2, 8.2 Hz, 3H), 7.30 (s, 1H), 6.63 (s, 1H), 4.92 (s, 2H), 3.77 (br s, 2H), 3.54-3.41 (m, 6H), 2.45 (s, 1H), 1.47 (s, 9H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.36, 169.91, 154.63, 140.43, 138.43, 138.31, 130.14, 129.34, 129.18, 128.30, 126.89, 125.76, 124.15, 109.56, 104.10, 80.58, 77.0(obsc), 74.30, 47.61, 42.20, 36.23, 28.48. **HRMS** *m/z*: cacld for $C_{28}H_{29}N_3O_4$ $[M + H]^+$ 472.2231, found: 472.2233.



***N*-cyclopropyl-4-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzamide (15).** Prepared using general procedure A from **S5b** and cyclopropylamine **S6b** and yielded **15** (7.9 mg, 0.023 mmol, 70%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.07 (d, *J* = 1.4 Hz, 1H), 7.85-7.81 (m, 5H), 7.48 (d, *J* = 8.6 Hz, 1H), 7.31 (d, *J* = 3.3 Hz, 1H), 6.63 (dd, *J* = 3.2, 0.7 Hz, 1H), 6.36 (s, 1H), 4.94 (s, 2H), 2.95 (m, 1H), 2.46 (t, *J* = 2.6 Hz, 1H), 0.93-0.88 (m, 2H), 0.68-0.65 (m, 2H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 197.15, 169.07, 142.44, 139.07, 137.83, 130.82, 130.16, 129.92, 129.06, 127.52, 126.48, 124.91, 110.30, 104.87, 77.0(obsc), 75.05, 37.00, 24.13, 7.73, 0.87. **HRMS** *m/z*: cacld for $C_{22}H_{18}N_2O_2$ $[M + H]^+$ 343.1441, found: 343.1442.

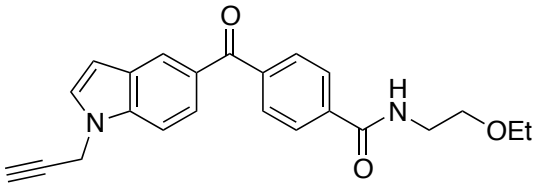


***N*-(2-morpholinoethyl)-4-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzamide (16).** Prepared using general procedure A from **S5b** and 4-(2-aminoethyl)morpholine **S6c** and yielded **16** (9.1 mg, 0.022 mmol, 67%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.08 (s, 1H), 7.86 (m, 5H), 7.48 (d, *J* = 8.4 Hz, 1H), 7.30 (s, 1H), 6.89 (s, 1H), 6.62 (s, 1H), 4.92 (s, 2H), 3.73 (s, 4H), 3.59 (s, 2H), 2.63 (s, 2H), 2.52 (s, 4H), 2.45 (s, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 197.14, 167.59, 142.40, 139.07, 138.04, 130.83, 130.16, 129.94, 129.06, 127.61, 126.47, 124.91, 110.31, 104.85, 77.90, 77.0(obsc), 75.07, 67.86, 57.68, 54.20, 37.00. **HRMS** *m/z*: cacld for $C_{25}H_{25}N_3O_3$ $[M + H]^+$ 416.1969, found: 416.1968.

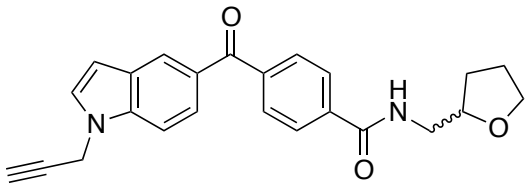


***N*-methoxy-4-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzamide (17).** Prepared using general procedure A from **S5b** and methoxyamine•HCl **S6d** and yielded **17** (7.5 mg, 0.023 mmol, 68%). **^{1}H-NMR** (600 MHz, MeOD-d_4): δ 8.05 (d, *J* = 1.0 Hz, 1H), 7.83 (d, *J* = 8.2 Hz, 2H), 7.80-7.77 (m, 3H), 7.46 (d, *J* = 8.7 Hz, 1H),

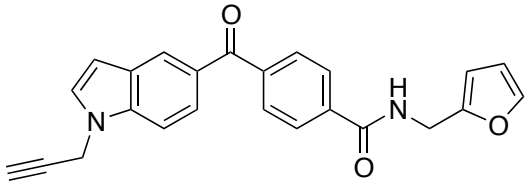
7.29 (d, *J* = 3.2 Hz, 1H), 6.60 (d, *J* = 3.1 Hz, 1H), 4.91 (d, *J* = 2.5 Hz, 2H), 3.89 (m, 3H), 2.44 (t, *J* = 2.5 Hz, 1H). **^{13}C-NMR** (151 MHz, MeOD-d_4): δ 197.53, 167.67, 142.81, 139.16, 135.46, 130.73, 130.03, 129.88, 129.07, 127.86, 126.61, 124.88, 110.37, 104.87, 77.81, 75.06, 65.33, 36.97. **HRMS** *m/z*: cacld for $C_{20}H_{16}N_2O_3$ [M + H]$^+$ 333.1234, found: 333.1232.



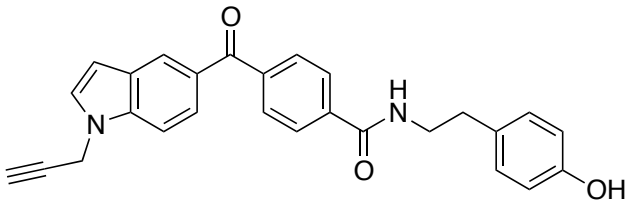
***N*-(2-ethoxyethyl)-4-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzamide (18).** Prepared using general procedure A from **S5b** and 3-ethoxypropylamine **S6a** and yielded **18** (9.8 mg, 0.025 mmol, 76%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.09 (d, *J* = 1.1 Hz, 1H), 7.89-7.87 (m, 2H), 7.83 (m, 3H), 7.49 (d, *J* = 8.6 Hz, 1H), 7.31 (m, 2H), 6.63 (dd, *J* = 3.2, 0.7 Hz, 1H), 4.94 (d, *J* = 2.5 Hz, 2H), 3.64 (td, *J* = 11.5, 5.5 Hz, 4H), 3.54 (q, *J* = 7.0 Hz, 2H), 2.46 (t, *J* = 2.5 Hz, 1H), 1.92 (dt, *J* = 11.4, 5.7 Hz, 2H), 1.25 (t, *J* = 7.0 Hz, 4H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.41, 166.33, 141.31, 138.18, 137.43, 129.92, 129.41, 129.01, 128.19, 126.63, 125.59, 124.07, 109.40, 104.00, 74.17, 70.87, 66.72, 39.90, 36.12, 28.67, 15.42. **HRMS** *m/z*: cacld for $C_{24}H_{24}N_2O_3$ [M + H]$^+$ 389.1860, found: 389.1858.



4-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)-*N*-((tetrahydrofuran-2-yl)methyl)benzamide (19). Prepared using general procedure A from **S5b** and (±)-2-(tetrahydrofuran-2-yl)ethanamine **S6f** and yielded **19** (8.1 mg, 0.021 mmol, 63%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.09 (z, 1H), 7.89 (m, 2H), 7.85-7.82 (m, 3H), 7.49 (d, *J* = 8.6 Hz, 1H), 7.31 (d, *J* = 3.3 Hz, 1H), 6.64-6.60 (m, 2H), 4.94 (d, *J* = 2.5 Hz, 2H), 4.12-4.08 (m, 1H), 3.93-3.89 (m, 1H), 3.84 (m, 1H), 3.79 (m, 1H), 3.37 (m, 1H), 2.06 (m, 1H), 1.95 (m, 2H), 1.67-1.62 (m, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.30, 166.87, 141.54, 138.20, 137.09, 129.95, 129.35, 129.02, 128.19, 126.79, 125.62, 124.07, 109.42, 104.01, 77.71, 74.18, 77.0(obsc), 68.21, 43.76, 36.13, 28.71, 25.94. **HRMS** *m/z*: cacld for $C_{24}H_{22}N_2O_3$ [M + H]$^+$ 387.1703, found: 387.1704.



