Development of Novel Small Molecule Inhibitors
of Neurotropic Alphavirus Replication

by

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List of Abbreviations

ADME   Absorption, distribution, metabolism, excretion

BBB   Blood-brain barrier

BSL   Bio-safety level

CC_{50} Cytotoxic concentration, half maximal

CEV   California encephalitis virus

CNMR   Carbon nuclear magnetic resonance spectroscopy

CNS   Central nervous system

CPE   Cytopathic effect

CYP   Cytochrome P450

DMSO   Dimethylsulfoxide

EEV   Equine encephalitis virus

EEEV   Eastern equine encephalitis virus

EMCV   Encephalomyocarditis virus

fLUC   Firefly luciferase

FMV	Fort Morgan virus
HNMR	Proton nuclear magnetic resonance spectroscopy
HPI	Hours after infection
HPLC	High-performance liquid chromatography
HTS	High-throughput screen
IC_{50}	Inhibitory concentration, half maximal
IR	Infrared spectroscopy
LC	Liquid chromatography
MDCK	Madin-Darby canine kidney cells
MDR1	Multidrug resistance protein 1
MLM	Mouse liver microsome
MOA	Mechanism of action
MOI	Multiplicity of infection
MS	Mass spectrometry
MS/MS	Tandem mass spectrometry
MTT	3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyl-2*H*-tetrazolium bromide

MW	Molecular weight
NMR	Nuclear magnetic resonance spectroscopy
PAMPA	Parallel artificial membrane permeability assay
PFU	Plaque-forming unit
Pgp	P-glycoprotein
Rho123	Rhodamine 123
SAR	Structure-activity relationship
SEAP	Secreted embryonic alkaline phosphatase
SEM	Standard error of the mean
SD	Standard deviation
SINV	Sindbis neuroadapted virus
TPSA	Topological polar surface area
VEEV	Venezuelan equine encephalitis virus
WEEV	Western equine encephalitis virus

Chapter I. **Introduction**

Arboviruses: An Emerging Public Health Threat

New and emerging viruses pose an immense public health risk to the world population,[1, 2] due in large part to a lack of effective vaccines and treatments, although inadequate public health responses may also play a significant role.[3] Mosquito-borne viruses, collectively called arboviruses (*ar*thropod-*bo*rne *viruses*), constitute a large portion of this risk because of the ubiquity of mosquitoes and the potential for mosquito-human transmission.[4-6] Increasing urbanization and mosquito range expansion further complicates the threat of potential arboviral pandemic.[4-6] Among emerging New World arboviruses, the equine encephalitis viruses (EEVs) (Western, Eastern, and Venezuelan) are of particular interest, and are listed as category B agents of biodefense concern due in part to their potential use in bioterrorism.[7-9]

EEVs are neurotropic arboviruses that infect the central nervous system (CNS) of vertebrates, causing debilitating inflammation and damage to the brain.[10-12] They are members of the genus *Alphavirus* of the Group IV family *Togaviridae*,[13] of which more than ten members cause significant disease in humans, and all are arboviruses.[1] They are also RNA viruses, and their emerging status may be attributed to this characteristic,[2] largely due to poor genome replication fidelity and high mutation rates common to RNA viruses.[14] In nature, EEV existence is maintained by a cycle between passerine birds and the mosquitoes that prey exclusively on them (**Figure 1**).[15, 16] Infection in humans and

animals occurs when other mosquitoes, the so-called bridging vectors, pass the virus along from birds.[17] The natural infection rate in humans, while underreported, is nonetheless low: since 1964, only 639 cases of Western equine encephalitis virus (WEEV)[18] and 220 cases of Eastern equine encephalitis virus (EEEV) infection have been confirmed.[19] Still, the clinical manifestations of infection can be severe. For victims of WEEV, the fatality rates may be as low as 3%,[20] but neural sequelae may persist well after infection.[21] Young children exhibit seizures and paralysis,[22-24] for example, and upwards of 60% continue to do so for months or years after the virus has disappeared.[25] Adults, too, may experience persistent disease-associated impairment.[24] Of greater concern is EEEV, which boasts a mortality rate in excess of 30-40%, and even as high as 70% among some strains,[19] and is also likely to cause permanent neurological damage in survivors.[26-28] On the other hand, Venezuelan equine encephalitis virus (VEEV), while least likely to cause debilitating symptoms in humans,[29-31] may have high agricultural costs associated with infection as equine mortality rates have been observed to exceed 80-90% during epizootic pandemics. Furthermore, unlike with WEEV and EEEV infections, horses and humans are not dead-end hosts and may continue to spread the virus.[16]
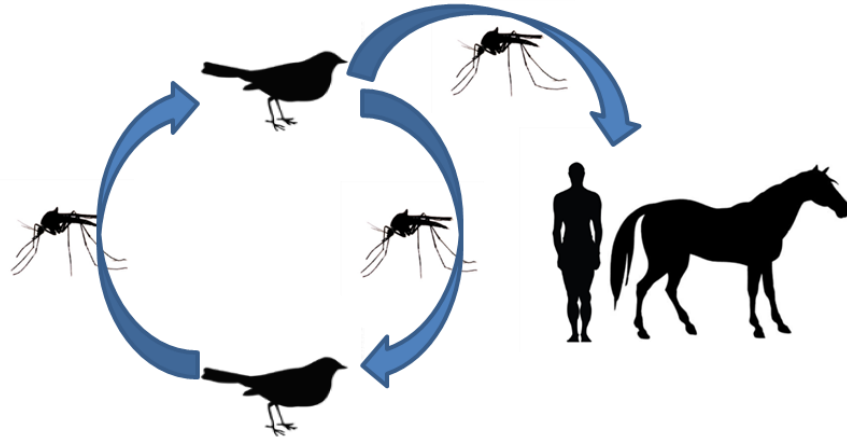
Figure 1. Equine Encephalitis Virus Maintenance Cycle.
Passerine birds serve as reservoirs for many of the New World alphaviruses, but all alphaviruses are transmitted to humans and susceptible animals by arthropods.

After World War II, many of the early developers of biological warfare found that the equine encephalitis viruses and related alphaviruses were highly amenable to weaponization.[9, 16, 32] As stated from a report by the US Army Medical Research Institute for Infectious Diseases, "Few [viruses] possess as many of the required characteristics for strategic or tactical weapon development as the alphaviruses."[16] The reasons are simple: they can be produced in large quantities, they are stable and many are infectious as aerosols, many are fatal (or at the very least, incapacitating), they are relatively simple to engineer (and make more lethal), and defensive vaccine development is hindered by the existence of multiple serotypes. Many nations have long since shut down their offensive bioweapon programs (the United States in 1969), but today there is a growing threat from small groups of bioterrorists.[9, 16, 33]

There currently exist vaccines for WEEV[34] and a few of the other alphaviruses,[35, 36] but they require multiple shots, confer little immunoprotection, and are available only

to researchers.[15, 16, 35] It has also been demonstrated that potential vaccines against one alphavirus may interfere with vaccination against other alphaviruses.[37-39] From a public health perspective, this underlines the need for an effective antiviral, but there are no therapeutics available for treating infection once the virus has colonized the CNS.[15, 40] Therefore, the focus of our research is the development of new, novel compounds for the pharmacological intervention of CNS infection by alphaviruses.

Neurotropic Alphavirus Virus Biology

The reproductive life cycle of neurotropic alphaviruses is among the most simple of viruses, and is largely localized to the cytoplasm of the host cell.[13] As shown in **Figure 2**, the first step (*step 1*) of reproduction involves binding of the virion to specific host receptors displayed on the exterior of the cell membrane, and receptor-mediated endocytosis draws the virion into the cell. Upon acidification of the vesicle, the virion is uncoated and released into the cellular environment (*step 2*), where it may release its simple positive-sense single-stranded RNA ((+)ssRNA) genome (*step 3*.) The genome may be directly translated upon cellular entry by cytoplasmic ribosomes to produce protein and enzyme products that facilitate amplification of the genome (*step 4*), or it may be transcribed to produce many copies of the original genome (*step 5*). The copies may be involved in further translation, or they may be packaged in the capsid of new virion particles (*steps 5*, *9*, and *10*.) Some copies are involved in the translation of membrane-bound proteins in the secretory pathway, where they will be involved in recruitment of capsid proteins and assembly of new virion particles, and will ultimately

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form the viral envelope (*steps 6* through *8*). Finally, budding occurs and the mature virion is released (*step 11*.)[13, 41-45]
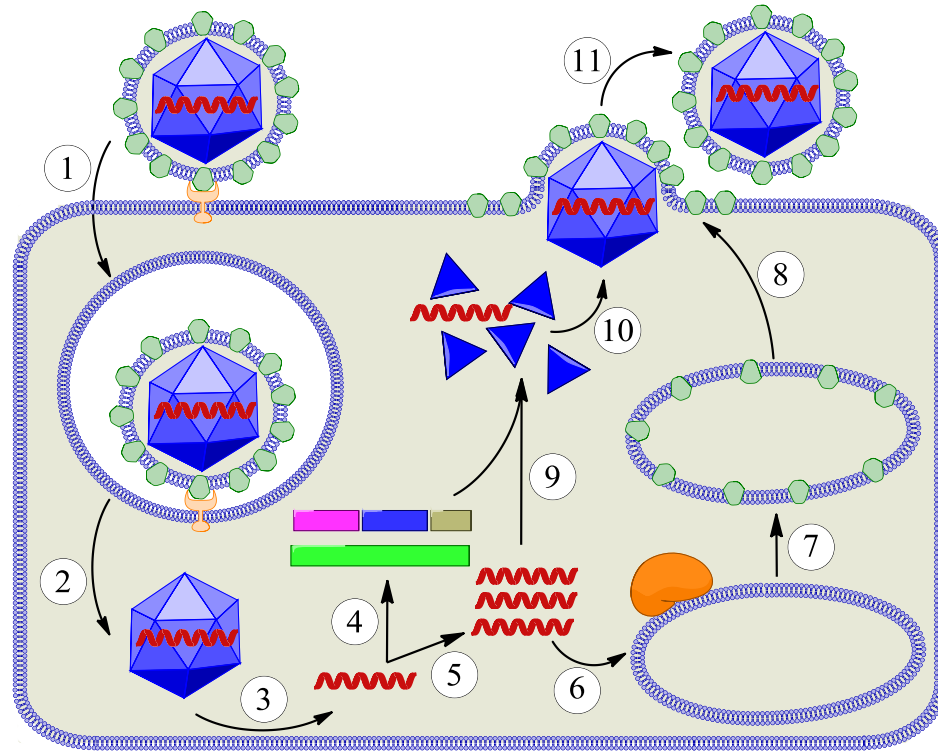
Figure 2. Alphavirus Single-Cell Reproductive Cycle.
Overview of the alphavirus life cycle. Our research focuses on the development of inhibitors of replication (step 5).

Mosquitoes transmit EEVs to hosts upon biting, causing a localized infection that may escalate to observable viremia after several days.[46-48] The mechanism of invasion of the CNS is a contentious subject, with some evidence suggesting that the olfactory nerves are the major route as opposed to the direct crossing of the blood-brain-barrier (BBB).[49-54] Nevertheless, once the virus is established in the CNS, the recruitment of immunological cells is responsible for encephalitis, but the majority of serious brain destruction is caused by the induction of neuronal cell death.[43, 55] Other cell types are

susceptible as well, but form of death is not uniform: glial cells chiefly undergo apoptosis, whereas neuronal cells usually die via necrotic pathways.[55-59]

Project History

High-Throughput Discovery. In search of potential antiviral leads, the David Miller lab executed a high-throughput screen (HTS) of a >50,000-compound library at the University of Michigan Center for Chemical Genomics (CCG).[60] Central to the screen was a cell-based assay that utilized WEEV replicon plasmids encoding the firefly luciferase reporter gene.
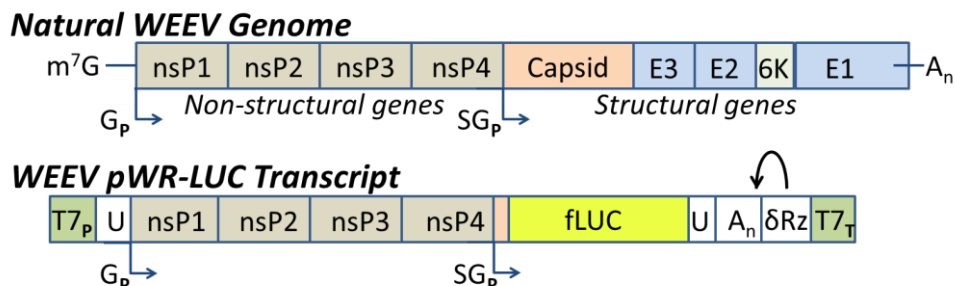
Figure 3. Natural WEEV Genome and WEEV Replicon.
Structural genes were excised and replaced with the reporter gene fLUC, coding for firefly luciferase. In this way, the WEEV replicon is incapable of producing infectious virus and may be used without special precautions.

WEEV Replicon Assay. Inhibitory concentrations (IC_{50} values) were determined using a WEEV replicon assay developed by our collaborators in the lab of Prof. David Miller.[60] BSR-T7/C3 cells were transfected with a complementary plasmid (pWR-LUC) encoding a majority of the WEEV genome (**Figure 3**); however, most of the structural genes are replaced with the reporter gene firefly luciferase.[61] In this way, the replicon cannot produce infectious virus and thus requires less stringent biosafety containment

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measures. BSR cells constitutively express bacteriophage T7 RNA polymerase, for which promoters and terminators exist in the plasmid to maintain high levels of transcription of the WEEV replicon. Therefore, control cells will produce a baseline luminescence while cells treated with active inhibitors will show a marked decrease from baseline, from which an IC_{50} can be calculated.

MTT Reduction Assay. Cytotoxic concentrations (CC_{50} values) were established using a standard 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-2*H*-tetrazol-3-ium bromide (MTT) assay in BSR cells used in the WEEV replicon assay by the lab of Prof. David Miller.[60, 62] In this classical measure of cellular viability, the tetrazolium compound MTT is reduced to a dark purple formazan dye by oxidoreductase enzymes in living cells. Cells stressed or killed by cytotoxic compounds exhibit diminished metabolic activity and, as a result, are less or not at all capable of reducing the MTT to the colorful dye. Therefore, CC_{50} values are calculated from the drop in absorbance relative to a healthy baseline control.

HTS Triage. Because luciferase served as a marker for disruption of genome replication, those compounds with the lowest IC_{50} values in the initial HTS were submitted to secondary and tertiary validation assays. The secondary assay utilized replicons containing the SEAP reporter gene to indicate transcriptional activity and confirm that activity was not an artificial construct of luciferase inhibition,[63] and the tertiary assay screened for compounds with selectivity indices (CC_{50}/IC_{50}) greater than five. One compound, **CCG-32091**, possessed favorable potency and selectivity, and its activity was further verified in cultured human neurons in the presence of the live alphaviruses SINV (Sindbis virus) and FMV (Fort Morgan Virus), which are surrogates

for WEEV that require lower level biosafety containment facilities. Limited structure-activity relationships (SAR) among commercial thienopyrroles related to **CCG-32091** showed promising activities and selectivity ratios, as well as a clearly defined SAR spanning two orders of magnitude potency,[60] so the compound was selected for further optimization.

CCG-32091
IC_{50} = 26 μM
CC_{50} > 100 μM

CCG-102516
IC_{50} = 15 μM
CC_{50} > 100 μM
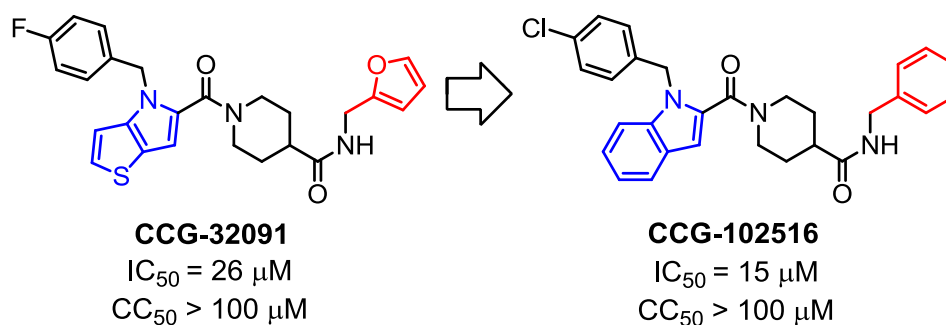
Figure 4. HTS Discovery Hit CCG-32091 and Initial Development.
The thienopyrrole was substituted with bioisosteric indole (blue), and the known metabolism-prone furan was replaced with benzyl functionality (red).

Initial SAR. The initial approach was replacement of the thienopyrrole core of **CCG-32091** with an indole core (**Figure 4**).[64] This was undertaken for several reasons: indoles are generally more metabolically stable than thienopyrroles,[65] are cheaper and synthetically more robust, and phenyl and thiophene are well established bioisosteres.[66, 67] **CCG-102516**, synthesized by Kyle Bolduc, was developed from this rationale. It contains an indole core, a more lipophilic chloro-substituted *N*-benzyl group, and a benzylamide predicted to be more metabolically stable relative to the furylmethylamide of **CCG-32091**.[68, 69] This initial strategy was successful, as **CCG-102516** proved to be nearly twice as potent as **CCG-32091**.

Chapter II. **Reduction of Polar Surface Area**

Rationale

  The Blood-Brain Barrier. Neurotropic alphaviruses replicate to high titer within the CNS,[70] necessitating development of CNS-penetrant antiviral agents. This is especially important because clinical manifestations may appear well after the systemic virus titer has dropped to immeasurable levels, days after initial transmission.[71-73] This places an enhanced emphasis upon retaining physical properties predictive of both good pharmacokinetics and CNS penetration while optimizing both *in vitro* and *in vivo* activity. The most challenging barrier to CNS entry is the blood-brain barrier (BBB).[74-77] The BBB is structurally distinct from other membrane obstacles; due to diminished pinocytosis and the presence of tight-junctions, most drugs must cross the BBB via transcellular passive diffusion alone.[74, 77] However, there are a number of common features among successful CNS-active drugs that enhance passive BBB transit, including low molecular weight (< 400-450), low topological polar surface area (< 60–70 Å^2), and positive logD (~1-3).[74-76, 78, 79]

  MDR1 Recognition Assays. Transporter-mediated efflux is a major hurdle to a CNS drug's biological activity *in vivo*.[80, 81] In its protective role, the blood-brain barrier (BBB) has a number of efflux transporters to facilitate removal of xenobiotics from the CNS, of which MDR1 is generally the most important.[80, 82] Neurotropic alphaviruses replicate to high titer in the CNS, thereby necessitating the development of transporter-

evasive inhibitors to elicit the greatest antiviral effect. It is therefore important to generate analogs with little to no affinity for MDR1 early in the drug development process.