***N*-(furan-2-ylmethyl)-4-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzamide (20).** Prepared using general procedure A from **S5b** and 2-(furan-2yl)ethanamine **S6g** and yielded **20** (5.8 mg, 0.015 mmol, 46%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.07 (s, 1H), 7.89-7.80 (m, 5H), 7.48 (d, *J* = 8.6 Hz, 1H), 7.39 (d, *J* = 0.8 Hz, 1H), 7.30-7.30 (m, 1H), 6.62-6.62 (m, 1H), 6.50 (s, 1H), 6.36-6.33 (m, 2H), 4.93 (s, 2H), 4.68 (s, 2H), 2.45 (s, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.39, 166.65, 150.98, 149.78, 142.62, 141.88, 138.36, 136.83, 130.13, 129.19, 128.34, 126.97, 125.77, 124.20, 110.73, 109.58, 108.10, 104.16, 77.0(obsc), 74.33, 37.28, 36.28. **HRMS** *m/z*: cacld for $C_{24}H_{18}N_2O_3$ $[M + H]^+$ 383.1390, found: 383.1389.



***N*-(4-hydroxyphenethyl)-4-(1-(prop-2-yn-1-yl)-1*H*-indole-5-carbonyl)benzamide (21).** Prepared using general procedure A from **S5b** and 4-(2-aminoethyl)phenol **S6h** and yielded **21** (9.1mg, 0.022 mmol, 67%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.08 (d, *J* = 1.2 Hz, 1H), 7.82-7.78 (m, 5H), 7.48 (d, *J* = 8.7 Hz, 1H), 7.30 (d, *J* = 3.2 Hz, 1H), 7.10 (d, *J* = 8.5 Hz, 2H), 6.82 (d, *J* = 8.5 Hz, 2H), 6.63 (dd, *J* = 3.2, 0.7 Hz, 1H), 6.26 (t, *J* = 5.5 Hz, 1H), 5.84 (s, 1H), 4.93 (d, *J* = 2.5 Hz, 2H), 3.71 (q, *J* = 6.4 Hz, 2H), 2.89 (t, *J* = 6.8 Hz, 2H), 2.45 (t, *J* = 2.5 Hz, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 196.66, 167.18, 155.04, 141.81, 138.50, 137.46, 130.66, 130.26, 130.19, 129.52, 129.34, 128.48, 126.94, 125.93, 124.35, 115.97, 109.73, 104.30, 77.0(obsc), 74.48, 41.74, 36.41, 34.99. **HRMS** *m/z*: cacld for $C_{27}H_{22}N_2O_3$ $[M + H]^+$ 423.1703, found: 423.1703.

Synthesis of 7-benzoyl-benzo-1,4-diazepin-2,5-dione (BzBDs).

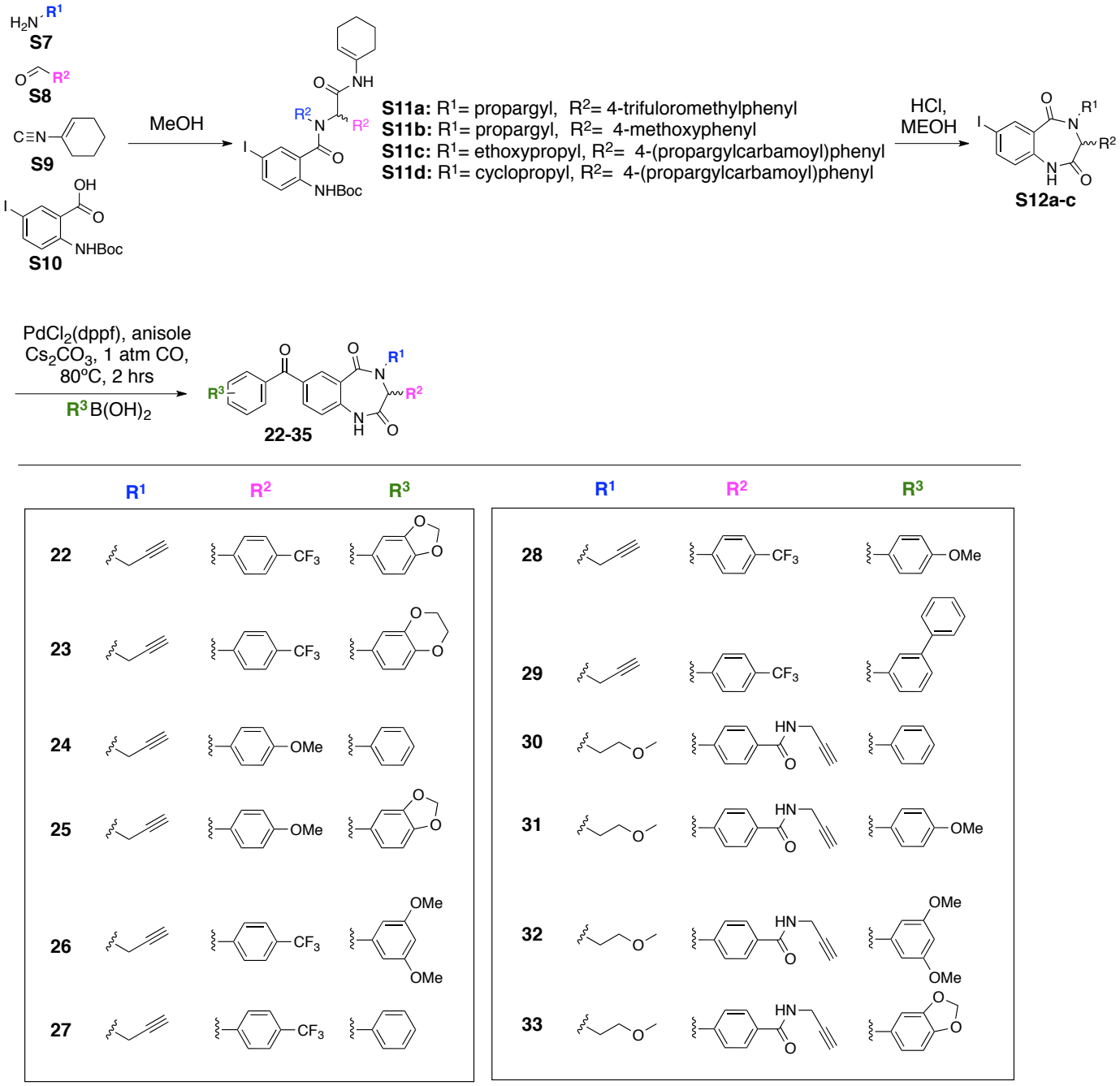
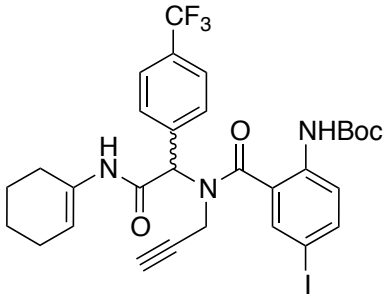
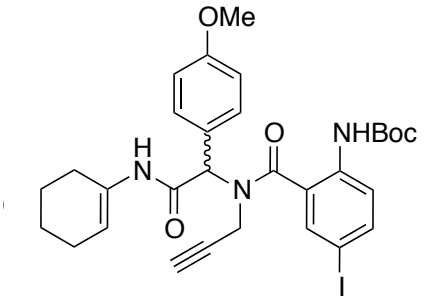


Figure S7. Synthesis of 7-benzoyl-benzo-1,4-diazepin-2,5-dione (BzBDs) 22-33. (Boc = *t*-butoxy-carbonyl).