Over the course of the project, two recognition assays were employed to evaluate MDR1 recognition of WEEV inhibitors in the lab of Prof. Richard Keep.[83] Initially, our collaborators utilized a simple experiment that measured the impact of our analogs on the efflux of tritiated (^{3}H) vinblastine, a known MDR1 substrate, from MDCK cells that expressed human MDR1 (MDR1-MDCKII cells).[83, 84] This specific assay was employed in early evaluations of WEEV inhibitor MDR1 interactions, such as those reported for low topological polar surface area (TPSA) analogs in this chapter, but later the Rhodamine 123 uptake assay was employed (see **Chapter III**). In the ^{3}H-vinblastine uptake assay, if the test compound of interest has no interaction with MDR1, the ^{3}H-vinblastine concentration inside the cell will drop as the substrate is pumped out. If the test compound does interact with MDR1, the measured ^{3}H-vinblastine concentration will be higher within the cell. However, due to the nature of the experiment, it cannot be said whether or not analogs are simple blockers of MDR1 or actual substrates. Regardless, the data are useful for identifying compounds with low recognition by MDR1 (and thus potential for efflux) and was used for the prioritization of compounds for further development.

Initially, we were most interested in maximizing potential for achieving CNS penetration by reducing the TPSA of the inhibitors. TPSA is a measure of the surface area of all polar atoms in a molecule, predominately heteroatoms like oxygen and nitrogen.[79, 85, 86] Additionally, polar substituents possess high desolvation penalties and reduce passive permeability due to repulsive interactions with highly nonpolar lipid

membranes.[87, 88] High TPSA is also associated with greater recognition by efflux transporters.[89, 90] The total heteroatom contribution to the TPSA of **CCG-102516** is presented in **Figure 5**; it is evident that heteroatoms with electrons engaged in resonance have little impact, whereas heteroatoms with a complement of free electrons contribute more to the TPSA.

CCG-102516
Total TPSA: 53 Å²
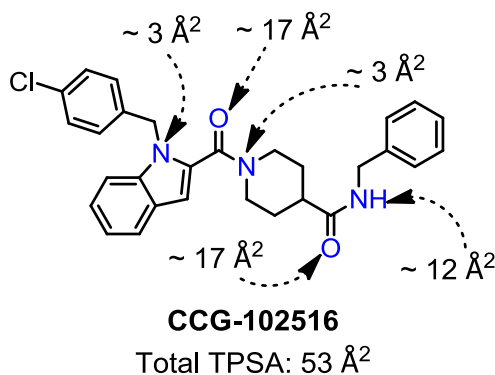
Figure 5. Polar Surface Area Contribution per Heteroatom.
Total TPSA and individual contributions were calculated using ChemDraw's chemical properties calculator.

BBB-PAMPA Assay. Due to the importance of BBB penetration to our compounds, and the role of TPSA in enhancing this property, we estimated the passive membrane permeability for select compounds using BBB-PAMPA (_b_lood-_b_rain _b_arrier _p_arallel _a_rtificial _m_embrane _p_ermeability _a_ssay) from pION, Inc.,[91, 92] and utilized a cosolvent system due to the poor aqueous solubility of early compounds.[93] The traditional PAMPA assay was developed as a useful low-cost tool for assessing potential passive membrane permeability for drug-like compounds in the gut, and was found to correlate well with higher-cost cellular assays.[94, 95] However, the BBB is structurally and compositionally distinct from other membranes, causing PAMPA to often report false positives; for this reason, BBB-PAMPA was developed and was found to possess

superior correlation with BBB passive permeability.[96] It is also an important complement to the MDR1 recognition assay because the MDR1 assay correlates poorly with BBB passive permeability – due to the different lipid composition of the cells – but contains efflux transporters not present in BBB-PAMPA.[97]

While the TPSA of **CCG-102516** is within the range of successful CNS drugs, which have TPSAs significantly lower than other drug classes,[74] it is on the higher end of that range, and we felt it was important to identify low TPSA features that could serve as new templates throughout other aspects of the SAR campaign in order to maintain optimal properties predictive of BBB-penetration. Furthermore, it was important to determine whether TPSA could be reduced with maintenance of anti-viral activity.

SAR

Initial analogs were based on **CCG-203880**, the equipotent *N*-4-fluorobenzyl analog of **CCG-102516** (**Figure 6**) because of its lower molecular weight (see **Chapter III**). A general feature of these analogs was the replacement of the right-hand secondary amide with alternative linkers.

Figure 6. Lead Compounds.
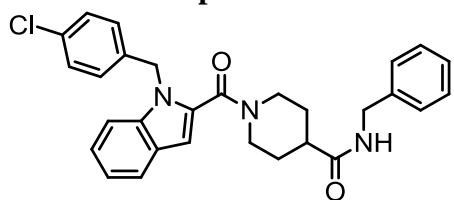
CCG-102516
IC_{50} = 15 μM
CC_{50} > 100 μM
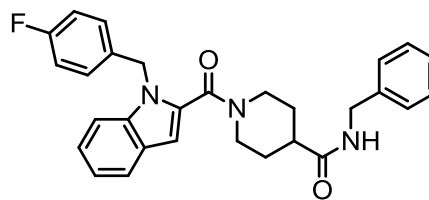
CCG-203880
IC_{50} = 15 μM
CC_{50} > 100 μM

CCG-102516 and its equipotent fluoro analog **CCG-203880**.

Table 1. WEEV Replicon Data for Alicyclic Low TPSA Analogs.
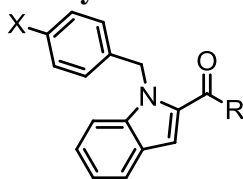
CCG No.	X	R	IC$_{50}$ (μM)[a]	CC$_{50}$ (μM)[b]	MDR1 (% vinblastine uptake)[c]	TPSA[d]
203880†	F		15 ± 2	>100	1,495	53
203941	F		>100	>100	---	27
205421	F		14 ± 1	24 ± 1	---	44
205475	F		>100	>100	---	41
203945	F		23 ± 2	>100	---	44
204055	F		12 ± 1	96	821	27
205476	Cl		10 ± 2	38	---	27

[a]Inhibition of luciferase expression in WEEV replicon assay. [b]Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. [d]TPSA was predicted using ChemDraw's chemical properties calculator. †Synthesized by Janice Sindac.

Alicyclic Low TPSA SAR. As seen in **Table 1**, we were able to demonstrate that TPSA could be reduced substantially in some cases without impacting activity or toxicity (*e.g.* alicyclic analogs **CCG-203945** and **204055**). Aromatic rings are capable of engaging in numerous types of bonding interactions – like ring-stacking or weak CH

hydrogen bonding – not available to alicycles. That the active low TPSA analogs bear non-aromatic heterocycles and carbocycles suggests that the binding in this region may be largely dependent on hydrophobic interactions between the ring and lipophilic residues in the unknown binding site. Also of interest is the difference in activities between **CCG-205421** and **CCG-205475**. Both compounds are hydrogen bond acceptors, but only **CCG-205421** is also a hydrogen bond donor, perhaps forming a key contact with an acceptor in the binding site. **CCG-205421** is also tetrahedral about the sp^3 alcohol carbon, unlike the sp^2 carbonyl carbon in **CCG-205475**. This could indicate that **CCG-205421** has geometry that enhances interactions with unknown binding site contacts.

Aromatic Low TPSA SAR. As seen in **Table 2**, like the alicyclic analogs, we found TPSA could be halved in the aromatic analogs while maintaining favorable potency and toxicity profiles (*e.g.* aromatic analogs **CCG-203942** and **CCG-203945**), placing WEEV inhibitor properties well within the range of successful CNS drugs. Intriguingly, the SAR of ketone **CCG-203943** and alcohol **CCG-204056** matched that of their alicyclic counterparts **CCG-205475** and **CCG-205421**, respectively (**Table 1**). We also noted that benzylamine **CCG-205420** was substantially more potent than its benzylether analog **CCG-203944**, but this result may be an artificial product of its high toxicity. Curiously, benzyl piperazine analogs (*e.g.* **CCG-203942**) were eqipotent with the lead, in contrast to the inactive cyclohexylmethyl piperazine **CCG-203941** (**Table 1**). This challenged the assumption that lipophilicity was the most important factor for binding in this region of molecule.

Table 2. WEEV Replicon Data for Aromatic Low TPSA Analogs.
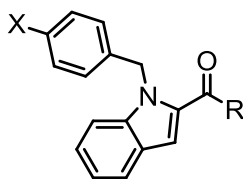
CCG No.	X	R	IC_{50} $(\mu M)^a$	CC_{50} $(\mu M)^b$	MDR1 (% vinblastine uptake)^c	TPSA^d
203880†	F		15 ± 2	>100	1,495	53
203943	F		>100	>100	---	41
204056	F		15 ± 0.4	40	---	44
204020	F		>100	>100	---	27
205420	F		22 ± 3	39	---	36
203944	F		>100	>100	---	33
203942	F		12 ± 1	>100	764	27
206372	Cl		12 ± 1	>100	---	27
211827	Cl		7.2	>100	---	27
211826	Cl		12.4	>100	---	39

^{a}Inhibition of luciferase expression in WEEV replicon assay. ^{b}Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. †Synthesized by Janice Sindac. ^{d}TPSA was predicted using ChemDraw's chemical properties calculator.

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Table 3. MDR1 Recognition Data for Indole and Thienopyrrole leads.
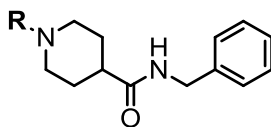
CCG No.	R	MDR1 Recognition (% vinblastine uptake)[a]
102516†		534.0
203880†		1495.6
203881†		433.3
32088*		694.1

[a]MDR1 recognition was assessed by measuring uptake ^{3}H-vinblastine at 30 μM of test anti-viral or vehicle. †Synthesized by Janice Sindac. *Synthesized by Bryan Yestrepsky.

MDR1 Recognition. It was hoped that the best low TPSA candidates from this assay may serve as new templates for further development. After our initial survey of the activity of these analogs was complete, **CCG-204055** and **CCG-203942** were graduated to the MDR1/vinblastine assay because they showed promising inhibition and low cytotoxicity. Both of these compounds were found to possess improved MDR1-efflux results over **CCG-203880** (which may be attributed to their lower TPSA compared to their amide counterparts), and were therefore considered potential new leads. However,

initial trends among other scaffolds suggested that chloro-substituted compounds may be more MDR1-evasive than their fluoro-substituted counterparts (**Table 3**), prompting us to prepare and evaluate chloro-substituted analogs of some of the better fluoro-substituted compounds (*e.g.* **CCG-205476** and **CCG-206372, Table 1** and **Table 2**, respectively). These compounds were found to be equipotent to their fluoro-substituted cousins. Unfortunately, these compounds were never submitted to the MDR1 recognition assay due to the discovery of the significantly more potent 4-pyridylethylamide inhibitor class, described briefly below and in detail in **Chapter III**.

TPSA Analog Length-Activity Dependence. While many of the TPSA analogs feature low polar surface area with retention of activity, it was apparent that there was still room for potency optimization. Janice Sindac, working on other aspects of the WEEV project, had established that the right-hand portion of the template must be of a specific length, where the intervening alkyl chain between the amide and the aromatic is optimally 2-carbons long (**Figure 7**).[83] Interestingly, propyl linkers abolished activity entirely. The low TPSA compounds in **Table 1** and **Table 2** were significantly truncated, so longer analogs were investigated. Interestingly, analogs with ethyl and propyl linkers did not perform significantly better than their shorter counterparts (*e.g.* **CCG-206372** compared to **CCG-211827**, **Table 2**), nor were they worse. Even **CCG-211826**, which bears a 4-pyridylethyl substituent crucial to the activity of later analogs (see **Chapter III**), merely maintained activity. This suggested that either important structural features were not present among the low TPSA analogs or that they were not binding in the same way as the amide leads. Indeed, later work established the importance of the right-hand

secondary amide to activity (see **Chapter IV**) which is lacking in the low TPSA inhibitor class.
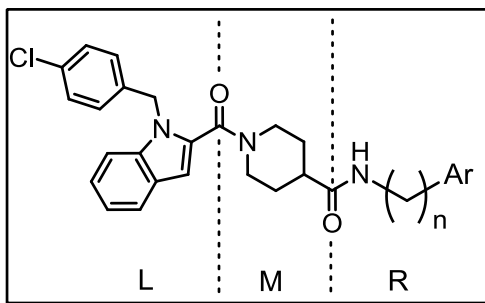
Figure 7. Right-Hand Chain Length-Dependence.
Chain-length dependence of the right-hand region "R" was found to decrease in activity such that n=2>1>>3, where n is an intervening number of methylene units.

Secondary Amine Analog SAR. The low TPSA analogs are highly lipophilic. While this aids in BBB permeation, it decreases water solubility such that aqueous delivery of compound is impeded. Interestingly, analogs containing ionizable functionalities, such as amines, exhibited the greatest activity (*e.g.* piperazine-containing analogs **CCG-203942** and **CCG-204055**), perhaps because of improved aqueous solubility. Alternatively, the analogs may simply possess improved permeability, as very lipophilic compounds may be retained by the membrane, inhibiting their ability to penetrate the cell. In an effort to exploit this trend, a new generation of right-hand amine-bearing analogs was prepared (**Table 4**), but these proved to be quite toxic, especially those containing secondary amines (*e.g.* **CCG-206486** and **CCG-206500**), perhaps via detergent activity (disruption of cellular membranes). On the other hand, tertiary amines (*e.g.* **CCG-206502** and **CCG-206503**) were better tolerated but displayed markedly decreased activity. All of the analogs possessed lower CC_{50} values than those seen in

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their amide counterparts, and there was no significant activity improvement. For these reasons, the amine-containing series was discontinued early. This did not undermine development efforts of the low TPSA WEEV inhibitor class; overall, we demonstrated that activity could be maintained with reduced TPSA and – potentially – increased hydrophilicity.

Table 4. WEEV Replicon Data for Amine Analogs.
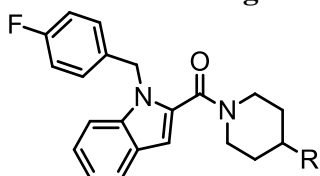
CCG No.	R	IC$_{50}$ (µM)^a	CC$_{50}$ (µM)^b
203880†		14.8 ± 2.1	>100
206486		6.1 ± 0.7	9
206499		>100	>100
206500		6.5 ± 0.3	12
206501		8.9 ± 0.4	16
206502		40.6 ± 0.5	96
206503		25.1 ± 1.9	85

[a]Inhibition of luciferase expression in WEEV replicon assay. [b]Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. †Synthesized by Janice Sindac.

Synthesis

Low TPSA analogs were generally prepared through the alkylation of ethyl 2-indolecarboxylate **1** with either 4-chloro- or fluorobenzyl chloride **2a** or **2b**. The *N*-alkylated ethyl ester intermediate **3a,b** was then saponified to the corresponding carboxylic acid **4a,b** and coupled with the desired amine to give the general structure **5a,b** (**Scheme 1**).

Scheme 1. General Preparation of Low TPSA Analogs.^{a}

X = Cl, 2a
X = F, 2b

1   3a,b   4a,b   5a,b
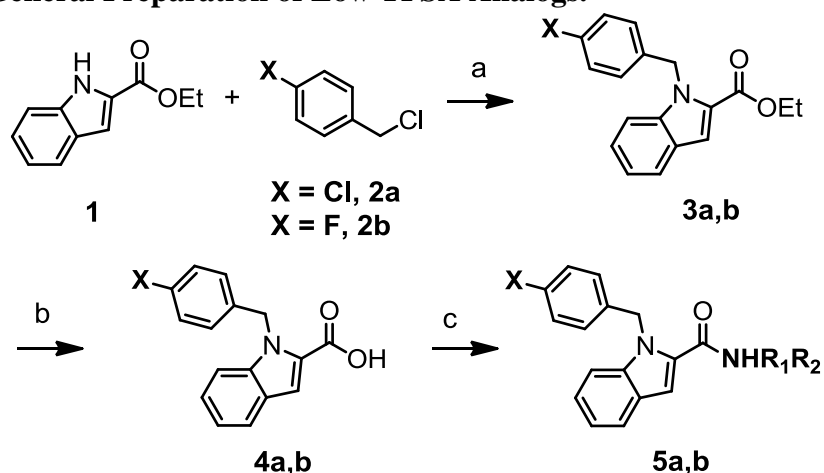
^{a}Reagents and conditions: (a) K$_2$CO$_3$, DMF, 60 °C, 20 h, 70%; (b) 7 M aq. NaOH, EtOH, 50 °C, 3 h, 97%; (c) **HNR$_1$R$_2$**, EDC, HOBt, DIPEA, DMF, rt, ~ 24 h.

Many amines **R$_1$R$_2$NH** had to be prepared due to commercial unavailability. The 1-(piperidin-4-ylmethyl)piperidine portion of the analogs **CCG-204055** and **CCG-205476** was first accessed through the Boc-protection and Swern oxidation of piperidinyl-4-methanol **6**. This provided the aldehyde **8** that underwent reductive amination with piperidine to give the boc-protected amine **9**, and this was subsequently

deprotected to reveal the amine as a trifluoroacetate salt and coupled with the appropriately halogenated indole carboxylic acid to yield the desired analogs (**Scheme 2**).

Scheme 2. Preparation of CCG-204055 and CCG-205476.[a]
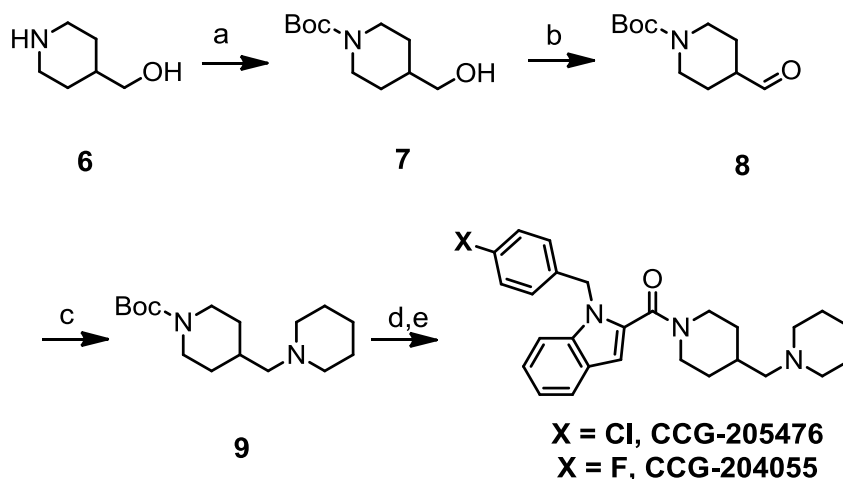
[a]Reagents and conditions: (a) Boc$_2$O, Na$_2$CO$_3$, H$_2$O, THF, reflux, 2 h, 87%; (b) DMSO, (COCl)$_2$, -78 °C, 30 min; *then* TEA, -78 °C→0 °C, 3 h, 89%; (c) piperidine, NaCNBH$_3$, TFE, 3 Å MS, RT, 20 h, 72%; (d) TFA, DCM, RT; (e) **4a** or **4b**, EDC·HCl, HOBT, DIPEA, DMF, RT, 15-24 h, 54-63%.

The alicyclic analogs **CCG-205421** and **CCG-205475** were accessed through a single linear route in which the latter was a direct synthetic derivative of the former (**Scheme 3**). In this fashion, the aldehyde **8** was treated with cyclohexylmagnesium bromide and the resulting Grignard reaction adduct **10** was deprotected to reveal the secondary amine, which was then coupled with the indole carboxylic acid to afford the first analog, hydroxyl-containing **CCG-205421**, which was contaminated with ester from competing *O*-acylation. The desired amide was separated from the ester side-product and confirmed by IR spectroscopy. **CCG-205475** was then prepared by simply oxidizing the alcohol to the ketone under Swern conditions.

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Scheme 3. Preparation of CCG-205421 and CCG-205475.[a]

8 → a → **10** → b → **11** → c →

CCG-205421 → d → CCG-205475
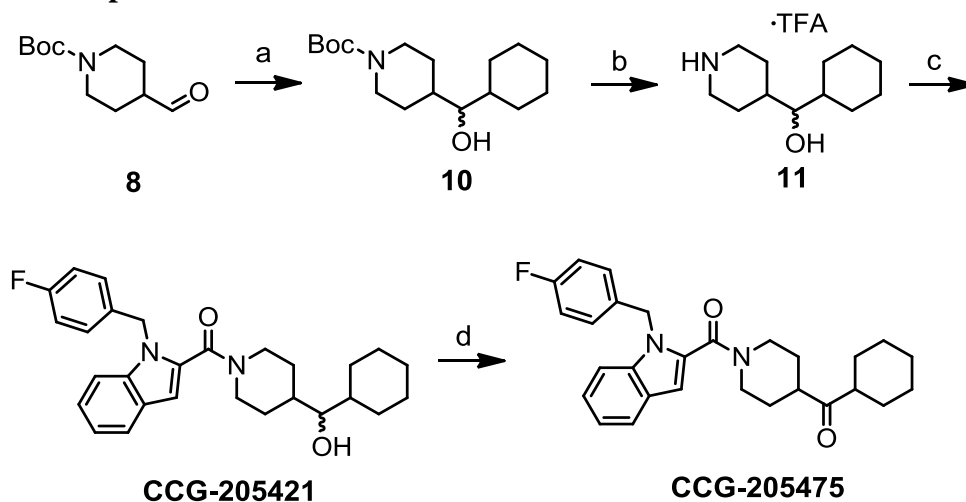
[a]Reagents and conditions: (a) cyclohexyl-MgBr, Et$_2$O, RT, 2 h, 85%; (b) TFA, DCM, RT, 15 h; (c) 1-(4-fluorobenzyl)-1H-indole-2-carboxylic acid, EDC·HCl, HOBT, DIPEA, DMF, rt, 24 h, 60%; (d) (COCl)$_2$, DMSO, DCM, -78 °C, 30 min; *then* TEA, -78 °→RT, 30 min, 58%.

Scheme 4. Preparation of CCG-204020 and CCG-205420.[a]

12 → a → **13** → b → **14** → c →

CCG-204020 → d → CCG-205420
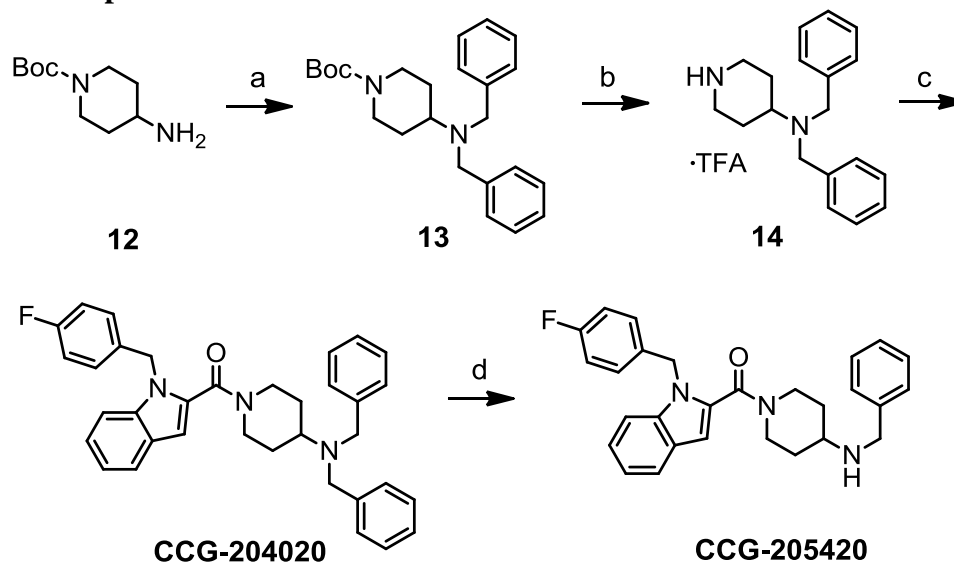
[a]Reagents and conditions: (a) BnBr, Na$_2$CO$_3$, H$_2$O, DCM, reflux, 3 h, 87%; (b) TFA, DCM, RT, 18 h; ; (c) 1-(4-fluorobenzyl)-1H-indole-2-carboxylic acid, EDC·HCl, HOBT, DIPEA, DMF, rt, 30 h, 52%; (d) Pd(OH)$_2$, H$_2$, HCl, EtOH, THF, RT, 1 h, 77%.

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The benzylated amine analogs **CCG-204020** and **CCG-205420** were prepared in a synthetically distinct, albeit similarly linear fashion, to the previous two compounds (**Scheme 4**). The protected 4-aminopiperidine **12** was doubly benzylated with benzyl bromide and deprotected with TFA to afford the amine salt **14**. Coupling with the indole carboxlic acid **4b** gave **CCG-204020**, and time-sensitive selective mono-debenzylation provided the singly substituted benzyl amine **CCG-205420**.

The synthesis of piperazine analog **CCG-211826** required prior preparation of a suitable alkylation partner (**Scheme 5**) by displacement of alcohol **15** under forcing conditions with refluxing hydrobromic acid. 1-Boc-piperazine was then alkylated with the resulting bromo compound **16**, deprotected under acidic conditions, and coupled with the indole carboxylic acid **4a** to provide the desired final analog.

Scheme 5. Preparation of CCG-211826.[a]
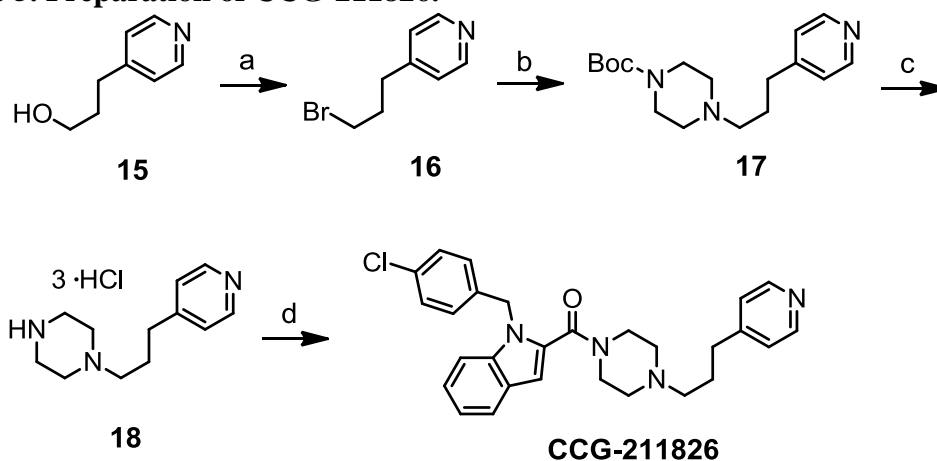
[a]Reagents and conditions: (a) conc. HBr, 130 °C, 2 h, 40%; (b) *t*-butyl piperazine-1-carboxylate, NaH, DMF, RT, 19 h, 66%; (c) HCl, 1,4-dioxane, RT, 15 min, 100%; (d) 1-(4-Chlorobenzyl)-1*H*-indole-2-carboxylic acid, TEA, EDC·HCl, HOBT, DCM, RT, 14 h, 63%.

Secondary amine analogs were generally prepared through the coupling of the indole carboxylic acid **4b** and piperidine-4-methanol **19** (**Scheme 6**). The resulting alcohol adduct was then oxidized under Swern conditions to the aldehyde **20**, which could undergo reductive amination with various amines to provide the desired diversity of structural type **21**.

Scheme 6. General Preparation of Secondary Amine Analogs.
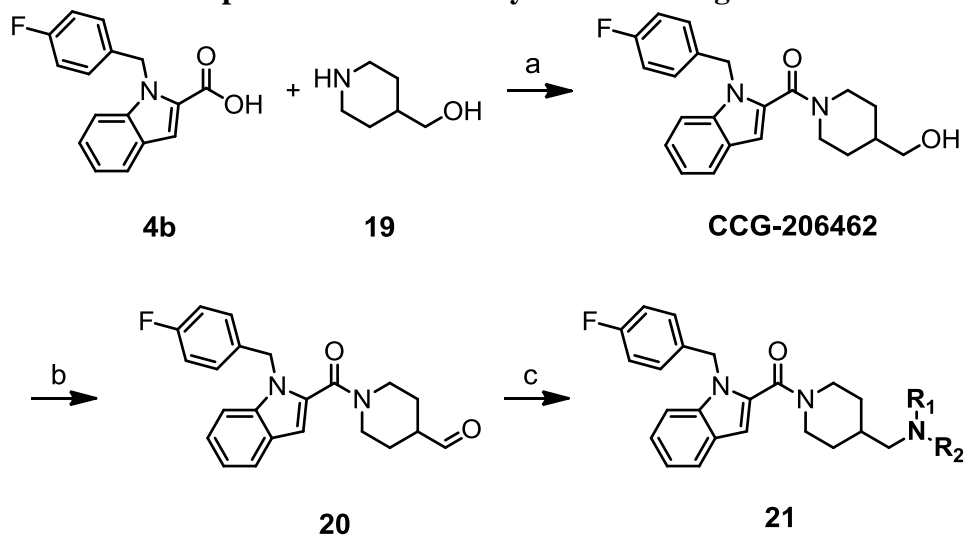
*Reagents and conditions: (a) EDC·HCl, HOBT, DIPEA, DMF, 94%; (b) DMSO, (COCl)$_2$, DCM, -78 °C; *then* TEA, -78 °C→0 °C, 66%; (c) **NHR$_1$R$_2$**, Na(CN)BH$_3$, AcOH, THF, EtOH.

Conclusion

The primary goal of this chapter was reduction of topological polar surface area. To this end, we succeeded in halving the TPSA while maintaining potency, bringing our compounds well within the optimal range of successful CNS drugs. This work also identified useful structural modifications (*e.g.* piperidine-to-piperazine substitutions) and

underlined the need to avoid secondary amine functionality due to cytotoxicity issues.

Notably, reduction of TPSA appeared to enhance MDR1 evasion, an important feature

for CNS-active drugs.

Chapter III. **Reduction of Molecular Weight**

Rationale

Janice Sindac, working on other aspects of the WEEV project, had identified **CCG-205432** as a viable new lead due to its substantially improved potency and solubility over the earlier lead **CCG-102516** (**Figure 8**).[83] However, it was clear that some properties of **CCG-205432** required optimization due to its increased molecular weight (MW) and TPSA, features associated with diminished passive permeability and thus reduced activity. Due to the rather flat SAR associated with the low TPSA analogs (see **Chapter II**), we elected to focus on molecular weight reduction as an alternative course of action for correcting the physicochemical properties of **CCG-205432**.

CCG-102516[†]
IC_{50} = 15 μM
CC_{50} > 100 μM
MW = 486

CCG-205432[†]
IC_{50} = 0.53 μM
CC_{50} = 65 μM
MW = 501
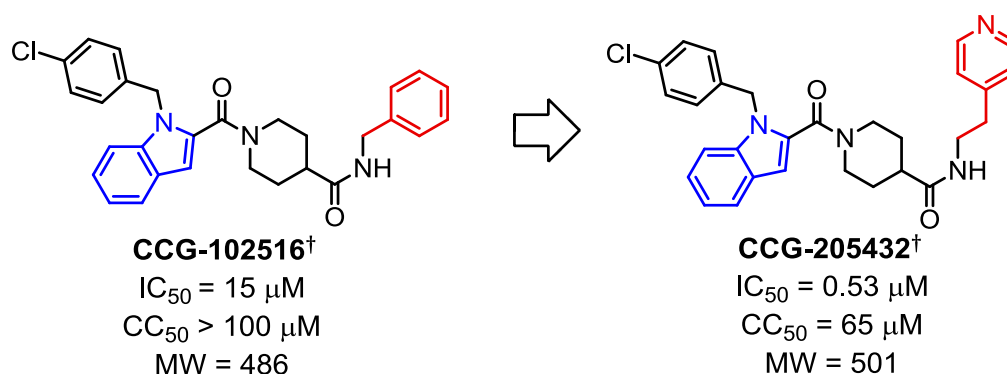
Figure 8. Development of Lead CCG-205432.

Molecular weight (MW) is a property of extreme importance for CNS-acting drugs, which tend to be of low molecular weight (<450).[74, 98] This is crucial, as reduction

of molecular weight promotes passive permeability into membranes non-linearly;[99] even a small change in molecular weight can have a large impact on membrane penetration. Furthermore, reduced molecular weight is associated with lower recognition by efflux transporters like MDR1,[79, 80, 100, 101] an attribute we considered essential for our WEEV inhibitors.

MDR1 Rho123 Uptake Assay. Our collaborators in the lab of Prof. Richard Keep discovered that the ^{3}H-vinblastine uptake assay suffered from poor reproducibility due to complications in acquiring the specific MDR1-MDCKII cell line used in previous work (see **Chapter II**). Fortunately, they found that the Rhodamine 123 (Rho123) uptake assay[102] was a superior substitute to the ^{3}H-vinblastine uptake assay, and this was utilized for the reduced molecular weight analog class. Not only did the Rho123 assay feature improved reproducibility, but Rho123 is a simple dye whose concentration may be measured by absorbance and avoids those complications associated with radioactivity. Both of these assays operate in the same manner; if the test compound of interest has no interaction with MDR1, the ^{3}H-vinblastine or Rho123 concentration inside the cell will drop as the substrate is pumped out. If the test compound does interact with MDR1, the measured ^{3}H-vinblastine or Rho123 concentration will be higher within the cell. Results for selected analogs are reported as percent effect on Rho123 intracellular concentration relative to tariquidar, where lower values indicate lesser interaction with MDR1 and therefore less potential for efflux. Moreover, the Rho123 assay was controlled by comparing WEEV inhibitors to the efflux of the known MDR1 inhibitor tariquidar.

We were also interested in undertaking MDR1 SAR studies in order to define an MDR1-recognition pharmacophore, but such work is not always trivial.[89] MDR1 binds

many different substrates, and traditional SAR, which focuses on biomolecules that bind specific substrates, is not always up to the task. Further complications arise when MDR1 binds only a few compounds from a class of analogs. There are very general SAR trends associated with the transporter, but most individual chemotypes must typically be optimized on a case-by-case basis.

Mouse Liver Microsome Metabolic Stability Assay. Metabolic stability is one of the most crucial aspects of early drug development, and instability is a potential killer of lead series.[81] It is therefore necessary to improve the stability of a compound to maximize its potential for *in vivo* efficacy, especially as half-life is related to a compound's biological availability. (See **Chapter V** for more information regarding metabolic stability in drug development.) To this end, the half-lives of compounds were determined by incubating in the presence of Balb/C mouse liver microsomes containing cytochrome p450 (CYP) enzymes and monitoring the decrease in parent compound concentration over time. Compounds more susceptible to CYP-mediated metabolism will decrease more rapidly than those that are more stable. Concentration was measured using an LC/MS/MS method developed specifically to maximize throughput, and all values were controlled against a known concentration of a structurally-related internal standard. The assay and analytical protocols were developed in our lab specifically to fit the needs of the WEEV project, and details appear in the experimental section.

Kinetic Solubility Assay. The kinetic solubility is a measure of the metastable solution equilibrium of a compound (pre-dissolved in a solvent like DMSO and added to aqueous media) that begins to precipitate as its concentration exceeds its ability to dissolve.[79, 103] This can be observed by measuring the absorbance of the sample. The

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concentration at which the compound precipitates is reported as the kinetic solubility, and is a useful measure of aqueous solubility that allows for prioritization of compounds of interest.

SAR

The average CNS drug has a molecular weight of about 319, but the lead compounds **CCG-102516** and **CCG-205432** (**Figure 8**) were much higher than that. We were therefore interested in exploring the indole and piperidine cores as potential sites for molecular weight reduction (**Figure 9**), largely due to the lack of attention these regions had received until this point. A number of analogs featuring monocyclic heterocycles lacking the fused phenyl ring of indole, such as pyrroles and imidazoles, were prepared (**Table 5**). Azetidine-containing analogs of the piperidine core were also synthesized and evaluated.
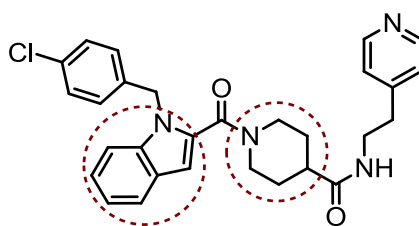
CCG-205432[†]
$IC_{50} = 0.53 \ \mu M$
$CC_{50} = 65 \ \mu M$
MW = 501
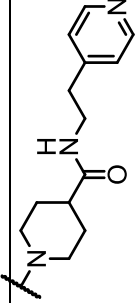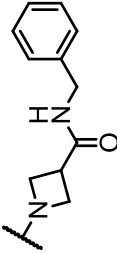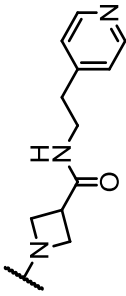
Figure 9. Sites for Molecular Weight Reduction.

Table 5. WEEV Replicon and ADME Data for Pyrrole and Imidazole Analogs.
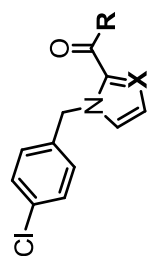
CCG No.	X	R	IC_{50} $(\mu M)^a$	CC_{50} $(\mu M)^b$	BBB-PAMPA $(\log P_{eff})$	Sol $(\mu M)^c$	MDR1 Recognition (% Rho123 uptake)^d	MLM $T_{1/2}$ $(min)^e$	MW
102516†	---	---	15.6 ± 1.8	>100	-3.6 ± 0.3	---	---	7.3 ± 3.9	486
205432†	---	---	0.53 ± 0.01	65.3	-4.18 ± 0.06	31-63	30.9 ± 1.0	11.5	501
204054	C	[structure]	77 ± 23	>100	---	---	---	---	437
206381	C	[structure]	0.68 ± 0.04	>100	-5.01 ± 0.03	125-250	-1.2 ± 0.6	2.9	451
206586	N	[structure]	>100	>100	---	---	---	---	437

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Table 5. Continued.

CCG No.	X	R	IC$_{50}$ (μM)[a]	CC$_{50}$ (μM)[b]	BBB-PAMPA (logP$_{eff}$)[c]	Sol (μM)[d]	MDR1 Recognition (% Rho123 uptake)[e]	MLM T$_{1/2}$ (min)[f]	MW
208916	N		>100	>100	-5.06 ± 0.04	>500	0.13 ± 0.37	---	437
208829	C		>100	>100	---	---	---	---	451
208915	C		22 ± 8	>100	---	---	---	---	437

[a]Inhibition of luciferase expression in WEEV replicon assay. [b]Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. [c]Log of effective permeability (cm/s) determined using PAMPA Explorer (pION) with BBB lipid mixture measured at pH = 7.4. [d]Kinetic solubility measured using the same assay media as WEEV replicon assay, except with 10% fetal bovine serum. [e]Rhodamine 123 uptake was measured in MDR1-MDCKII cells utilizing Glomax Multi Detection System (Promega). 'MDR1 recognition' was assessed by measuring uptake of Rhodamine 123 in the presence of MDR inhibitor, tariquidar (5 μM), 30 μM of test anti-viral or vehicle, and calculating: (C$_{av}$ − C$_{veh}$)*100/(C$_{tar}$-C$_{veh}$), where C$_{av}$ = concentration of rhodamine 123 in the presence of anti-viral, C$_{veh}$ = concentration in the presence of vehicle, C$_{tar}$ = concentration of rhodamine 123 in the presence of tariquidar. In the presence of tariquidar, rhodamine 123 uptake was 1123 ± 54% of vehicle controls (n=44). [f]Half-life in BALB/c mouse liver microsome incubation. Values are half-lives calculated from the equation T$_{1/2}$=ln(2)/slope, where the slope was ln(% detected) against time, plotted from the mean of at least n=3 independent experiments. †Synthesized by Janice Sindac.