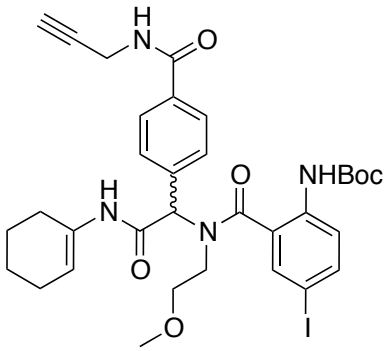


***tert*-butyl (2-((2-(cyclohex-1-en-1-ylamino)-2-oxo-1-(4-(trifluoromethyl)phenyl)ethyl)(prop-2-yn-1-yl)carbamoyl)-4-iodophenyl)carbamate (S11a).** A solution of 4-trifluoromethylbenzaldehyde (40 µL, 0.3 mmol, 1.0 equiv), propargyl amine (19 µL, 0.3 mmol, 1.0 equiv) in MeOH (5mL) was stirred at rt for 30 min. To this solution was added scandium trifluoromethanesulfonate (20 mg, 0.003 mmol, 0.1 equiv), isonitrile[4,5] **S9** (300 µL of a 1M solution, 0.3 mmol, 1.0 equiv), and anthranilic acid **S10** (108 mg, 0.3 mmol, 1.0 equiv). After 18 hr at rt, the reaction was concentrated under reduced pressure. Purification by silica gel chromatography (5:1 hexanes/EtOAc) yielded **S11a** (133 mg, 0.20 mmol, 65%). **^{1}H-NMR** (500 MHz, $CDCl_3$): δ 7.95 (m, 2H), 7.68-7.62 (m, 6H), 7.12 (br s, 1H), 6.13 (s, 1H), 5.89 (br s, 1H), 4.12-4.00 (m, 2H), 2.10 (m, 5H), 1.65 (m, 2H), 1.59-1.53 (m, 2H), 1.51 (s, 9H). **^{13}C-NMR** (126 MHz, $CDCl_3$): δ 170.43, 166.63, 153.10, 140.46, 138.10, 137.20, 135.69, 132.66, 131.84, 131.58, 130.59, 126.36, 125.26, 123.09, 81.35, 78.43, 73.64, 60.83, 53.86, 42.38, 28.66, 28.20, 27.42, 24.37, 22.82, 22.22. **ESI-MS** *m/z* (neg): cacld for $C_{30}H_{31}F_3IN_3O_4$ $[M - H]^-$ 680.1, found: 680.1.

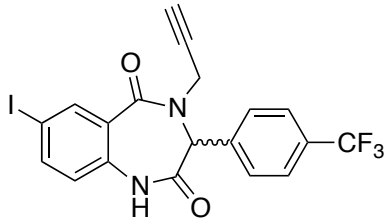


***tert*-butyl (2-((2-(cyclohex-1-en-1-ylamino)-1-(4-methoxyphenyl)-2-oxoethyl)(prop-2-yn-1-yl)carbamoyl)-4-iodophenyl)carbamate (S11b).** A solution of anisaldehyde (132 mg, 1.0 mmol, 1.0 equiv), propargyl amine (64 µL, 1.0 mmol, 1.0 equiv) in MeOH (5mL) was stirred at rt for 30 min. To this solution was added scandium trifluoromethanesulfonate (49 mg, 0.1 mmol, 0.1 equiv), isonitrile[5] **S9** (1 mL of a 1M solution, 1.0 mmol, 1.0 equiv), and anthranilic acid **S10** (362 mg, 1.0 mmol, 1.0 equiv). After 18 hr at rt, the reaction was concentrated under reduced pressure. Purification by silica gel chromatography (4:1 hexanes/EtOAc) yielded **S11b** (345 mg, 0.53 mmol, 53%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.23-7.99 (m, 2H), 7.76-7.64 (m, 2H), 7.42 (t, *J* = 1.0 Hz, 2H),

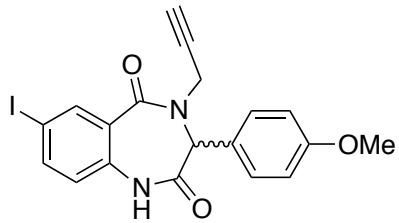
6.93 (d, *J* = 8.3 Hz, 2H), 6.73 (br s, 1H), 6.12 (s, 1H), 5.89 (br s, 1H), 3.93 (m, 2H), 3.63 (s, 3H), 2.09-2.04 (m, 5H), 1.65 (m, 2H), 1.57-1.54 (m, 2H), 1.51 (s, 9H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 169.41, 167.37, 160.39, 152.96, 143.24, 142.44, 140.07, 139.67, 136.60, 135.23, 132.36, 131.80, 125.13, 114.63, 83.24, 81.01, 72.74, 63.58, 55.47, 34.78, 31.70, 28.40, 27.91, 22.78, 22.52, 21.95. **ESI-MS** *m/z* (neg): cacld for $C_{30}H_{34}IN_3O_5$ $[M - H]^-$ 642.1, found: 642.1.



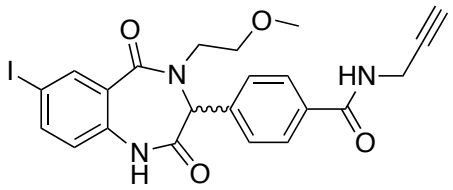
***tert*-butyl(2-((2-(cyclohex-1-en-1-ylamino)-2-oxo-1-(4-(prop-2-yn-1-ylcarbamoyl)phenyl)ethyl)(2-methoxyethyl)carbamoyl)-4-iodophenyl)carbamate (S11c).** A solution of 4-(propargylcarbamoyl)benzaldehyde (186 mg, 1.0 mmol, 1.0 equiv), 2-methoxyethylamine (87 µL, 1.0 mmol, 1.0 equiv) in MeOH (5mL) was stirred at rt for 30 min. To this solution was added scandium trifluoromethanesulfonate (49 mg, 0.1 mmol, 0.1 equiv), isonitrile[5] **S9** (1 mL of a 1M solution, 1.0 mmol, 1.0 equiv), and anthranilic acid **S10** (362 mg, 1.0 mmol, 1.0 equiv). After 18 hr at rt, the reaction was concentrated under reduced pressure. Purification by silica gel chromatography (1:4 hexanes/EtOAc) yielded **S11c** (400 mg, 0.56 mmol, 56%). **^{1}H-NMR** (500 MHz, $CDCl_3$): δ 8.04 (br s, 1H), 7.75 (m, 4H), 7.54 (dd, *J* = 8.8, 1.2 Hz, 1H), 7.42 (m, 3H), 6.06 (s, 1H), 5.64 (br s, 1H), 4.09 (m, 2H), 3.33 (br s, 2H), 3.01 (br s, 5H), 2.19 (t, *J* = 2.4 Hz, 1H), 2.05-2.01 (m, 4H), 1.58 (m, 2H), 1.50 (m, 2H), 1.43 (s, 9H). **^{13}C-NMR** (126 MHz, $CDCl_3$): δ 171.10, 166.97, 166.39, 152.84, 139.01, 138.06, 135.92, 135.27, 133.93, 132.59, 129.57, 127.74, 127.01, 122.69, 85.08, 83.00, 80.68, 79.65, 71.42, 69.44, 69.32, 60.31, 58.53, 29.52, 28.22, 27.57, 23.93, 22.40, 21.79. **ESI-MS** *m/z* (neg): cacld for $C_{33}H_{39}IN_4O_6$ $[M - H]^-$ 713.2, found: 713.2.