Heterocyclic Monocycle SAR. The imidazole analogs (**CCG-206586** and **CCG-208916**) were completely inactive, presumably due to the enhanced ionizability and diminished lipophilicity arising from the presence of two azine nitrogens (**Table 5**). These properties would be expected to impede passive membrane permeability. Gratifyingly, the pyrrole analog **CCG-206381** was nearly equipotent with indole **CCG-205432**, and boasted improved aqueous solubility and a 50 Da improvement in molecular weight (**Table 5**). Attempts to reduce molecular weight of **CCG-206381** further, by replacing the central piperidine with azetidine, returned significantly less active compounds (**Table 5**). This may be attributed to the length-activity dependence discussed in **Chapter II**, as the small 4-membered ring is much smaller and shorter than the larger 6-membered piperidine. Curiously, the pyrrole analog of **CCG-102516** (**CCG-204054**) was substantially less active than its lead.

Potential for BBB Penetration. Passive permeability is an excellent predictor of successful BBB penetration, and high permeability may also overcome the effect of efflux transporters.[79] Therefore, compounds with good BBB permeability are of central importance to our development of CNS active inhibitors, and the pyrrole and imidazole analogs were evaluated for their potential to permeate the BBB. The notion that the significantly diminished activity of the imidazole analogs was related to increased ionizability and lipophobicity was supported by the high aqueous solubility and low BBB-PAMPA permeability of **CCG-208916** seen in **Table 5** (compounds with log P_{eff} > −4.7 are considered to be highly permeable, whereas those with log P_{eff} ≤ −6 are considered poorly permeable). **CCG-206381**, as expected, possessed improved aqueous solubility (**Table 5**), but appeared to have reduced permeability as measured by the BBB-

PAMPA assay. It is important to note that PAMPA assays are not perfect models of membranes because of reduced surface area and the presence of multiple (>100) lipid layers. Therefore, it could not be confidently concluded that **CCG-206381** was in fact less permeable than **CCG-205432**.

Recognition by efflux transporters is highly correlated with diminished BBB permeation, so selected analogs were tested for recognition by MDR1 in the Rho123 assay, using the known MDR1 substrate tariquidar as a control (**Table 5**). Significantly, **CCG-205432** interacted with MDR1 to an extent equal to 30% that of tariquidar, whereas **CCG-206381** had virtually no detectable interaction with the transporter. This indicated that **CCG-206381** could potentially evade MDR1 and thus achieve a higher *in vivo* brain concentration than **CCG-205432**.

Antiviral Activity. Due to its improved aqueous solubility and reduced molecular weight, **CCG-206381** was advanced into live virus studies and compared to **CCG-205432.** In these assays, cells were infected with live WEEV and the number of plaques – often visually distinct zones caused by infection – were counted and used to extrapolate viral concentration after treatment with varying concentrations of test compound. Viability was determined by measuring the number of living cells with the MTT assay. **CCG-205432** reduced viral titer by over two log units, and **CCG-206381** was able to successfully reduce viral titer by about one log unit (**Figure 10A**). Both compounds also comparably conferred protection to infected cells (**Figure 10B**). Together, these results indicated that, while only modestly less active, pyrrole could serve as a viable reduced molecular weight alternative to indole.

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Figure 10. WEEV Antiviral Activity for CCG-205432 and CCG-206381.
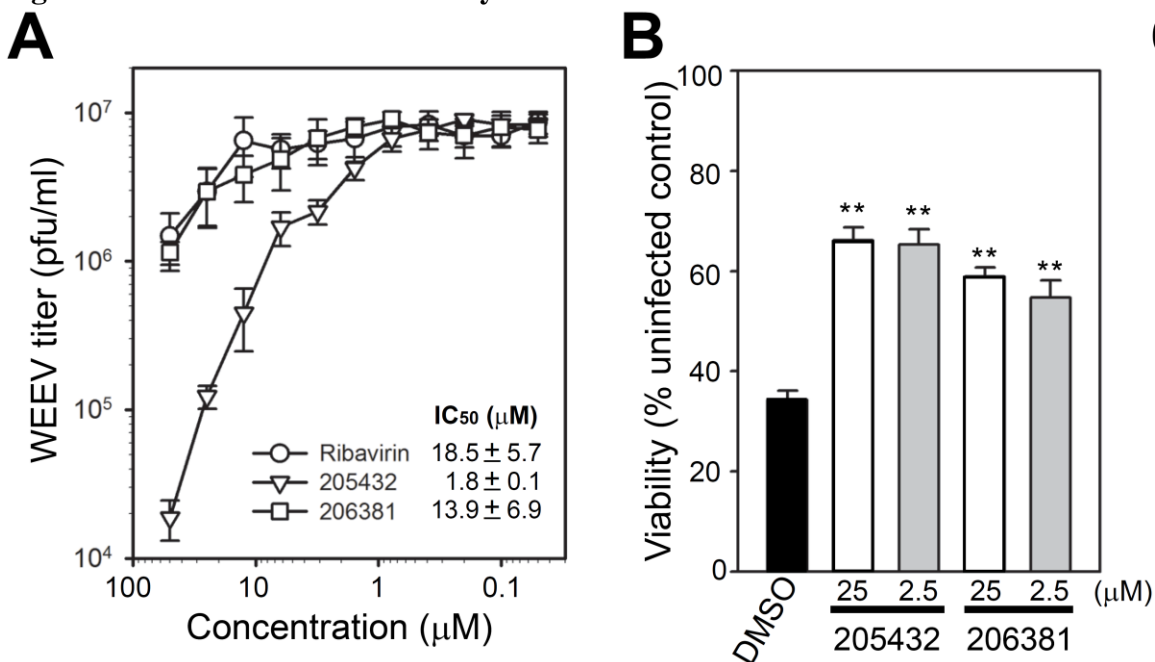
(A) HEK293 cells were infected with WEEV at a multiplicity of infection (MOI) of 1 and simultaneously treated with decreasing concentrations of the indicated compound. Virus titers in cell culture supernatants were determined at 24 hpi by plaque assay. Calculated IC_{50} values for **CCG-205432** and **CCG-206381** were 1.8 ± 0.1 and 13.9 ± 6.9, respectively. (B) Assay utilized the alphavirus WEEV. Infections were done in cultured human BE(2)-C neuronal cells. Viability was measure using an MTT assay, and viral titers were measured using a plaque assay. Values are mean $\pm$ SEM of n = 3-4 independent experiments. P value $<0.05^*$ or 0.005^{**} compared to DMSO control.

In Vivo Pharmacokinetic Properties. Despite its reduced molecular weight, **CCG-206381** was less permeable in the BBB-PAMPA assay (**Table 5**) than its indole analog **CCG-205432**. However, while lead **CCG-205432** could not achieve a measurable brain concentration in mouse pharmacokinetic studies, **CCG-206381** did register brain permeation at the early time points (**Figure 11**). This confirmed our prediction that reduction of molecular weight would enhance BBB permeability; due to significantly reduced MDR1 recognition, we can credit **CCG-206381**'s BBB penetration with enhanced evasion of efflux transporters despite its worse BBB-PAMPA permeability.

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However, the instability of **CCG-206381** towards CYP-mediated metabolism compared to **CCG-205432** in the mouse liver microsomal (MLM) assay (see **Chapter V** for more information) was reflected in its pharmacokinetic profile (**Figure 11**). **CCG-205432** had an MLM half-life of 11.5 minutes and was measurable in rat plasma for over 20 hours, whereas **CCG-206381** had an MLM half-life of 2.9 minutes and was measurable in rat plasma or brain samples only up to one hour. Overall, this data indicated that reduction of molecular weight was a viable tactic in achieving brain penetration and, therefore, *in vivo* efficacy.

Figure 11. In Vivo Pharmacokinetic Data for CCG-205432 and CCG-206381.
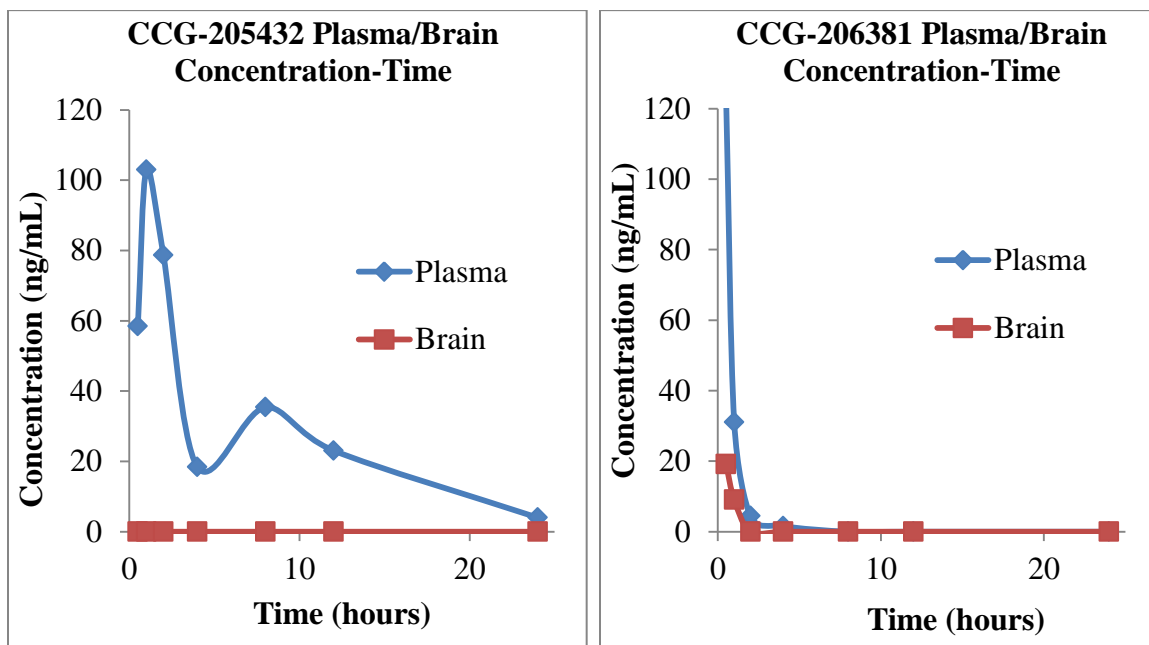
CCG-205432 and CCG-206381 possess different pharmacokinetic profiles. Balb/C mice were dosed and sacked at various time points (David Irani lab). Compound concentrations in plasma and brain samples were evaluated via LC/MS/MS by the lab of Prof. Duxin Sun.

Table 6. WEEV Replicon Data for Acyclic Low MW Analogs.

No.	R	IC$_{50}$ (μM)^a	CC$_{50}$ (μM)^b	MW
206381	---	0.68 ± 0.04	>100	451
208825		60.2 ± 4.6	>100	401
208827		40.9 ± 5	>100	415
208848		>100	>100	386
208846		>100	>100	400

^{a}Inhibition of luciferase expression in WEEV replicon assay. ^{b}Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments.

Acyclic Analog SAR. Having established that a smaller scaffold than indole could retain antiviral activity and improve BBB permeability, a variety of additional analogs was synthesized (**Table 6**) that featured complete excision of the left-hand heterocycle. Many of these analogs possess molecular weights in the low 400 to high 300 g/mol range, bringing these values substantially closer to successful CNS drugs than any WEEV inhibitor synthesized to this point. However, while the pyrrole analogs indicated that the indole left-hand core was not a necessary component of the SAR, it was immediately evident that complete removal of the heterocycle eliminated activity. Only ureas **CCG-208825** and **CCG-208827** retained any activity in the WEEV replicon assay, and those were greatly diminished relative to pyrrole **CCG-206381**. Amides **208848** and

208846 were completely inactive. These results suggested that some degree of rigidity is likely necessary to retain good antiviral activity.

Table 7. WEEV Replicon Data for Pyrrolidine Analogs.
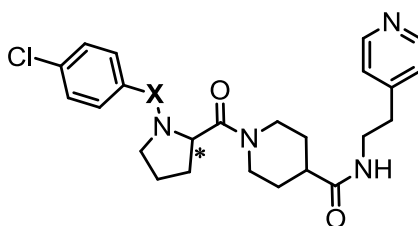
No.	X	IC_{50} $(\mu M)^a$	CC_{50} $(\mu M)^b$	MW
206381	---	0.68 ± 0.04	>100	451
(*S*)-211758	CH_2	13.5 ± 2	91.6	455
(*R*)-211757	CH_2	53.5 ± 12	>100	455
(*S*)-211754	CO	>100	>100	467
(*R*)-211756	CO	>100	>100	467

^{a}Inhibition of luciferase expression in WEEV replicon assay. ^{b}Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments.

Pyrrolidine Analog SAR. We therefore turned our attention to cyclic, but non-aromatic, pyrrolidine analogs (**Table 7**). Our rationale was that the saturated heterocycles would possess greater stability towards oxidation than pyrrole, yet provide the rigidity that was apparently required. While significantly less active than pyrrole **CCG-206381**, *N*-benzyl pyrrolidines **CCG-211758** and **CCG-211757** still retain some activity. Worth noting is the small but significant chiral preference for the *S*-enantiomer, which is four times more potent than the *R*-enantiomer. This was consistent with our previous

observation that the unknown molecular target for this series of antiviral compounds possesses some degree of enantiospecificity.[64, 83] *N*-benzoyl pyrrolidines **CCG-211754** and **CCG-211756** by contrast were completely inactive. This could suggest a need for a basic nitrogen in the linker, but more likely reflects an unfavorable conformational bias induced by the planar nature of the amide.

Arene Monocycle SAR. We then synthesized and evaluated a series of arene derivatives (**Table 8**) to more closely mimic the pyrrole scaffold but with greater predicted stability to oxidative metabolism. Several of these compounds possessed low micromolar activity in the replicon assay, including salicylamide **CCG-212392** and anthranilamide **CCG-211824**. We anticipated that the aniline moiety might be the most suitable bioisosteric replacement for pyrrole based on its electronic similarity, and indeed **CCG-211824** was only two-fold less potent than **CCG-206381**. The benzyl aniline homologue **CCG-222660** proved to be equipotent with the lead (compound **CCG-206381**), while further homologation of the anthranilamide (analog **CCG-222983**) resulted in a diminishment of activity. This observation of an optimal length is consistent with what we observed while developing **CCG-205432**[83] and suggests either a well-defined limit to the size of the unknown binding site, or simply reflects an excessive loss of entropy needed for binding a long acyclic substituent. Removal of the chloride (intended to improve metabolic stability by reducing lipophilicity) resulted in compounds (**CCG-222981** and **CCG-222982**) with greatly reduced potency. Replacement of the aniline nitrogen of **CCG-211824** with oxygen (ether analog **CCG-212392**) resulted in a modest loss of activity, while replacement with methylene (benzyl analog **CCG-222821**) provided an equipotent analogue.

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Table 8. WEEV Replicon and In Vitro ADME Data for Arene Analogs.

No.	G	IC$_{50}$ (μM)[a]	CC$_{50}$ (μM)[b]	BBB-PAMPA (log P$_{eff}$)[c]	MDR1 Recognition (% Rho123 uptake)[d]	MLM T$_{1/2}$ (min)[e]	Sol (μg/mL)[f]	MW
205432	---	0.53 ± 0.08	49.9	-4.18 ± 0.06	30.9 ± 1.0	9	31-63	501
206381	---	0.68 ± 0.04	>100	-5.01 ± 0.03	-1.2 ± 0.6	3	125-250	451
212392	(4-chlorophenoxy)	6.1 ± 0.8	56.0	-4.68 ± 0.09	9.3 ± 3.2	18		464
211824	(4-chlorophenyl NH)	1.6 ± 0.2	>100	-4.45 ± 0.11	47.7 ± 7.2	19		463
222981	(phenyl NH)	28.8 ± 5.2	>100	-4.75 ± 0.02	10.9 ± 1.7	---	>250	429

Table 8. Continued.

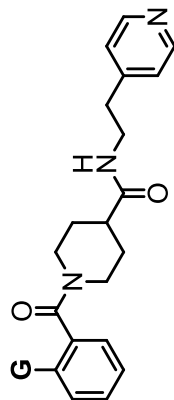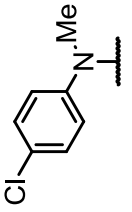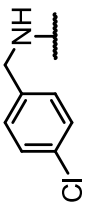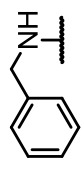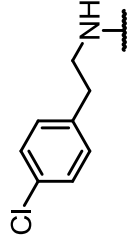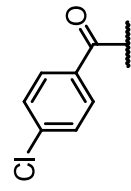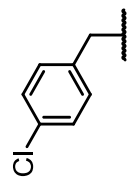| No.	G	IC$_{50}$ (µM)[a]	CC$_{50}$ (µM)[b]	BBB-PAMPA (log P$_{eff}$)[c]	MDR1 Recognition (% Rho123 uptake)[d]	MLM T$_{1/2}$ (min)[e]	Sol (µg/mL)[f]	MW
212393		15.4 ± 2.8	>100	-4.71 ± 0.09	3.9 ± 1.5	---	>250	477
222660		0.56 ± 0.08	75.2	-4.39 ± 0.05	59.6 ± 7.5	15		477
222982		19.0 ± 1.7	92.2	-4.93 ± 0.03	2.5 ± 0.3	---	>250	443
222983		3.49 ± 0.1	86.0	-4.49 ± 0.35	30.7 ± 4.7	---	>250	491
212391		>50	>50	-5.08 ± 0.02	1.8 ± 1.3	---		476
222821		1.46 ± 0.09	70.7	-4.40 ± 0.06	22.6 ± 2.7	3		462

Table 8. Continued.

[a]Inhibition of luciferase expression in WEEV replicon assay. [b]Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. [c]Log of effective permeability (cm/s) determined using PAMPA Explorer (pION) with BBB lipid mixture measured at pH = 7.4. [d]Rhodamine 123 uptake was measured in MDR1-MDCKII cells utilizing Glomax Multi Detection System (Promega). 'MDR1 recognition' was assessed by measuring uptake of Rhodamine 123 in the presence of MDR inhibitor, tariquidar (5 μM), 30 μM of test anti-viral or vehicle, and calculating: $(C_{av} - C_{veh})*100/(C_{tar}-C_{veh})$, where C_{av} = concentration of rhodamine 123 in the presence of anti-viral, C_{veh} = concentration in the presence of vehicle, C_{tar} = concentration of rhodamine 123 in the presence of tariquidar. In the presence of tariquidar, rhodamine 123 uptake was 1123 ± 54% of vehicle controls (n=44) . [e]Half-life in BALB/c mouse liver microsome incubation. Values are half-lives calculated from the equation $T_{1/2}$=ln(2)/slope, where the slope was ln(% detected) against time, plotted from the mean of at least n=3 independent experiments. [f]Kinetic solubility measured using the same assay media as WEEV replicon assay, except with 10% fetal bovine serum. See Experimental Section for detailed methods and synthetic procedures.

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Interestingly, the *N*-methylated analogue **CCG-212393** was ten times less active than its parent anthranilamide **CCG-211824**, suggesting that the aniline nitrogen may be engaged in internal hydrogen bonding with the ortho carbonyl oxygen, resulting in a specific conformation favorable for activity. Further evidence for this conformational hypothesis is provided by comparing benzylbenzamide analogue **CCG-222821**, with inactive benzoylbenzamide **CCG-212391.** The latter would not be expected to be able to achieve a conformation similar to the internally hydrogen bonded conformation of **CCG-211824**.
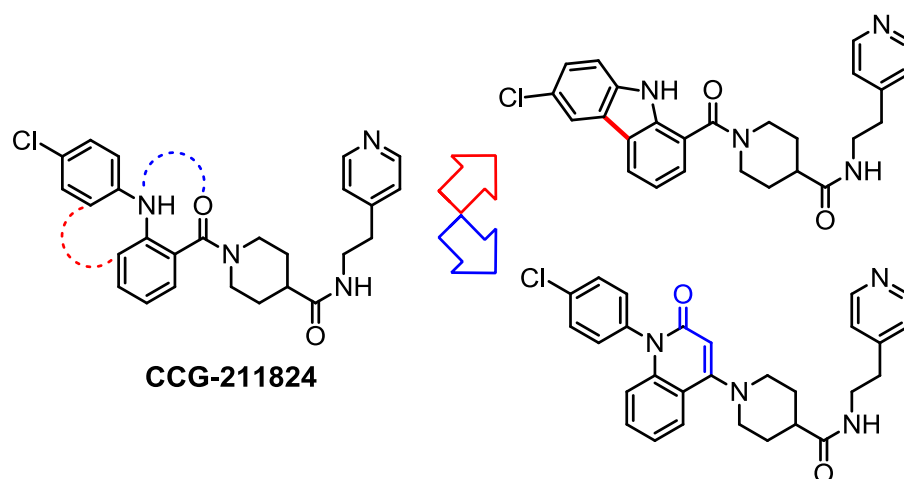
Figure 12. Conformational Restriction Plan for Arene Analogs.
Analogs were designed to tether the arene rings (red, carbazole derivatives) or tie the amide to the aniline nitrogen (blue, quinolone derivatives).

Conformational Restriction. Several rigid analogues were designed to evaluate our conformational hypothesis (**Figure 12** and **Table 9**). Carbazole derivatives (**CCG-222824** and **CCG-222825**) were intended to tether the two arenes in the molecule and prevent rotation around the two C-N bonds. Surprisingly, these compounds were remarkably less active than their more flexible parent, which may indicate that the putative internal NH hydrogen bond is not sufficient for achieving the biologically active

conformation and that in fact the two tethered aromatic rings need to be orthogonal rather than planar. It is also possible that tight binding to the unknown molecular target entails induced fit, which the rigid carbazole cannot accommodate. The quinolone derivatives (**CCG-222823** and **CCG-222822**), on the other hand, were designed to freeze rotation about the aniline-amide C-C bond. Their poor activity, however, indicates either a poor match for the biologically active conformation, or a key role for the benzamide carbonyl of **CCG-211824** that has been disrupted.

Table 9. WEEV Replicon Date for Rigid Arene Analogs.
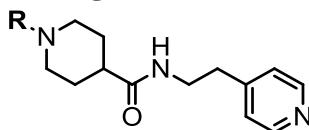
No.	G	IC$_{50}$ (µM)^{a}	CC$_{50}$ (µM)^{b}
222824		59.2 ± 6.5	>100
222825		18.7 ± 5.4	66.1
222823		88.3 ± 11.7	>100
222822		10.9 ± 2.0	34.1

[a]Inhibition of luciferase expression in WEEV replicon assay.
[b]Cell viability determined by the MTT reduction assay.
Values are mean of at least n=3 independent experiments.

Potential for CNS Penetration. To measure the potential for MDR1 recognition of the arene analogs, we used the Rho123 assay.[83] In **Table 8**, replacement of the pyrrole of **CCG-206381** with phenyl (**CCG-211824**) markedly increased recognition by MDR1. Surprisingly, simple *N*-methylation of **CCG-211824** (**CCG-212393**) almost completely abated MDR1 recognition. Reductions in MDR1 recognition were also observed upon replacement of the NH of **CCG-211824** with O, C=O, or CH$_2$ (**CCG-212393**, **CCG-212391** and **CCG-222821**, respectively) suggesting that the NH may be a key factor for recognition. This is supported by the strong MDR1 recognition of NH homologs **CCG-222660** and **CCG-222983**. However, it is notable that simple removal of the aromatic chloro group strongly attenuated MDR1 recognition (**CCG-222981** and **CCG-222982**), indicating that overall lipophilicity may also be a factor.

Permeability is an excellent predictor of potential BBB penetration, so selected compounds were evaluated using pION's BBB-PAMPA assay and PAMPA Explorer program as previously described.[83] Compounds with log $P_{\text{eff}} > -4.7$ are considered to be highly permeable, whereas those with log $P_{\text{eff}} \leq -6$ are considered poorly permeable. In general, our compounds exhibit moderate to high BBB permeability, and all but one (**CCG-212391**) possess modestly better permeability than pyrrole lead **CCG-206381** (**Table 8**).

In vitro Metabolic Stability. Based on their WEEV replicon activities, selected compounds were advanced into the mouse liver microsome (MLM) assay to assess their stabilities towards Phase I oxidative metabolism (**Table 8**). (See **Chapter V** for more information regarding metabolism.) The stability of aniline **CCG-211824** was improved with a half-life of 19 minutes, a six-fold improvement over the slightly more electron-rich

pyrrole **CCG-206381** (3 minutes) and a two-fold improvement over the indole **CCG-205432** (9 minutes). The chlorobenzyl homologue **CCG-222660** was slightly less stable towards metabolism than **CCG-211824**, perhaps because of the introduction of a benzylic carbon and increased lipophilicity. Phenolic ether **CCG-212392** was as stable towards microsomal metabolism as aniline **CCG-211824**. Interestingly, replacement of the NH of **CCG-211824** with CH_2 (**CCG-222821**) markedly attenuated metabolic stability, due either to increased lipophilicity or the creation of an additional site for oxidation.

Table 10. Antiviral Data for Select Analogues[a]

| CCG No. | Conc. (μM) | FMV | | WEEV |
		Titer (x 10^6 pfu/ml)	Viability (% uninfected control)	Titer (x 10^4 pfu/ml)
DMSO	NA	30.7 ± 9.0	35.6 ± 4.7	37.3 ± 7.3
205432	25	ND	$69.5 \pm 10.0^*$	$1.1 \pm 0.1^{**}$
211824	25	$3.7 \pm 1.4^*$	58.1 ± 9.0	$3.2 \pm 2.2^*$
	5	10.6 ± 4.1	54.6 ± 9.2	49.2 ± 19.4
222660	25	$3.2 \pm 1.0^{**}$	$66.6 \pm 9.5^*$	$1.6 \pm 0.5^{**}$
	5	$9.4 \pm 5.6^*$	$73.3 \pm 9.1^*$	$11.7 \pm 3.6^*$

[a]Assay utilized the alphaviruses Fort Morgan Virus (FMV) and western equine encephalitis virus (WEEV). Infections were done in cultured human BE(2)-C neuronal cells. Viability was measure using an MTT assay, and viral titers were measured using a plaque assay. Values are mean ± SEM of n = 3-4 independent experiments. P value <0.05[*] or 0.005[**] compared to DMSO control. NA, not applicable. ND, not determined. Data from the David Miller lab.

Antiviral Activity. Based on their WEEV replicon potencies, **CCG-211824** and **CCG-222660** were advanced into assays to evaluate their ability to inhibit cellular replication of infectious live virus. Our collaborators used two parallel assays to measure activity against infectious virus: reduction in cytopathic effect (CPE) and extracellular

virus titers, and examined activity against both WEEV and Fort Morgan Virus (FMV), a WEEV-serogroup alphavirus that can be used safely under reduced biosafety level conditions compared to WEEV. Against FMV, both **CCG-211824** and **CCG-222660** reduced virus titers by ten-fold when used at 25 μM, and also reduced FMV-induced CPE similar to previous lead **CCG-205432** (**Table 10**). Furthermore, both **CCG-211824** and **CCG-222660** also reduced WEEV titers by at least 10-fold when used at 25 μM. Although we did not do full titration curves with these experiments, **CCG-222660** maintained antiviral activity against both FMV and WEEV when used at 5 μM, whereas **CCG-211824** had reduced activity at this lower concentration (**Table 10**).
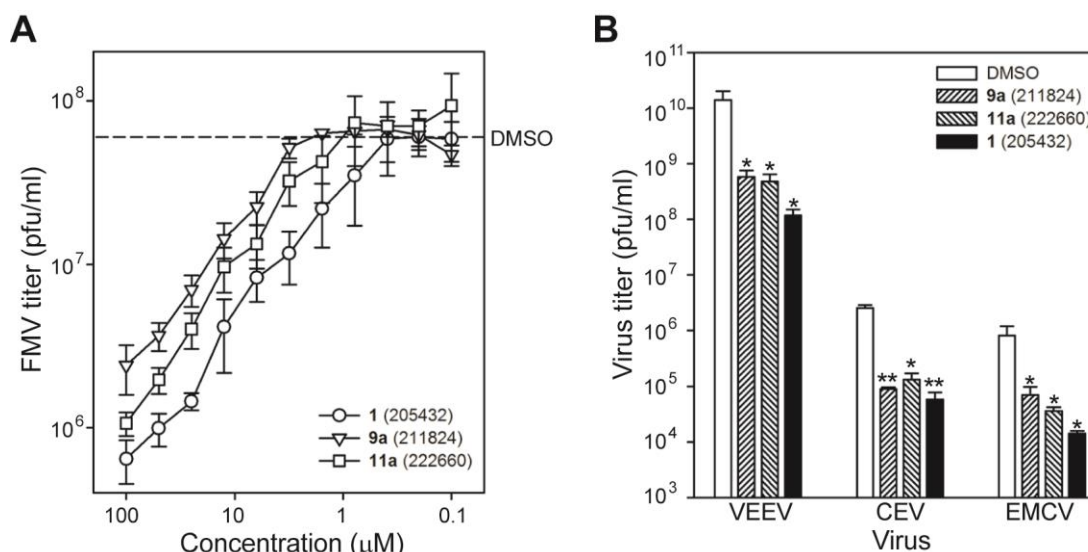
Figure 13. Spectrum Antiviral Data for Selected Analogues.
CCG-211824 and **CCG-222660** have potent and broad spectrum antiviral activity. (A) HEK293 cells were infected with FMV at an MOI of 1 and simultaneously treated with decreasing concentrations of the indicated compound. Virus titers in cell culture supernatants were determined at 24 hpi by plaque assay. The horizontal dashed line indicates FMV titers in cells treated with control DMSO. Calculated IC_{50} values for **CCG-205432**, **CCG-211824**, and **CCG-222660** were 1.0 ± 0.5, 5.2 ± 1.0, and 2.3 ± 1.2 μM, respectively. (B) HEK293 cells were infected with the indicated viruses at an MOI of 0.1, simultaneously treated with the indicated compound at 25 μM, and virus titers in tissue culture supernatants were determined by plaque assay at 24 hpi. P <0.05* or 0.005** compared to DMSO-treated controls. Data from the David Miller Lab.

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To further evaluate the antiviral potency and breadth of activity for **CCG-211824** and **CCG-222660**, we completed both full titration studies with FMV in HEK293 cells (**Figure 13A**) and examined their inhibitory activity against viruses derived from three different families: Venezuelan equine encephalitis virus (VEEV, TC-83 vaccine strain; *Togaviridae*), California encephalitis virus (CEV; *Bunyaviridae*), and encephalomyocarditis virus (EMCV; *Picornaviridae*) (**Figure 13B**). Both **CCG-211824** and **CCG-222660** had dose-dependent antiviral activity against FMV, where **CCG-222660** had an IC_{50} similar to previous lead **CCG-205432** (**Figure 13A**). In addition, both **CCG-211824** and **CCG-222660** had demonstrable antiviral activity against VEEV, CEV, and EMCV in cultured cells, with an approximate 10-fold reduction in infectious virus titers for all three pathogens, similar to previous lead **CCG-205432** (**Figure 13B**).

Mechanism of Action Studies. The use of the WEEV replicon assay, a phenotypic cell-based assay, increases the probability of selecting compounds having a cellular rather than a viral target for antiviral activity. There are several advantages of targeting host factors over viral proteins. The most significant advantage is a decreased probability of the targeted virus developing resistance to therapeutics.[105] Our collaborator's studies with the lead **CCG-205432** and related carboxamide analogs indicated that the mechanism of action for their antiviral activity is through the modulation of cellular cap-dependent translation.[104] To examine whether our monocyclic analogs used a similar mechanism, we evaluated the ability of **CCG-211824** and **CCG-222660** to modulate cellular cap-dependent translation (**Figure 14A**). For these experiments, we used BSR-T7 cells and two reporter plasmids: pSV40-LUC, which produces a capped mRNA through nuclear transcription and export, and pCITE-LUC,

which produces an uncapped mRNA containing a cap-independent translational element (CITE) through cytoplasmic transcription that is driven by a T7 promoter (**Figure 14B**).[106] Similar to lead **CCG-205432** and previous generation compounds, both **CCG-211824** and **CCG-222660** had no effect upon cap-independent translation, but potently inhibited cellular cap-dependent translation of transcripts produced by the pSV40-LUC reporter plasmid (**Figure 14B**). These results suggest that the anthranilamide analogs **CCG-211824** and **CCG-222660** also function as antiviral compounds, in part, through suppression of host cell cap-dependent translation.
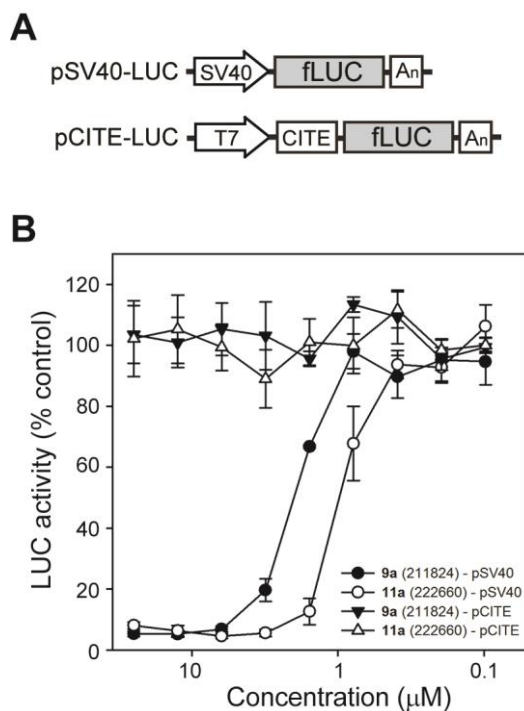
Figure 14. Effect of Selected Analogs on Transfected Gene Expression in BSR-T7 Cells.[a]
9a and **11a** modulate cellular gene expression. BSR-T7 cells were transfected with the expression plasmids pSV40-LUC (circles) or pT7/CITE-LUC (triangles), treated with decreasing concentrations of the indicated compound, and fLUC activity was measured 20 h later. Calculated IC_{50} values for suppression of pSV40-LUC activity for **9a** and **11a** were 2.2 ± 0.2 and 1.0 ± 0.1 μM, respectively. **1** has an IC_{50} value of 0.1 μM in this assay.[104] N=3 or 4 for all data, and results are the mean $\pm$ SEM. Data from the David Miller lab.

Synthesis

The piperidine-containing pyrrole analogs **CCG-204054**, **CCG-206381**, and **CCG-208914** and the imidazole analogs **CCG-206586**, **CCG-208913**, and **CCG-208916** were first prepared by alkylating the appropriate azole with 4-chlorobenzyl chloride to provide the methyl ester. Saponification of this ester and amide coupling with ethyl isonipecotate gave the intermediate. This ethyl ester was then hydrolyzed and coupled with the appropriate amine (**Scheme 7**).

Scheme 7. Preparation of Pyrrole and Imidazole Analogs.

X = CH, R = Me, **22a**
X = N, R = Et, **22b**

23a,b

24a,b

25a,b

26

27

X = CH, R = Bn, **CCG-204054**
X = CH, R = pyr, **CCG-206381**
X = CH, R = ind, **CCG-208914**
X = N, R = Bn, **CCG-206586**
X = N, R = ind, **CCG-208913**
X = N, R = pyr, **CCG-208916**

"Bn"

"pyr"

"ind"
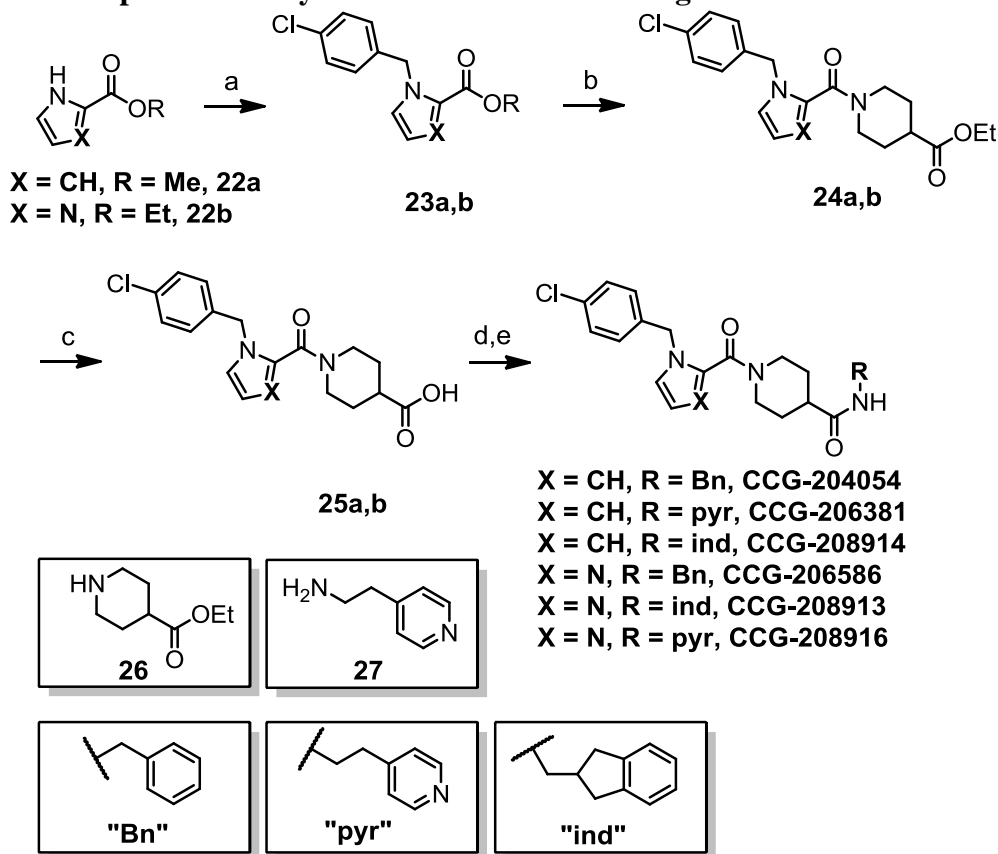
Reagents and conditions: (a) *p*-chlorobenzyl chloride, K_2CO_3, DMF, 60 °C, 36 h; (b) 7M NaOH, EtOH, 70 °C, 4 h; (c) **26**, EDC·HCl, HOBt, DIPEA, DMF, rt, 24 h; (d) 7M NaOH, EtOH, 60 °C, 8 h; (e) **27**, EDC·HCl, HOBt, DIPEA, DMF, rt, 24 h.