7-iodo-4-(prop-2-yn-1-yl)-3-(4-(trifluoromethyl)phenyl)-3,4-dihydro-1*H*-benzo[*e*][1,4]diazepine-2,5-dione (S12a). A solution of **S11a** (589 mg, 0.87 mmol) dissoved in a solution of 1:4 AcCl:MeOH (5 mL). After stirring at rt for 18 hrs, the solution was poured into EtOAc and the organic layer was washed with 1 N NaOH and sat aq NaCl, dried with Na_2SO_4 and concentrated under reduced pressure. Purification by silica gel chromatography (150:1→100:1 CH_2Cl_2/MeOH) yielded **S12a** (200 mg, 0.41 mmol, 48%). (**^{1}H-NMR** (600 MHz, MeOD-d_4): δ 7.87 (d, *J* = 1.9 Hz, 1H), 7.57 (dd, *J* = 8.5, 2.0 Hz, 1H), 7.49 (d, *J* = 8.3 Hz, 2H), 7.35 (d, *J* = 7.7 Hz, 2H), 6.63 (d, *J* = 8.5 Hz, 1H), 5.70 (s, 1H), 4.90 (d, *J* = 2.4 Hz, 1H), 4.59 (dd, *J* = 17.3, 0.4 Hz, 1H), 2.85 (t, *J* = 2.3 Hz, 1H). **^{13}C-NMR** (151 MHz, MeOD-d_4): δ 170.70, 166.36, 142.06, 139.72, 138.82, 135.87, 130.70, 130.66, 129.04, 126.25, 126.06, 126.04, 122.85, 87.57, 77.85, 74.65, 66.47, 39.61, 28.12. **ESI-MS** *m/z* (neg): cacld for $C_{19}H_{12}F_3IN_2O_2$ $[M - H]^-$ 483.0, found: 483.0.

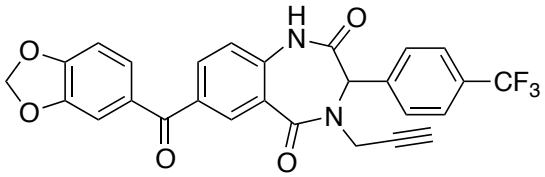


7-iodo-3-(4-methoxyphenyl)-4-(prop-2-yn-1-yl)-3,4-dihydro-1*H*-benzo[*e*][1,4]diazepine-2,5-dione (S12b). A solution of **S11b** (345 mg, 0.54 mmol) dissoved in a solution of 1:4 AcCl:MeOH (5 mL). After stirring at rt for 18 hrs, the solution was poured into EtOAc and the organic layer was washed with 1 N NaOH and sat aq NaCl, dried with Na_2SO_4 and concentrated under reduced pressure. Purification by silica gel chromatography (150:1→100:1 CH_2Cl_2/MeOH) yielded **S12b** (106 mg, 0.238 mmol, 44%). **^{1}H-NMR** (500 MHz, $CDCl_3$): δ 9.74 (m, 1H), 8.04 (s, 1H), 7.48 (d, *J* = 8.1 Hz, 1H), 7.08-7.06 (m, 2H), 6.68 (d, *J* = 7.4 Hz, 2H), 6.62 (d, *J* = 8.4 Hz, 1H), 5.57 (s, 1H), 4.97 (d, *J* = 17.4 Hz, 1H), 4.52-4.42 (m, 1H), 3.70 (s, 3H), 2.36 (s, 1H). **^{13}C-NMR** (126 MHz, $CDCl_3$): δ 171.42, 165.33, 159.27, 141.18, 139.57, 136.00, 134.39, 128.43, 126.21, 124.49, 121.83, 114.11, 88.10, 77.50, 73.84, 55.26, 30.99. **ESI-MS** *m/z* (neg): cacld for $C_{19}H_{15}IN_2O_3$ $[M - H]^-$ 445.0, found: 445.0.



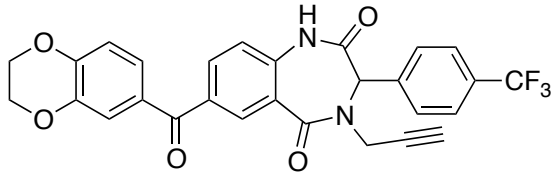
7-iodo-4-(2-methoxyethyl,)-3-(4-(prop-2-yn-1-ylcarbamoyl)phenyl)-3,4-dihydro-1*H*-benzo[*e*][1,4]diazepine-2,5-dione (S12c). A solution of **S11c** (400 mg, 0.79 mmol) dissoved in a solution of 1:4 AcCl:MeOH (5 mL). After stirring at rt for 18 hrs, the solution was poured into EtOAc and the organic layer was washed with 1 N NaOH and sat aq NaCl, dried with Na_2SO_4 and concentrated under reduced pressure. Purification by silica gel chromatography (150:1→100:1 CH_2Cl_2/MeOH) yielded **S12c** (250 mg, 0.484 mmol, 61%). **^{1}H-NMR** (600 MHz, MeOD-d_4): δ 7.86 (s, 1H), 7.64 (d, *J* = 8.3 Hz, 2H), 7.52-7.51 (m, 1H), 7.19 (d, *J* = 8.0 Hz, 2H), 6.60 (d, *J* = 8.5 Hz, 1H), 5.58 (s, 1H), 4.15-3.99 (m, 4H), 3.77-3.67 (m, 2H), 3.33 (m, 3H), 2.56 (t, *J* = 2.5 Hz, 1H). **^{13}C-NMR** (151 MHz, MeOD-d_4): δ 171.47, 168.26, 167.24, 141.74, 139.69, 138.55, 135.81, 134.11, 129.63, 128.15, 125.50, 122.64, 87.47, 80.01, 71.50, 70.86, 68.10, 58.42, 51.18, 29.32. **ESI-MS** *m/z* (neg): cacld for $C_{22}H_{20}IN_3O_4$ $[M - H]^-$ 516.0, found: 516.0.

General Procedure B: Coupling of aryl iodides S12a-c to boronic acids. To a 4 mL vial with a screwtop septa top was added aryl iodide **S12a** (7.4 mg, 15 µmol, 1 equiv), 3,4-methylene-dioxyphenyl boronic acid (3.8 mg, 0.023 mmol, 1.5 equiv), Cs_2CO_3 (9.8 mg, 0.030 mmol, 2.0 equiv) and $PdCl_2$(dppf) (1.1 mg, 1.5 µmol, 0.1 equiv). Anisole (0.5 mL, purged with CO) was added and the vial was flushed with CO gas (toxic) for 30 sec. After heating to 80 °C for 2hrs, the solution was poured into EtOAc and the organic layer was washed with 1 N NaOH and sat aq NaCl, dried with Na_2SO_4 and concentrated under reduced pressure. Purification by silica gel chromatography (150:1→100:1 CH_2Cl_2/MeOH) yielded **22** (4.2mg, 8.2 µmol, 55%).

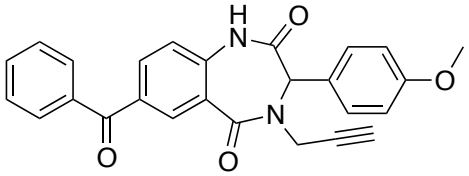


7-(benzo[*d*][1,3]dioxole-5-carbonyl)-4-(prop-2-yn-1-yl)-3-(4-(trifluoromethyl)phenyl)-3,4-dihydro-1*H*-benzo[*e*][1,4]diazepine-2,5-dione (22). Prepared as described above. **^{1}H-NMR** (400 MHz, $CDCl_3$): δ 8.33 (br s, 1H), 7.99 (d, *J* = 2.0 Hz, 1H), 7.68 (dd, *J* = 8.3, 2.0 Hz, 1H), 7.48 (d, *J* = 8.3 Hz, 2H), 7.40-7.36 (m, 2H), 7.04 (m, 1H), 6.86 (d, *J* = 8.2 Hz, 1H), 6.83 (d, *J* = 8.2 Hz, 1H), 6.10-6.09 (m, 2H), 5.71 (s, 1H), 5.13 (dd, *J* = 17.6, 2.5 Hz, 1H), 4.41 (dd, *J* = 17.6, 2.5 Hz, 1H), 2.40 (t, *J* = 2.5 Hz, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 193.65,

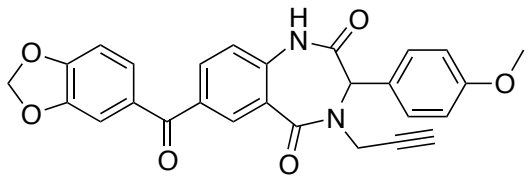
170.54, 166.55, 152.75, 148.91, 137.32, 135.71, 134.58, 134.19, 132.13, 132.00, 129.13, 127.34, 126.78, 126.56, 125.25, 120.92, 110.50, 109.37, 108.72, 102.88, 77.81, 75.19, 65.85, 40.06. **HRMS** *m/z*: cacld for $C_{27}H_{17}F_3N_2O_5$ $[M + H]^+$ 507.1162, found: 507.1159.