Scheme 8. Preparation of Azetidine Analogs.[a]
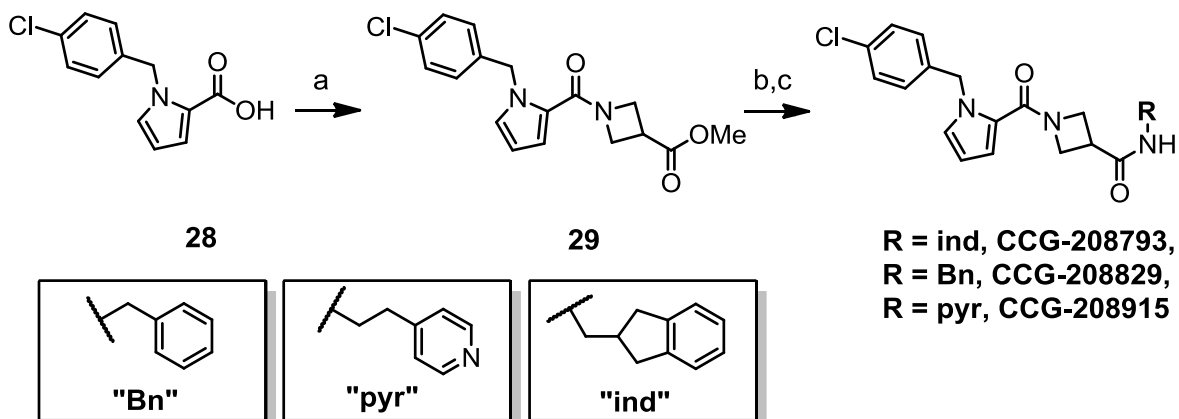
28   **29**

R = ind, CCG-208793,
R = Bn, CCG-208829,
R = pyr, CCG-208915

"Bn"   "pyr"   "ind"

[a]Reagents and conditions: (a) methyl 4-azetidinecarboxylate hydrochloride, EDC, HOBT, DIPEA, DCM, rt; (b) 10% aq. NaOH, EtOH; (c) **R-NH₂**, EDC, HOBT, TEA, DCM, rt.

The azetidine-containing pyrroles **CCG-208793**, **CCG-208829**, and **CCG-208915** were prepared through the union of *N*-benzyl pyrrole **28** and methyl azetidine-3-carboxylate, followed by base hydrolysis and amidation with the appropriate amine (**Scheme 8**). Urea analogs **CCG-208825**, **CCG-208826**, **CCG-208827**, and **CCG-208828** were synthesized by the addition of ethyl isonipecotate **26** to isocyanates **30a,b**, followed by hydrolysis and amidation with the appropriate amine (**Scheme 9**). The acyclic amides **CCG-208846**, **CCG-208847**, **CCG-208848**, and **CCG-208849** were synthesized from the amide coupling of the appropriate carboxylic acid **32a,b** and ethyl isonipecotate **26**, followed by hydrolysis/amidation with the appropriate amine similar to the previous syntheses (**Scheme 9**).

Scheme 9. Preparation of Acyclic Amide and Urea Analogs.[a]

n = 1, 30a
n = 2, 30b

31a,b
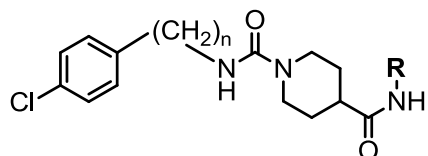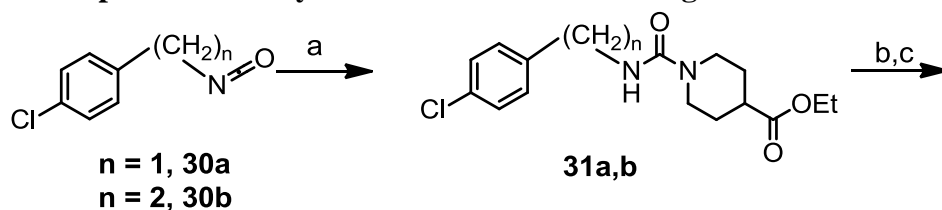
n = 1, R = pyr, CCG-208825
n = 1, R = ind, CCG-208826
n = 2, R = pyr, CCG-208827
n = 2, R = ind, CCG-208828

n = 1, 32a
n = 2, 32b

33a,b
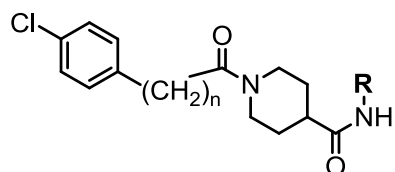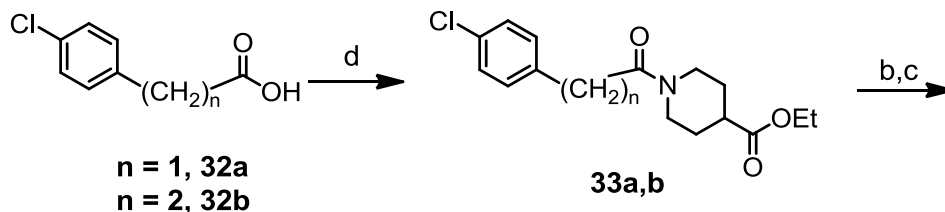
n = 1, R = pyr, CCG-208848
n = 1, R = ind, CCG-208849
n = 2, R = pyr, CCG-208846
n = 2, R = ind, CCG-208847

"pyr"

"ind"
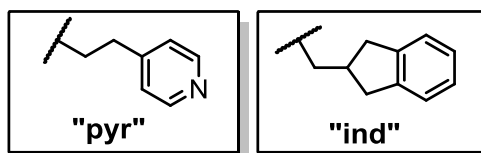
[a]Reagents and conditions: (a) **26**, DCM, rt; (b) 10% aq. NaOH, EtOH, rt; (c) **R-NH$_2$**, EDC, HOBT, TEA, DCM, rt; (d) **26**, EDC·HCl, HOBT, TEA, DCM, RT.

The right-hand portion of the pyridine-bearing analogs, *N*-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide dihydrochloride **36** (**Scheme 10**), was assembled by first boc-protecting ethyl isonipecotate **26**. The protected ethyl ester was then hydrolyzed to the carboxylic acid and coupled with 4-(2-aminoethyl)pyridine **27**. The protecting

group was then removed under dry acidic conditions to yield the dihydrochloride salt **36** of the desired amine.

Scheme 10. Preparation of the Right-Hand Amine Dihydrochloride 36.[a]
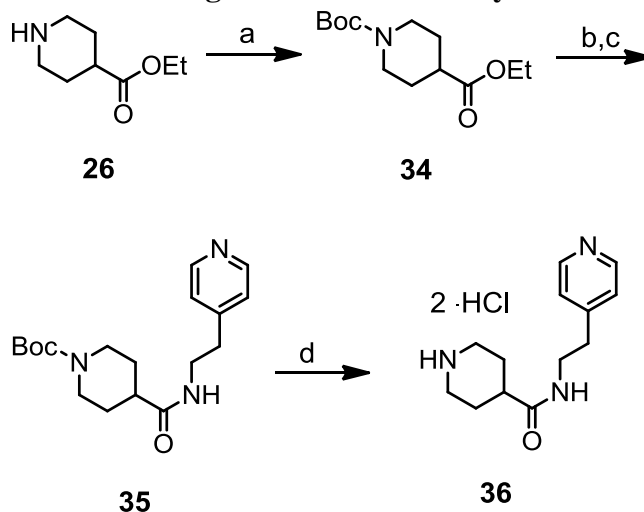
[a]Reagents and conditions: (a) Boc$_2$O, Na$_2$CO$_3$, H$_2$O, THF, reflux; (b) NaOH, H$_2$O, EtOH, RT; (c) 4-(2-aminoethyl)pyridine, EDC·HCl, HOBT, DCM, RT; (d) HCl, dioxane, RT.

N-benzylpyrrolidine enantiomers **CCG-211757** and **CCG-211758** were prepared through base-catalyzed alkylation of proline with 4-chlorobenzylchloride to protect stereochemical integrity via the carboxylate anion,[107] followed by amide coupling with amine dihydrochloride **36**[83] to yield the appropriate (*R*)- and (*S*)- analogs (**Scheme 11**). Pyridine-bearing enantiomers **CCG-211754** and **CCG-211756**, and indane-bearing enantiomers **CCG-211753** and **CCG-211755**, were synthesized in similar fashion, first by acylating proline under classical Schotten-Baumann conditions[108, 109] followed by amide coupling with ethyl isonipecotate **26** (**Scheme 11**). Hydrolysis of the ethyl ester **(R)-** or **(S)-40** through a non-racemizing protocol[110] provided the carboxylic acid, and amidation with amine **27** gave the desired (*R*)- and (*S*)- benzoyl analogs.

Scheme 11. Preparation of *N*-Benzyl and *N*-Benzoyl Pyrrolidine Analogs.[a]

(R)-37
(S)-37

(R)-38
(S)-38

(R)-CCG-211757
(S)-CCG-211758

(R)-39
(S)-39

(R)-40
(S)-40

R = ind, (R)-CCG-211753
R = pyr, (S)-CCG-211754
R = ind, (S)-CCG-211755
R = pyr, (R)-CCG-211756

"pyr"

"ind"
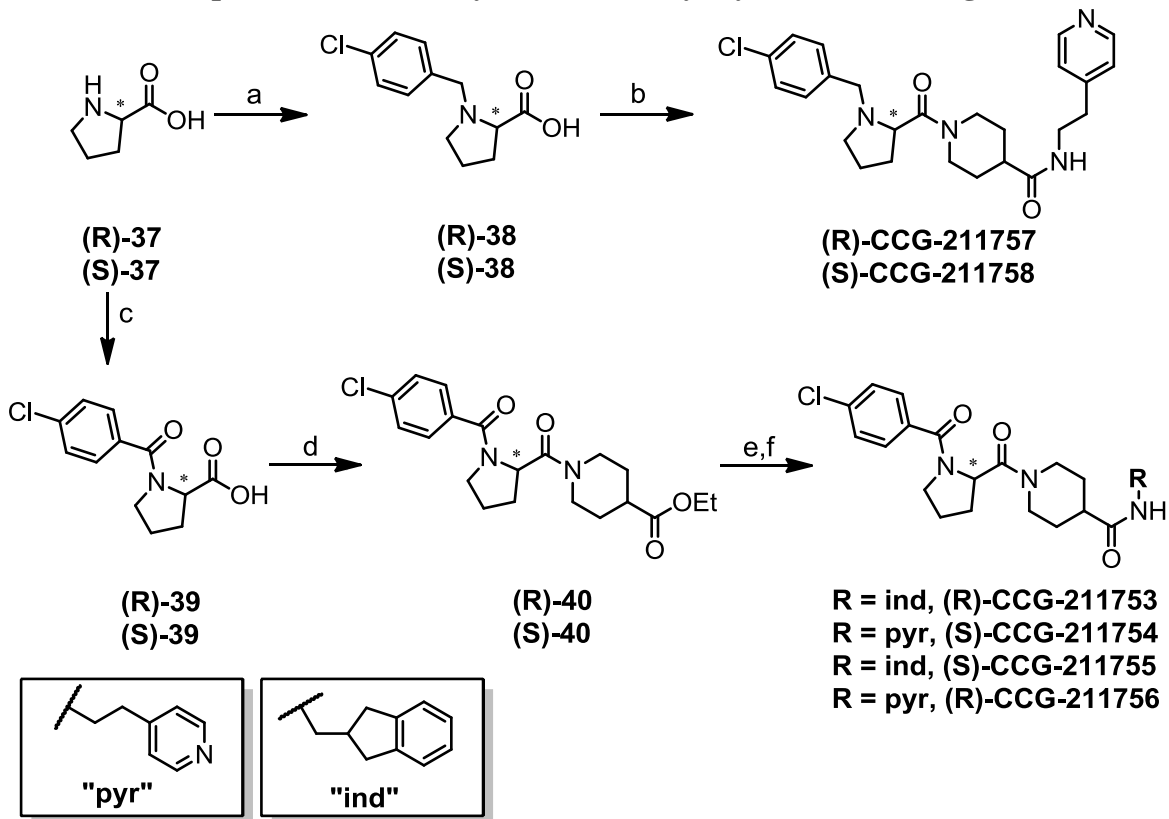
[a]Reagents and conditions: (a) **2a**, KOH, IPA, 40 °C; (b) **36**, HATU, TEA, DCM, RT; (c) 4-chlorobenzoylchloride, IPA, KOH, H_2O, 0 °C; (d) **26**, HATU, TEA, DCM, RT; (e) 9:1 DCM:MeOH, NaOH, rt; (f) **27**, HATU, TEA, DCM, RT.

Salicylamide analogue **CCG-212392** was prepared from the Chan-Lam coupling[111] of methyl salicylate **41** and 4-chlorophenyl boronic acid, providing the ether intermediate **42** in low yield, followed by ester hydrolysis and amidation (**Scheme 12**). Secondary phenyl anthranilamides **CCG-211824** and **CCG-222981** were assembled from Buchwald amination[112, 113] of triflated methyl salicylate and the appropriate aniline, followed by saponification/amidation. The *N*-methyl anthranilamide derivative **CCG-**

212393 was accessed through methylation of coupling intermediate **43a**, followed by the usual saponification and amide coupling.

Scheme 12. Preparation of Anthranilamide and Salicylamide Analogues.[a]

Benzyl anthranilamides **CCG-222660** and **CCG-222982** were obtained from the alkylation of methyl 2-aminobenzoate **46** and the corresponding benzyl halide, followed by the base hydrolysis and amidation (**Scheme 13**).

Phenethyl anthranilamide **CCG-222983** could not be prepared through direct *N*-alkylation of methyl 2-aminobenzoate because the electrophile was highly prone to elimination. Conveniently, the transformation could be achieved through a Jourdan-Ulmann coupling[114] of 2-bromobenzoic acid **48** and 1-(2-aminoethyl)-4-chlorophenyl amine in ethylene glycol dimethyl ether (**Scheme 14**). Base hydrolysis and amidation then provided the desired analog.

Scheme 13. Preparation of Benzyl Anthranilamide Analogues.[a]

R = Cl, 47a
R = H, 47b

R = Cl, CCG-222660
R = H, CCG-222982

46
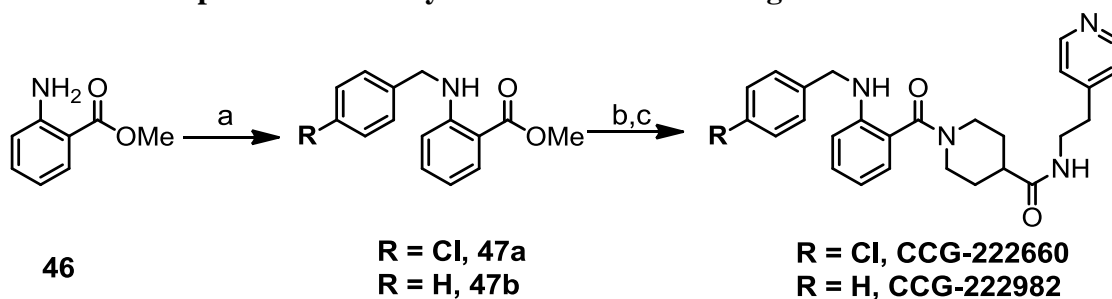
[a]Reagents and conditions: (a) 4-chlorobenzyl chloride or benzyl bromide, *t*BuOK or Cs$_2$CO$_3$, DMF; (b) 10% aq. NaOH, EtOH, rt; (c) **36**, EDC, HOBT, TEA, DCM, rt.

Scheme 14. Preparation of Phenethyl Anthranilamide Analogue CCG-222983.[a]

48   **49**   **CCG-222983**
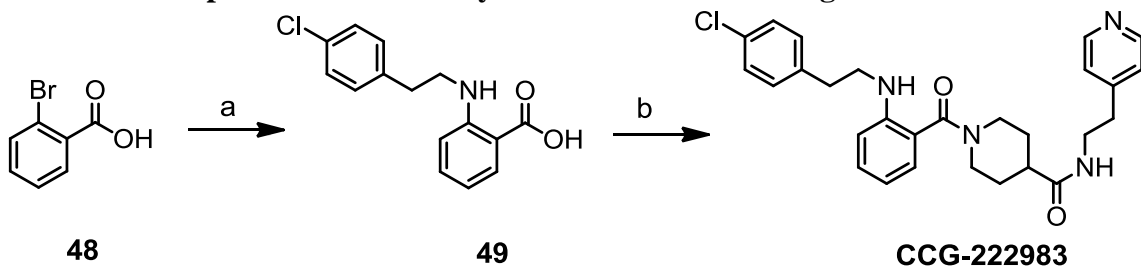
[a]Reagents and conditions: (a) 1-(2-aminoethyl)-4-chlorophenyl amine, Cu$_2$O, Cu^0, K$_2$CO$_3$, EDME, 130 °C; (b) **36**, EDC, HOBT, TEA, DCM, rt.

Scheme 15. Preparation of Benzyl and Benzoyl Benzoate Analogues.[a]
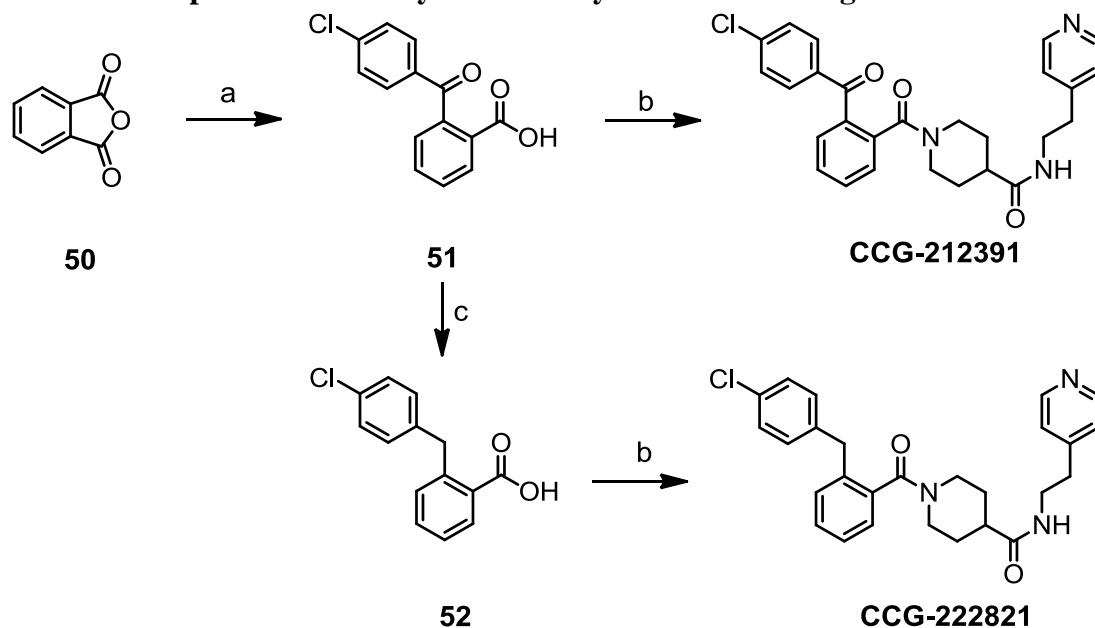
[a]Reagents and conditions: (a) AlCl$_3$, PhCl, reflux; (b) **36**, EDC, HOBT, TEA, DCM, rt; (c) Zn^0, CuSO$_4$·5H$_2$O, 28% aq. NH$_3$, reflux.

The preparation of benzyl- and benzoyl benzamide derivatives (**CCG-222821** and **CCG-212391** respectively) entailed the known Friedel-Crafts acylation of chlorobenzene with phthalic anhydride, [115] which delivered ketone intermediate **51** from which a simple amidation with **36** provided the benzoylbenzamide analogue **CCG-212391** (**Scheme 15**). The related benzylbenzamide analogue **CCG-222821** was accessed through a dissolving metal reduction of the ketone intermediate **51**[116, 117] to afford benzyl benzoic acid **52**, which was followed by amidation with amine **36**. Interestingly, the ketone could not be reduced without concomitant reduction of the chloride under several heterogenous hydrogenation conditions.[115, 118] Also, attempts to reduce the ketone to the respective alcohol with NaBH$_4$ failed,[119] even with excess borohydride and vigorous conditions. Thionation with Lawesson's Reagent and subsequent Raney-Ni reduction also could be

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used to generate the desired benzyl intermediate **52**, but the dissolving metal reduction in **Scheme 15** was found to be more direct and efficient.

Scheme 16. Preparation of Carbazole Analogues.[a]
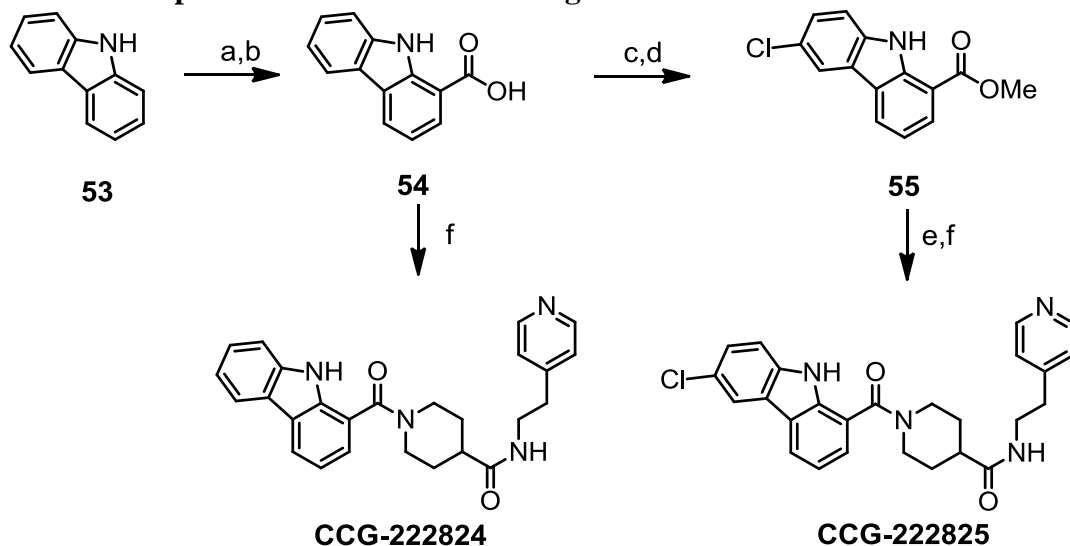
[a]Reagents and conditions: (a) pyrrolidine, 37% aq. HCHO, EtOH, reflux; (b) s-BuLi, THF, $CO_{2(g)}$, then 1 M aq. HCl, reflux; (c) sat. methanolic HCl, reflux; (d) SO_2Cl_2, DCM, 0 °C; (e) 10% aq. NaOH, EtOH, THF, rt; (f) **36**, EDC, HOBT, TEA, DCM, rt.

Carbazole analogues were synthesized as shown in **Scheme 16**. Ortho-directed lithiation of carbazole **53** followed by carboxylation with carbon dioxide in a manner similar to that reported by Katritzky provided key intermediate **54**.[120] Direct coupling with amine **36** provided analogue **CCG-222824**. The carbazole carboxylic acid **54** could not be chlorinated with NCS under a variety of conditions,[121, 122] while the methyl ester yielded a complex mixture of variously chlorinated carbazoles. Ultimately, sulfuryl chloride[123] gave clean regioselective conversion to chloro-substituted carbazole ester **55** as confirmed by ^{1}HNMR. This ester was then hydrolyzed to the corresponding carboxylic acid and coupled with amine **36** to give the desired analogue **CCG-222825**.

Scheme 17. Preparation of Quinolone Analogues.[a]
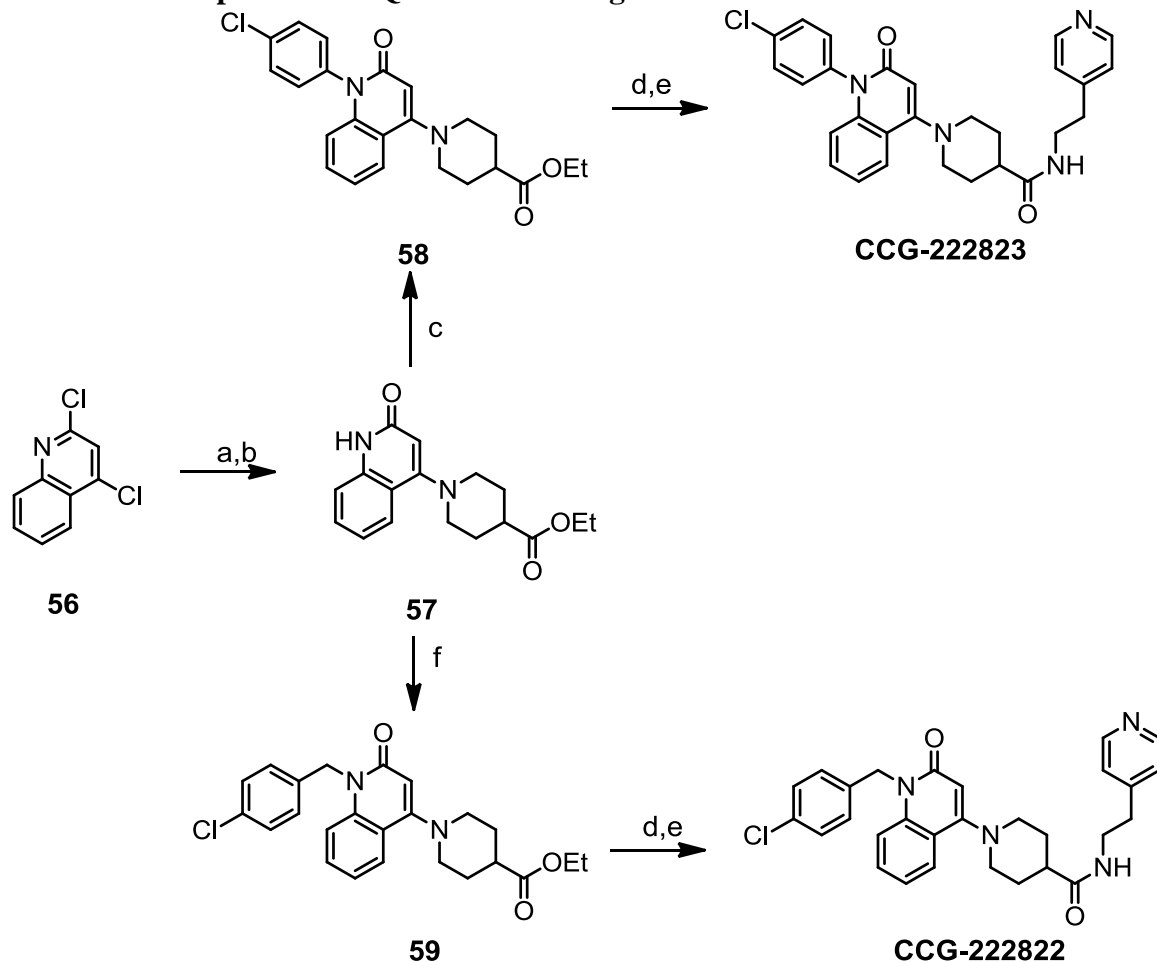
[a]Reagents and conditions: (a) 6 M aq. HCl, 1,4-dioxane, reflux; (b) ethyl isonipecotate, NaI, Na$_2$CO$_3$, DMF; 80 °C; (c) 4-chlorophenylboronic acid, Cu(OAc)$_2$, TEA, pyridine, DCM, rt; (d) 10% aq. NaOH, EtOH, THF, rt; (e) 4-(2-aminoethyl)pyridine, EDC, HOBT, TEA, DCM, rt; (f) 4-chlorobenzyl chloride, KOtBu, DMF, 0 °C to rt.

Finally, quinolone analogues were accessed as shown in **Scheme 17**. Hydrolysis of 2,4-dichloroquinoline **56**[124, 125] and subsequent addition/elimination with ethyl isonipecotate **26** gave intermediate **57** which subsequently could be *N*-arylated via a Chan-Lam coupling,[126] then saponified and coupled with amine **27** to give phenylquinolone **CCG-222823**. Conversely, **57** could be alkylated with 4-chlorobenzyl

chloride **2a**, hydrolyzed and coupled under similar conditions to the phenylquinolone to provide the benzylquinolone **CCG-222822**.

Conclusion

The primary goal of this chapter was the reduction of molecular weight, as low MW is associated with improved BBB-permeability and is a feature of successful CNS drugs. To this end, we successfully developed a novel MDR1-evasive pyrrole compound (**CCG-206381**) with sub-micromolar potency and demonstrable *in vivo* CNS penetration. This marks the first successful alphavirus antiviral with measurable CNS exposure, and confirmed our predictions that reduction of MW was a viable tactic in achieving favorable CNS permeability. However, this compound was remarkably susceptible to CYP-mediated metabolism; therefore, an additional goal was improvement of metabolic stability, and we identified the aniline moiety as a suitable substitute for pyrrole. We unveiled phenylaniline **CCG-211824** and benzylaniline **CCG-222660** as low-to-sub-micromolar inhibitors with MLM half-lives twice that of the original indole lead **CCG-205432** and five-to-six times that of the pyrrole lead **CCG-206381**. Importantly, these aniline compounds demonstrated excellent activity (almost two-fold titer reduction) against live viruses from three different families. This is an important step towards the development of antivirals that are both potent and bioavailable.

Chapter IV. **Conformational Restriction**

Rationale

Potency is important for many aspects of drug delivery; potent compounds are generally more amenable to oral administration because they require less material to achieve the desired therapeutic effect, and they are less prone to off-target effects (toxicity) because of the low doses required.[79] Therefore, as with any drug, a high therapeutic index, representing high potency and low toxicity, is a major goal for WEEV inhibitors. Potency is also especially important for the successful action of many CNS-active drugs; due to the brain-blood barrier (BBB), very little of the available drug will actually penetrate the brain, as described later.

An important task in potency optimization is the identification of the bioactive conformation of the inhibitors.[127-130] In the absence of structural information, this can only be accomplished by synthesizing and evaluating rigid analogs.[131, 132] Of the multiple conformations that a drug can adopt, only a small subset will have maximal activity if the binding site is well defined and not highly flexible.[128, 133] It is therefore informative to lock out or freeze certain conformations and observe their effect on activity in an effort to pinpoint the active one, especially when the binding site is unknown.[134] It is also a very useful strategy for improving selectivity versus off-target effects.[135-137] When designing such analogs, it is important to minimize the number of additional atoms used in locking a conformation because adding molecular weight or introducing new interactions with the

binding site may have their own effects on activity, thereby confounding interpretation of the actual impact of conformational restriction. Rigid analogs may be prepared utilizing several approaches to restricting rotatable bonds: a) substituting small functionality with sterically demanding functionality; b) bridging nonadjacent atoms; and c) restricting ring conformations.
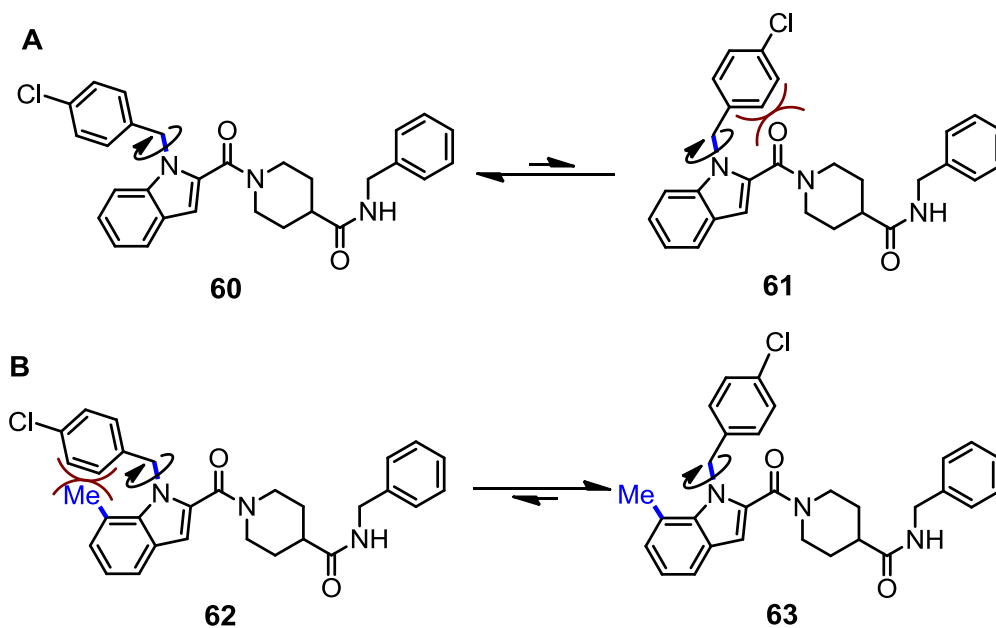
Figure 15. Steric Approach to Conformational Restriction.

Installation of sterically restricting functionality is most often the simplest approach of the three tactics and retains some degree of flexibility. In the case of lead **CCG-102516**, the *N*-benzyl encounters minimal resistance to rotation from the small indole hydrogens and would be expected to adopt a configuration that minimizes steric clashes with the amide carbonyl (**Figure 15 A**). On the other hand, methylation of the indole 7-position hinders its ability to rotate over this region and would be expected to

greatly limit the available conformers, inducing a conformation (**63**) that is distinct from

61 (**Figure 15 B**).
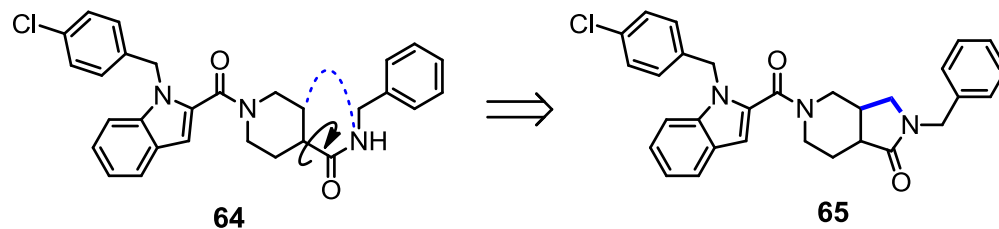
Figure 16. Tethering Approach to Conformational Restriction.

The second tactic, bridging of nonadjacent atoms, is a much more restrictive approach. For example, the distal amide of **CCG-102516** is free to rotate about piperidine-amide C-C bind (**Figure 16**), but inclusion of a bridging methylene between the piperidine 3-position and the amide nitrogen provides a compound with an immobilized carboxamide conformer.
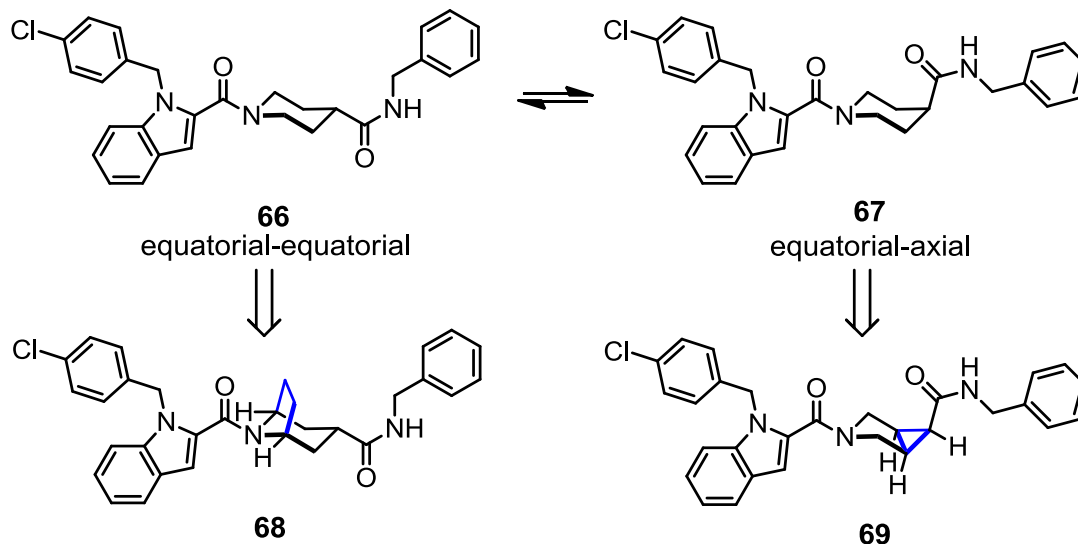
Figure 17. Ring-Locking Approach to Conformational Restriction.

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The third approach, the locking or steric restriction of ring conformers, is similar to that of the previous tactic in that it most frequently involves bridging of nonadjacent and (often) transannular atoms (**Figure 17**). The lead compound **CCG-102516** features a 1,4-disubstituted piperidine, and due to the meso C2 symmetry about the ring, there predominantly exist two major conformers: equatorial-equatorial (**66**) and axial-equatorial (**67**). Either of these can be conveniently locked. An ethylene bridge across the piperidine 2- and 6-positions can only axial-axial (**68**), resulting in a locked equatorial-equatorial relationship between the piperidine substituents. Similarly, preparation of the fused cyclopropane/pyrrolidine (**69**) allows access to the axial-equatorial analog, with minimal impact on molecular weight.

SAR

Initial SAR focused on the restricting the indole and benzamide portions of the lead **CCG-102516**. Biological results for indole core analogs appear in **Table 11**. Utilizing the steric approach to conformational restriction, two compounds were designed on the notion that strategically placed methyl groups would impede bond rotations of interest while only contributing a small increase in molecular weight. Both the 7-methylindole **CCG-206447** and the 3-methylindole **CCG-205431** possessed substantially reduced potency, indicating that the methyl groups may in fact be blocking access to the active conformations.