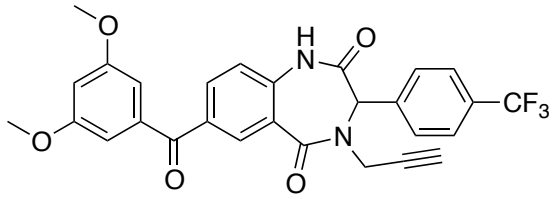
7-(2,3-dihydrobenzo[*b*][1,4]dioxine-6-carbonyl)-4-(prop-2-yn-1-yl)-3-(4-(trifluoromethyl)phenyl)-3,4-dihydro-1*H*-benzo[*e*][1,4]diazepine-2,5-dione (23). Prepared using general procedure B from **S12a** (7.4 mg, 15 µmol, 1 equiv) using 1,4-benzodioxane-6-phenyl boronic acid (4.1 mg, 23 µmol, 1.5 equiv), Cs_2CO_3 (9.8 mg, 30 µmol, 2.0 equiv) and $PdCl_2$(dppf) (1.1 mg, 1.5 µmol, 0.1 equiv) and Anisole (0.5 mL) that yielded **23** (3.6 mg, 6.9 µmol, 46%). **^{1}H-NMR** (600 MHz, MeOD-d_4): δ 7.79 (d, *J* = 1.8 Hz, 1H), 7.62 (dd, *J* = 8.4, 1.2 Hz, 1H), 7.55 (d, *J* = 8.3 Hz, 2H), 7.40 (d, *J* = 8.0 Hz, 2H), 7.01-6.98 (m, 3H), 6.91 (dd, *J* = 8.4, 1.1 Hz, 1H), 5.76 (s, 1H), 4.92 (dd, *J* = 17.5, 2.4 Hz, 1H), 4.64-4.59 (m, 1H), 4.34-4.29 (m, 4H), 2.87 (t, *J* = 2.2 Hz, 1H). **^{13}C-NMR** (151 MHz, MeOD-d_4): δ 194.35, 170.86, 167.18, 149.33, 144.27, 139.10, 134.61, 133.78, 133.20, 130.78, 130.60, 126.73, 126.39, 126.20, 126.18, 124.49, 123.75, 121.16, 119.82, 117.71, 77.86, 74.73, 65.55, 65.23, 64.94, 39.56. **HRMS** *m/z*: cacld for $C_{28}H_{19}F_3N_2O_5$ $[M + H]^+$ 521.1319, found: 521.1322.



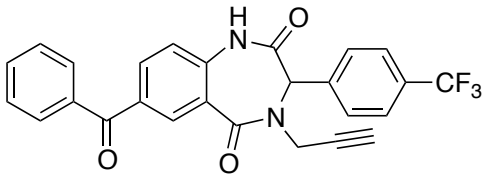
7-benzoyl-3-(4-methoxyphenyl)-4-(prop-2-yn-1-yl)-3,4-dihydro-1*H*-benzo[*e*][1,4]diazepine-2,5-dione (24). Prepared using general procedure B from **S12b** (10 mg, 27 µmol, 1 equiv) using phenyl boronic acid (4.9 mg, 41 µmol, 1.5 equiv), Cs_2CO_3 (18 mg, 54 µmol, 2.0 equiv) and $PdCl_2$(dppf) (2.0 mg, 2.7 µmol, 0.1 equiv) and anisole (0.5 mL) that yielded **24** (5.0 mg, 12 µmol, 44%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.25 (m, 2H), 8.06 (s, 1H), 7.77-7.76 (m, 1H), 7.60-7.45 (m, 3H), 7.10 (m, 2H), 6.86 (d, *J* = 8.3 Hz, 1H), 6.70 (d, *J* = 8.7 Hz, 2H), 5.62 (s, 1H), 5.00-4.97 (m, 1H), 4.45-4.42 (m, 1H), 3.67 (s, 4H), 2.36 (s, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 195.45, 171.57, 166.76, 161.76, 144.55, 141.87, 136.51, 134.84, 134.43, 134.22, 133.61, 132.11, 131.82, 130.75, 129.29, 128.84, 120.77, 114.82, 77.0(obsc), 76.48, 75.64, 74.70, 39.52. **HRMS** *m/z*: cacld for $C_{26}H_{20}N_2O_4$ $[M + H]^+$ 425.1496, found: 425.1500.



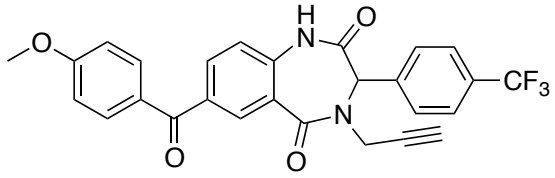
7-(benzo[*d*][1,3]dioxole-5-carbonyl)-3-(4-methoxyphenyl)-4-(prop-2-yn-1-yl)-3,4-dihydro-1*H*-benzo[*e*][1,4]diazepine-2,5-dione (25). Prepared using general procedure B from **S12b** (10 mg, 27 µmol, 1 equiv) using 2,3-methylenedioxyphenyl boronic acid (6.8 mg, 41 µmol, 1.5 equiv), Cs_2CO_3 (18 mg, 54 µmol, 2.0 equiv) and $PdCl_2$(dppf) (2.0 mg, 2.7 µmol, 0.1 equiv) and Anisole (0.5 mL) that yielded **25** (4.6 mg, 10 µmol, 37%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.12 (s, 1H), 7.99 (s, 1H), 7.70 (d, *J* = 8.2 Hz, 1H), 7.10 (m, 4H), 6.86-6.85 (m, 2H), 6.71 (d, *J* = 8.1 Hz, 2H), 6.08 (s, 2H), 5.63 (s, 1H), 5.02-4.99 (m, 1H), 4.46-4.43 (m, 1H), 3.69 (s, 3H), 2.37 (s, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 193.79, 171.44, 166.83, 160.19, 152.59, 151.26, 148.77, 137.71, 135.19, 134.34, 134.24, 132.32, 127.42, 127.17, 127.04, 120.73, 114.87, 110.69, 108.79, 102.80, 77.88, 74.69, 56.06, 31.82. **HRMS** *m/z*: cacld for $C_{27}H_{20}N_2O_6$ $[M + H]^+$ 469.1394, found: 469.1395.



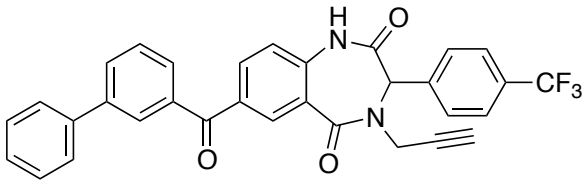
7-(3,5-dimethoxybenzoyl)-4-(prop-2-yn-1-yl)-3-(4-(trifluoromethyl)phenyl)-3,4-dihydro-1*H*-benzo[*e*][1,4]diazepine-2,5-dione (26). Prepared using general procedure B from **S12a** (7.4 mg, 15 µmol, 1 equiv) using 3,5-dimethoxyphenyl boronic acid (4.2 mg, 23 µmol, 1.5 equiv), Cs_2CO_3 (9.8 mg, 30 µmol, 2.0 equiv) and $PdCl_2$(dppf) (1.1 mg, 1.5 µmol, 0.1 equiv) and Anisole (0.5 mL) that yielded **26** (4.1 mg, 7.9 µmol, 52%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.40 (s, 1H), 8.08 (s, 1H), 7.76 (d, *J* = 7.6 Hz, 1H), 7.50 (d, *J* = 8.2 Hz, 2H), 7.36 (d, *J* = 7.9 Hz, 2H), 6.87 (d, *J* = 8.2 Hz, 1H), 6.66 (d, *J* = 1.8 Hz, 1H), 6.63 (s, 2H), 5.71 (s, 1H), 5.11 (dd, *J* = 17.7, 1.6 Hz, 1H), 4.42 (d, *J* = 17.7 Hz, 1H), 3.82 (s, 6H), 2.39 (s, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 194.89, 170.52, 166.53, 161.53, 139.44, 137.80, 137.61, 135.09, 134.84, 134.78, 131.41, 126.75, 126.74, 126.24, 123.46, 120.91, 111.65, 108.75, 105.66, 104.31, 77.78, 75.19, 56.46, 56.24, 40.08. **ESI-MS** *m/z* (neg): cacld for $C_{28}H_{21}F_3N_2O_5$ $[M - H]^-$ 521.1, found: 521.1.



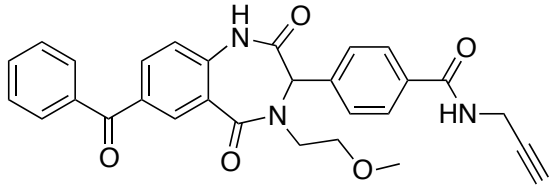
7-benzoyl-4-(prop-2-yn-1-yl)-3-(4-(trifluoromethyl)phenyl)-3,4-dihydro-1*H*-benzo[*e*][1,4]diazepine-2,5-dione (27). Prepared using general procedure B from **S12a** (7.4 mg, 15 µmol, 1 equiv) using phenyl boronic acid (2.8 mg, 23 µmol, 1.5 equiv), Cs_2CO_3 (9.8 mg, 30 µmol, 2.0 equiv) and $PdCl_2$(dppf) (1.1 mg, 1.5 µmol, 0.1 equiv) and Anisole (0.5 mL) that yielded **27** (3.3 mg, 7.1 µmol, 48%). **^{1}H-NMR** (600 MHz, $CDCl_3$): δ 8.35 (s, 1H), 8.04 (s, 1H), 7.75 (d, *J* = 8.2 Hz, 1H), 7.59 (t, *J* = 7.2 Hz, 1H), 7.50-7.41 (m, 6H), 7.36 (d, *J* = 8.0 Hz, 2H), 6.88 (d, *J* = 8.3 Hz, 1H), 5.72 (s, 1H), 5.12 (dd, *J* = 17.6, 1.7 Hz, 1H), 4.41 (d, *J* = 17.7 Hz, 1H), 2.40 (s, 1H). **^{13}C-NMR** (151 MHz, $CDCl_3$): δ 195.33, 170.50, 166.23, 137.80, 136.51, 135.28, 134.75, 134.65, 134.33, 133.75, 130.75, 130.62, 129.32, 128.93, 126.88, 126.53, 126.38, 120.92, 77.50, 75.21, 69.29, 42.7. **HRMS** *m/z*: cacld for $C_{26}H_{17}F_3N_2O_3$ $[M + H]^+$ 463.1264, found: 463.1270.



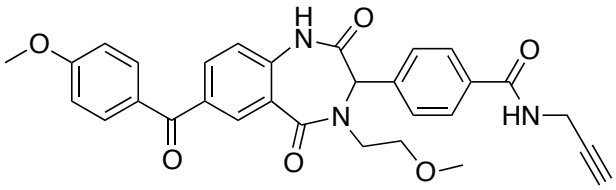
7-(4-methoxybenzoyl)-4-(prop-2-yn-1-yl)-3-(4-(trifluoromethyl)phenyl)-3,4-dihydro-1*H*-benzo[*e*][1,4]diazepine-2,5-dione (28). Prepared using general procedure B from **S12a** (7.4 mg, 15 µmol, 1 equiv) using 4-methoxyphenyl boronic acid (3.5 mg, 23 µmol, 1.5 equiv), Cs_2CO_3 (9.8 mg, 30 µmol, 2.0 equiv) and $PdCl_2$(dppf) (1.1 mg, 1.5 µmol, 0.1 equiv) and Anisole (0.5 mL) that yielded **28** (1.5 mg, 3 µmol, 20%). **^{1}H-NMR** (600 MHz, CDCl3): δ 8.09 (s, 1H), 8.01 (s, 1H), 7.70 (d, *J* = 8.2 Hz, 1H), 7.51-7.47 (m, 4H), 7.37 (dd, *J* = 7.7, 0.3 Hz, 2H), 6.93-6.92 (m, 2H), 6.85 (d, *J* = 8.2 Hz, 1H), 5.71 (s, 1H), 5.12 (d, *J* = 17.6 Hz, 1H), 4.45-4.39 (m, 1H), 3.90 (d, *J* = 1.8 Hz, 3H), 2.39 (d, *J* = 2.0 Hz, 1H).**^{13}C-NMR** (151 MHz, $CDCl_3$): δ 193.30, 169.62, 165.80, 163.66, 136.45, 135.24, 133.88, 133.52, 133.49, 132.41, 132.37, 129.70, 126.08, 125.79, 125.77, 125.67, 120.10, 113.87, 80.8, 74.44, 65.11, 55.69, 39.33. **HRMS** *m/z*: cacld for $C_{27}H_{19}F_3N_2O_4$ $[M + H]^+$ 493.1370, found: 493.1369.



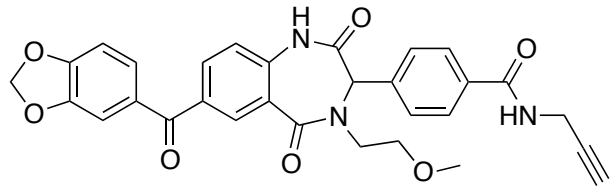
7-([1,1'-biphenyl]-3-carbonyl)-3-(4-methoxyphenyl)-4-(prop-2-yn-1-yl)-3,4-dihydro-1*H*-benzo[*e*][1,4]diazepine-2,5-dione (29). Prepared using general procedure B from **S12a** (7.4 mg, 15 µmol, 1 equiv) using biphenyl-3-boronic acid (3.5 mg, 23 µmol, 1.5 equiv), Cs_2CO_3 (9.8 mg, 30 µmol, 2.0 equiv) and $PdCl_2$(dppf) (1.1 mg, 1.5 µmol, 0.1 equiv) and Anisole (0.5 mL) that yielded **29** (4 mg, 8.1 µmol, 54%). **^{1}H-NMR** (600 MHz, MeOD-d_4): δ 7.99 (d, *J* = 1.9 Hz, 1H), 7.88 (dd, *J* = 7.8, 1.2 Hz, 1H), 7.79 (s, 1H), 7.76 (dd, *J* = 8.4, 2.0 Hz, 1H), 7.63-7.62 (m, 3H), 7.58 (dd, *J* = 9.6, 5.8 Hz, 1H), 7.49-7.47 (m, 4H), 7.40 (dd, *J* = 8.8, 7.8 Hz, 4H), 7.03 (d, *J* = 8.4 Hz, 1H), 5.77 (s, 1H), 4.94-4.90 (m, 1H), 4.64-4.59 (m, 1H), 2.88-2.86 (m, 1H). **^{13}C-NMR** (151 MHz, MeOD-d_4): δ 195.31, 170.71, 167.06, 142.54, 140.85, 139.68, 138.31, 134.22, 134.02, 133.84, 132.02, 129.59, 129.54, 129.35, 129.11, 128.56, 128.48, 128.02, 127.66, 127.55, 126.95, 126.81, 126.10, 121.26, 77.87, 74.74, 66.40, 39.65. **ESI-MS** *m/z* (pos): cacld for $C_{32}H_{21}F_3N_2O_3$ $[M + H]^+$ 496.2, found: 496.2; (neg) $[M - H]^-$ 494.2, found: 494.1.



7-benzoyl-4-(2-methoxyethyl,)-3-(4-(prop-2-yn-1-ylcarbamoyl)phenyl)-3,4-dihydro-1*H*-benzo[*e*][1,4]diazepine-2,5-dione (30). Prepared using general procedure B from **S12c** (7.8 mg, 16 µmol, 1 equiv) using phenyl boronic acid (2.9 mg, 24 µmol, 1.5 equiv), Cs_2CO_3 (10.4 mg, 32 µmol, 2.0 equiv) and $PdCl_2$(dppf) (1.2 mg, 1.6 µmol, 0.1 equiv) and anisole (0.5 mL) that yielded **30** (4.1 mg, 8.2 µmol, 52%). **^{1}H-NMR** (600 MHz, MeOD-d_4): δ 7.82 (s, 1H), 7.68 (d, *J* = 8.4 Hz, 2H), 7.64-7.62 (m, 2H), 7.50 (m, 2H), 7.46 (m, 2H), 7.25-7.24 (m, 2H), 6.99-6.98 (m, 1H), 5.65 (s, 1H), 4.19-4.15 (m, 2H), 4.07-4.02 (m, 4H), 3.75 (d, *J* = 48.4 Hz, 4H), 3.37-3.32 (m, 3H), 2.54 (s, 1H). **^{13}C-NMR** (151 MHz, MeOD-d_4): δ 195.90, 171.65, 168.06, 167.98, 141.74, 138.83, 137.88, 134.01, 133.66, 133.60, 133.41, 130.22, 129.10, 128.15, 127.49, 125.73, 122.64, 120.92, 80.02, 71.53, 70.86, 67.99, 58.42, 51.04, 29.30. **ESI-MS** *m/z* (pos): cacld for $C_{19}H_{13}NO_3$ $[M + H]^+$ 496.2, found: 496.2; (neg) $[M - H]^-$ 494.2, found: 494.1.

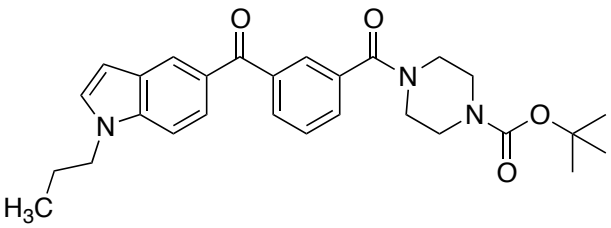


7-(4-methoxybenzoyl)-4-(2-methoxyethyl)-3-(4-(prop-2-yn-1-ylcarbamoyl)phenyl)-3,4-dihydro-1*H*-benzo[e][1,4]diazepine-2,5-dione (31). Prepared using general procedure B from **S12c** (7.8 mg, 16 µmol, 1 equiv) using 4-methoxyphenyl boronic acid (3.6 mg, 24 µmol, 1.5 equiv), Cs_2CO_3 (10.4 mg, 32 µmol, 2.0 equiv) and $PdCl_2$(dppf) (1.2 mg, 1.6 µmol, 0.1 equiv) and anisole (0.5 mL) that yielded **31** (3.7 mg, 7.1 µmol, 44%). **^{1}H-NMR** (600 MHz, MeOD-d_4): δ 7.76 (s, 1H), 7.69 (d, *J* = 8.4 Hz, 2H), 7.57 (dd, *J* = 8.3, 1.8 Hz, 1H), 7.44 (d, *J* = 8.7 Hz, 2H), 7.25 (d, *J* = 7.8 Hz, 2H), 7.02-7.01 (m, 2H), 6.97 (d, *J* = 8.3 Hz, 1H), 5.65 (s, 1H), 4.19-4.15 (m, 1H), 4.09-4.02 (m, 3H), 3.89 (d, *J* = 12.9 Hz, 3H), 3.80 (m, 1H), 3.73-3.70 (m, 1H), 3.35 (s, 3H), 2.52 (s, 1H). **^{13}C-NMR** (151 MHz, MeOD-d_4): δ 194.94, 171.73, 168.14, 167.94, 164.77, 139.01, 138.92, 134.75, 134.02, 133.41, 133.03, 132.85, 130.15, 128.14, 127.43, 125.75, 120.88, 114.39, 80.03, 71.54, 70.87, 68.00, 58.43, 55.64, 51.01, 29.31. **ESI-MS** *m/z* (pos): cacld for $C_{30}H_{27}N_3O_6$ $[M + H]^+$ 526.2, found: 526.2; (neg) $[M - H]^-$ 524.2, found: 524.2.



7-(benzo[*d*][1,3]dioxole-5-carbonyl)-4-(2-methoxyethyl)-3-(4-(prop-2-yn-1-ylcarbamoyl)phenyl)-3,4-dihydro-1*H*-benzo[e][1,4]diazepine-2,5-dione (32). Prepared using general procedure B from **S12c** (7.8 mg, 16 µmol, 1 equiv) using 3,4-methylenedioxyphenyl boronic acid (4.0 mg, 24 µmol, 1.5 equiv), Cs_2CO_3 (10.4 mg, 32 µmol, 2.0 equiv) and $PdCl_2$(dppf) (1.2 mg, 1.6 µmol, 0.1 equiv) and anisole (0.5 mL) that yielded **32** (4.0 mg, 7.4 µmol, 46%). **^{1}H-NMR** (600 MHz, MeOD-d_4): δ 7.75 (s, 1H), 7.68 (d, *J* = 8.4 Hz, 2H), 7.56 (d, *J* = 8.4 Hz, 1H), 7.24 (d, *J* = 7.9 Hz, 2H), 7.03 (s, 1H), 6.97 (d, *J* = 8.3 Hz, 1H), 6.91 (m, 2H), 6.08 (s, 2H), 5.65 (s, 1H), 4.16 (d, *J* = 3.5 Hz, 1H), 4.07-4.03 (m, 3H), 3.80-3.79 (m, 1H), 3.71 (t, *J* = 5.2 Hz, 1H), 3.35 (s, 3H), 2.53 (s, 1H). **^{13}C-NMR** (151 MHz, MeOD-d_4): δ 194.38, 171.71, 168.13, 167.95, 153.02, 149.10, 139.69, 138.90, 134.61, 134.08, 133.42, 132.99, 131.99, 128.17, 127.46, 127.41, 125.73, 120.88, 109.52, 108.33, 103.09, 80.02, 71.56, 70.87, 68.00, 58.43, 51.00, 29.31. **ESI-MS** *m/z* (pos): cacld for $C_{30}H_{25}N_3O_7$ $[M + H]^+$ 540.1, found: 540.1; (neg) $[M - H]^-$ 538.1, found: 538.1.

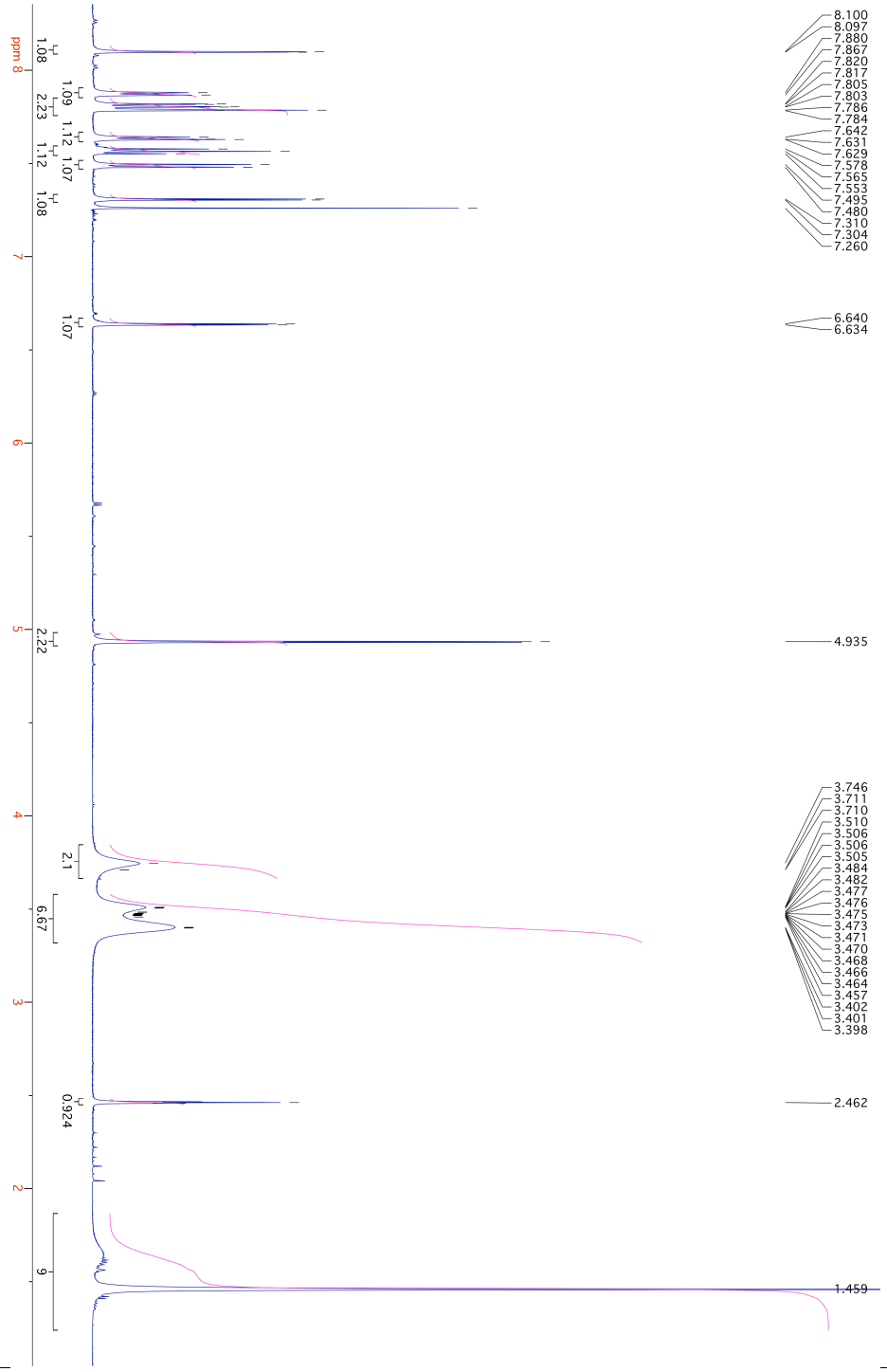
Synthesis of nonclickable analog 37.



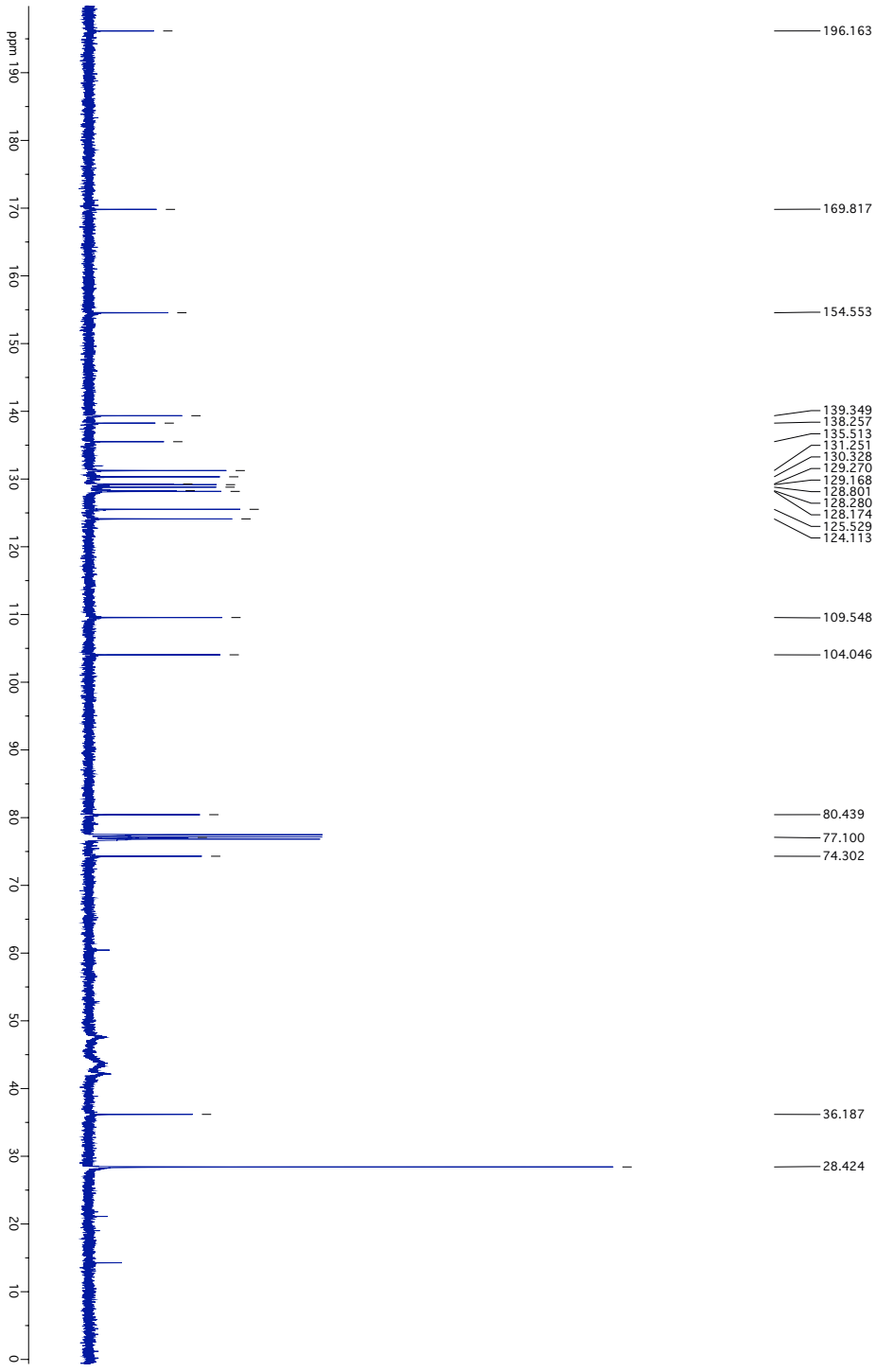
***tert*-butyl 4-(3-(1-propyl-1*H*-indole-5-carbonyl)benzoyl)piperazine-1-carboxylate (37).** A solution of **1** (8 mg, 17 µmol) in anhydrous EtOH was treated with 8mg 5% wt. % loading palladium on barium sulfate (8 mg). The solution was stirred under 1 atm H_2 for 2 hrs. The solution was filtered through celite, concentrated, and purified by silica chromatography (1:1 hexanes/EtOAc) and yielded **37** (7.0 mg, 14.7 µmol, 86%). **^{1}H-NMR** (600 MHz, CDCl3): δ 8.09 (s, 1H), 7.88 (d, *J* = 7.1 Hz, 1H), 7.79 (m, 2H), 7.63 (d, *J* = 6.8 Hz, 1H), 7.56 (t, *J* = 7.5 Hz, 1H), 7.41 (d, *J* = 8.6 Hz, 1H), 7.36 (m, 1H), 7.19 (m, 1H), 6.60 (s, 1H), 4.14 (t, *J* = 6.8 Hz, 2H), 3.76-3.74 (m, 2H), 3.52-3.39 (m, 6H), 1.90 (q, *J* = 7.1 Hz, 2H), 1.46 (s, 9H), 0.95 (t, *J* = 7.3 Hz, 3H). **^{13}C-NMR** (151 MHz, CDCl3): δ 196.36, 169.98, 154.65, 139.64, 138.68, 131.33, 130.28, 129.82, 128.86, 128.64, 128.47, 128.20, 127.94, 125.79, 123.69, 109.64, 103.22, 80.54, 77.16, 53.57, 48.47, 28.50, 23.77, 11.63. **HRMS** *m/z*: cacld for $C_{28}H_{33}N_3O_4$ $[M + H]^+$ 476.2544, found: 476.2542.

Spectra for Compound 1

Compound 1: ^{1}H spectra



Compound 1: ^{13}C Spectra



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