Table 11. WEEV Replicon Data for Methylindole Analogs.
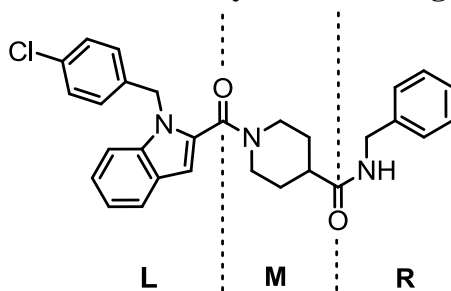
CCG No.	L	M-R	IC$_{50}$ (μM)[a]	CC$_{50}$ (μM)[b]
102516†			15.6 ± 1.8	>100
205431			>100	>100
206447			61 ± 39	>100
206485			17.1 ± 1.3	>100

[a]Inhibition of luciferase expression in WEEV replicon assay. [b]Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. †Synthesized by Janice Sindac.

Table 12. WEEV Replicon Data for Rigid Benzamide Analogs.
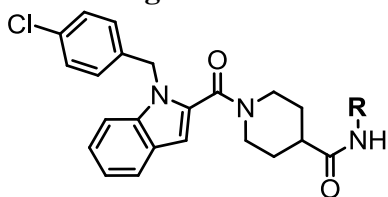
CCG No.	R	IC$_{50}$ (μM)^a	CC$_{50}$ (μM)^b
102516†		15.6 ±1.8	>100
205470		>100	>100
205471		>100	>100
205472		>100	>100
205473		>100	>100
205474		>100	>100
206328		>100	>100
206382		0.53 ± 0.04	>100
206549		1.7 ± 0.1	>100
206550		7.1 ± 0.9	>100
206397†		15.2 ± 4.7	62 ± 13
206587		>100	>50
206382		0.53 ±0.04	>100

^{a}Inhibition of luciferase expression in WEEV replicon assay. ^{b}Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. †Synthesized by Janice Sindac.

Restricted of the Terminal Carboxamide. We then synthesized and evaluated analogs of the benzamide featuring phenylpyrrolidines, isoindolines, indolines, and indane substructures (**Table 12**) and a bicyclic compound **CCG-206485** (**Table 11**), but the majority of analogs exhibited no activity, which was not entirely undesirable. Uniformly active compounds would indicate a non-selective binding site – one to which binding optimization would be difficult. Excitingly, one compound (**CCG-206382**) possessed significantly better activity than others in this series, and constituted the most active compound in the analog arsenal at that time. This result was most likely not the product of increased lipophilicity or chain extension because analogs of similar logP (**CCG-206550**) and length (**CCG-206587**) have higher IC_{50} values (**Table 12**). Interestingly, the racemic methyl analog of **CCG-206382** (**CCG-206549**) was also active, though just over two-fold less so than its more potent relative. Curiously, **CCG-206550** was similarly active to the lead **CCG-102516**, but **CCG-205472** was completely inactive despite being shorter by only a single methylene unit. Overall, the results in **Table 12** suggest that the amide binding pocket of the unknown molecular target is very well defined and discriminating.

Indane SAR. Due to the superb *in vitro* potency of **CCG-206382** in the WEEV replicon assay, the indane moiety was incorporated into other WEEV inhibitor analogs, including those of the low molecular weight class (**Chapter III**). As previously discussed in that chapter, Janice Sindac had discovered the equally potent compound **CCG-205432** bearing a pyridine in place of the indane. Curiously, related analogs differing only by the indane or pyridine functionality did not possess similar activities (**Table 13**); in some cases, the IC_{50} values differed in excess of one hundred-fold (*e.g.* **CCG-206381**

compared to **CCG-208914**, and **CCG-209021** compared to **CCG-209020**.) The indane compounds also behaved differently than their pyridine counterparts in the replicon assay. For example, an IC_{50} value for **CCG-209793** could not be deduced because the concentration-inhibition plot was uncharacteristically linear, and more than one compound's plot oddly plateaued in a manner inconsistent with every other analog and class synthesized to-date. These results questioned the validity of **CCG-206382**'s activity, but our collaborators found no evidence of direct luciferase inhibition. The compound was consequently promoted for further studies to evaluate its true efficacy against live virus.

Table 13. WEEV Replicon Data Comparisons of Analogs Based on CCG-205432 and CCG-206382.
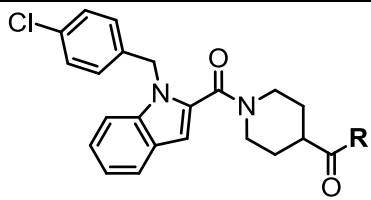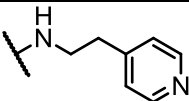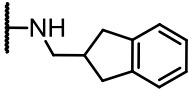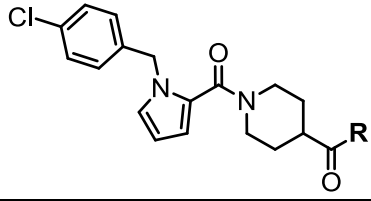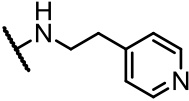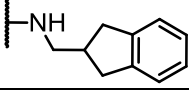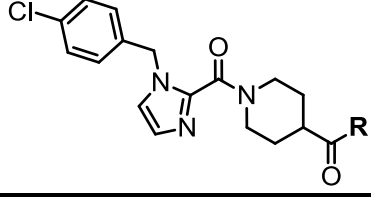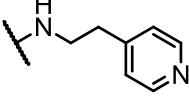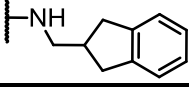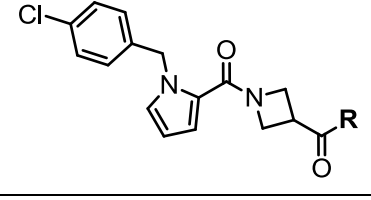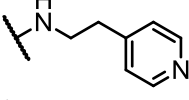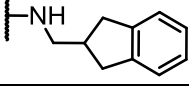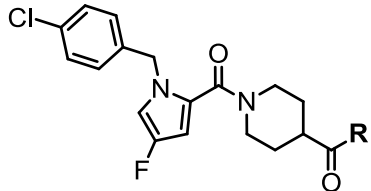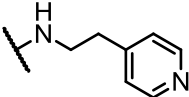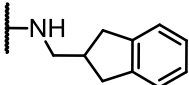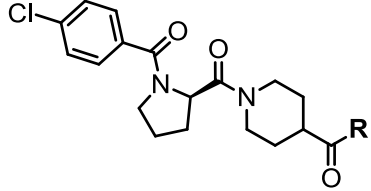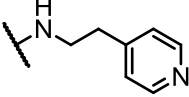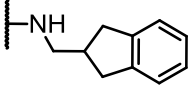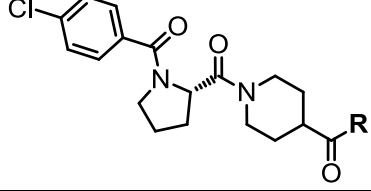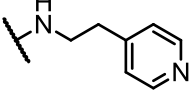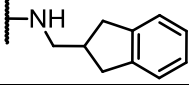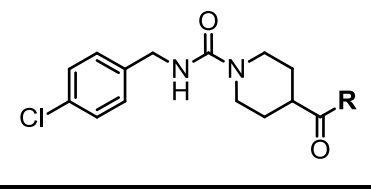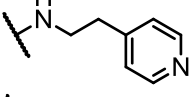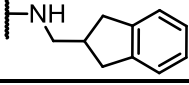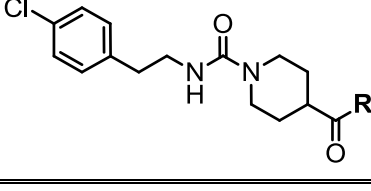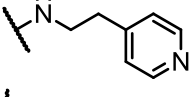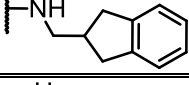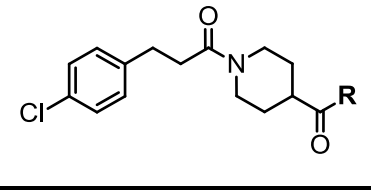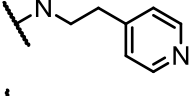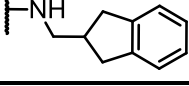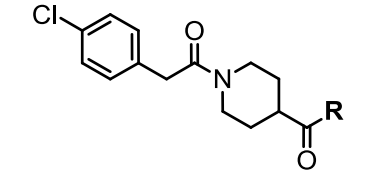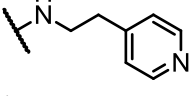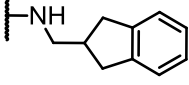
	CCG #	R	$IC_{50} (\mu M)^a$	$CC_{50} (\mu M)^b$
	205432†		0.53 ± 0.08	65.0
	206382		0.53 ± 0.04	>100
	206381		0.68 ± 0.04	>100
	208914		61.8^c	70.3
	208916		>100	>100
	208913		>100	>100
	208915		22.0 ± 8	>100
	208793		$CNBD^d$	>100

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Table 13. Continued.

	CCG #	R	IC_{50} $(\mu M)^a$	CC_{50} $(\mu M)^b$
	209021		0.93 ± 0.2	8.0
	209020		>100	>100
	211756		>100	>100
	211753		>100	>100
	211754		>100	>100
	211755		>100	>100
	208825		60 ± 5	>100
	208826		>100	>100
	208827		41 ± 5	>100
	208828		>100	>100
	208846		>100	>100
	208847		>100	>100
	208848		>100	>100
	208849		33 ± 15	>100

[a]Inhibition of luciferase expression in WEEV replicon assay. [b]Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. [c]Value is n=1 experiment. [d]Could not be determined. †Synthesized by Janice Sindac.

Antiviral Activity. When cells infected with WEEV or NSV (neuro-adapted sinbis virus) were treated with the pyridine analog **CCG-205432**, viral titer was reduced by nearly two log units, and cell viability was significantly improved (**Figure 18**). However, the indane analog **CCG-206382** (despite being equipotent in the WEEV replicon assay) neither decreased viral titer nor improved cell viability relative to the negative control, suggesting that the compound was truly not active. Analogs featuring the indane moiety were subsequently dropped.
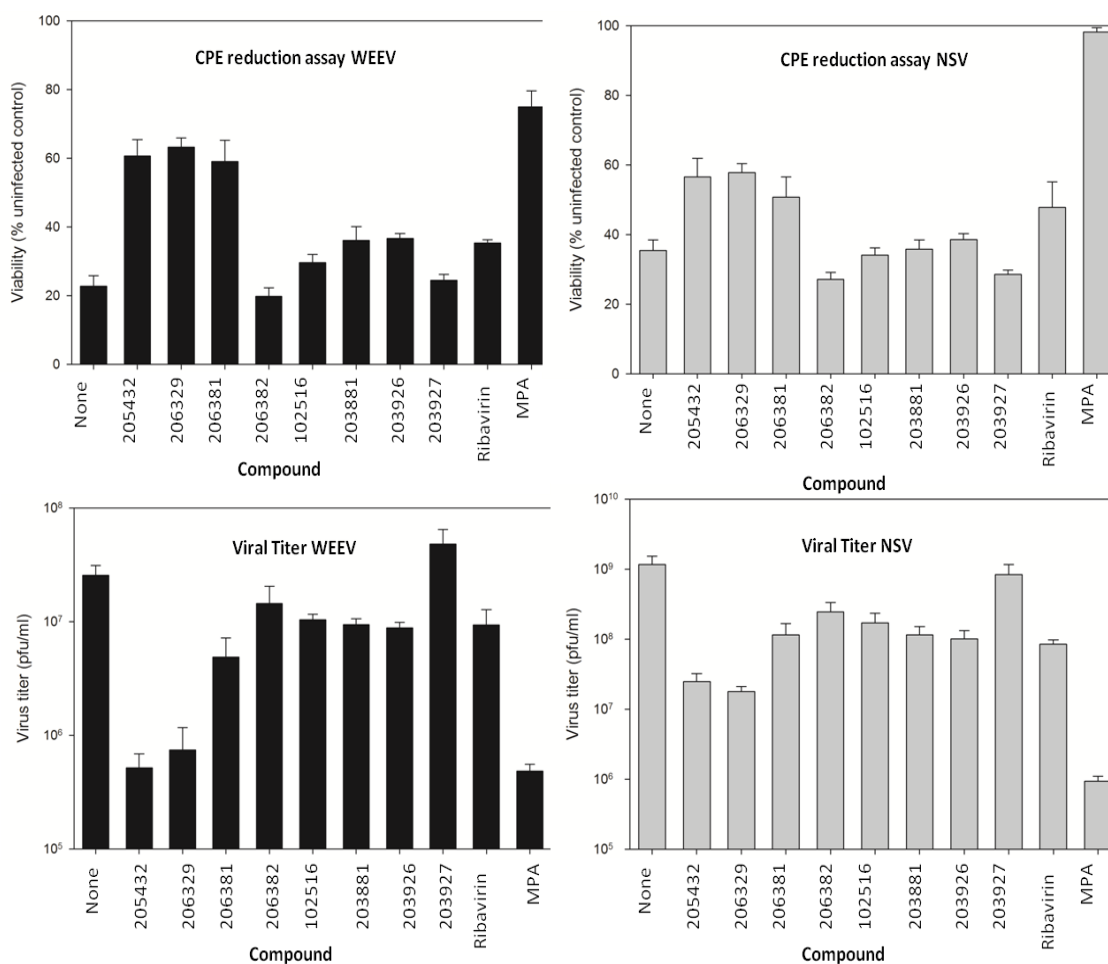
Figure 18. Activity of Various Analogs Against Live Virus.
Data from the David Miller Lab.
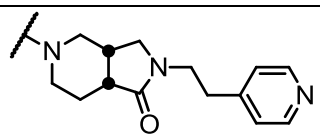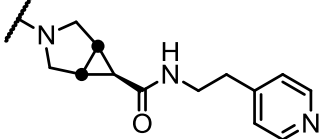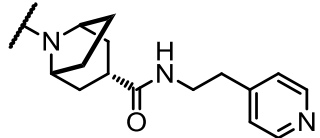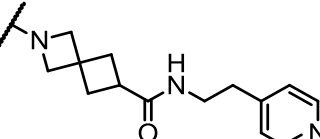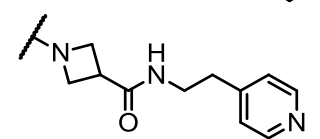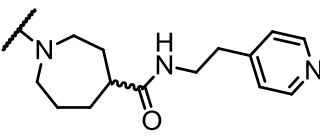
Piperidine Amide SAR. Having determined the terminal carboxamide portion of the analogs to be a challenging site for conformational restriction, our attention was turned towards the central piperidine ring. As in **Chapter III** (low molecular weight series), **CCG-205432** was employed as the lead due to its potency and demonstrable activity against live virus, and thus this series features the right-hand pyridylethyl amide. Biological results appear in **Table 14**. In **Chapter II** (reduced TPSA series), it was noted that there appeared to be a limited tolerance for TPSA reduction, perhaps because certain structure-activity requirements were not met. It was noted that compounds like **CCG-211826** (**Table 2**), despite being structurally similar to potent leads, lacked the right-hand secondary amide. To evaluate the postulated importance of this functionality, four compounds were prepared (**Table 14**). Activities of the inverse amide **CCG-2120532** and the shifted amide **CCG-224001** are over 30-fold less than **CCG-205432**, confirming that the amide is important to the unknown pharmacophore. In particular, both of these analogs retain the amide carbonyl in the same relative location, but differ in the location of the amide nitrogen, which indicates that the position of the carbonyl is apparently more important than the NH. The amide carbonyl may be engaged as a hydrogen-bond acceptor, and, due to the dependence on the acceptor-donor angle, translocation of the carbonyl by one methylene unit may be sufficient to deprive the molecule of a productive hydrogen bond in the unknown target, accounting for the diminished activity. Furthermore, complete excision of the amide (*e.g. E*- and *Z*-alkenes **CCG-211823** and **CCG-211825**) returned analogs with no activity, complementing this hypothesis.

Table 14. WEEV Replicon Data for Central Piperidine Analogs.
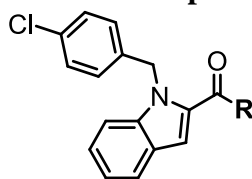
CCG No.	R	IC$_{50}$ (µM)^a	CC$_{50}$ (µM)^b
205432†		0.53 ± 0.08	65.0
212052		17.6^c	66.7^c
224001		24.7 ± 3.5	77.3
211823		>50	>50
211825		>50	>50
212390		24.4 ± 4.0	78.0
211822		3.9 ± 0.4	74.2
211829		>50	>50
211761†		1.3 ± 0.4	63.2

Table 14. Continued.

CCG No.	R	IC_{50} $(\mu M)^{a}$	CC_{50} $(\mu M)^{b}$
212394		24.4 ± 6.3	61.0
222980		24.6 ± 1.6	>100
224000		0.96 ± 0.06	50.9
224220		10.9 ± 0.6	56.5
224002		73.1 ± 29.1	88.1
222661		0.49 ± 0.09	41.1

[a]Inhibition of luciferase expression in WEEV replicon assay. [b]Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. [c]Value is n=1 experiment. †Synthesized by Janice Sindac.

Core Piperidine SAR. The significantly reduced activity of the urea analog **CCG-212390** and eneamide **CCG-211822** are also consistent with the postulated importance of the secondary amide (**Table 14**). Both analogs were built to rigidify and flatten the core ring, and such modifications may have changed the projection of the amide carbonyl. The bicyclic analog **CCG-212394** may display the amide in a similarly nonproductive manner, especially in light of the better activity of the very similar (but

less restricted) analog **CCG-211761**. The α-methyl analog **CCG-211829** was much less active, presumably because the methyl was either impeding favorable chair conformations or occluding the amide carbonyl. The *exo*-bicycle **CCG-222980** was also less potent than the lead. Interestingly, the ethylene-bridged piperidine **CCG-224000** was a mere two-fold less potent than **CCG-205432**, suggesting that the locked equatorial-equatorial relationship between the 1- and 4-substituents may be indicative of the unknown bioactive conformation.

The importance of length to the activity of WEEV inhibitors was discussed in **Chapter II** and **Chapter III**, and, unsurprisingly, the azetidine-containing **CCG-224002** was less active than its piperidine-containing counterpart **CCG-205432** (**Table 14**). To determine the effect of larger rings on activity, the azapane analog **CGG-222661** was synthesized and tested. Interestingly, this compound was equally potent with the lead despite being less rigid and possessing an offset right-hand amide. Furthermore, the active enantiomer could potentially be two-fold more active than the racemate, marking this compound as the most active to-date in the WEEV replicon assay.

Synthesis

Restricted analogs based upon the benzylamide lead **CCG-102516** were generally prepared through amide coupling of various amines with the core carboxylic acid **71**, which was itself accessed via coupling of the indole-2-carboxylic acid **4a** with ethyl isonipecotate **26** followed by saponification (**Scheme 18**).

Scheme 18. General Preparation of Rigid Benzamide Analogs.[a]

4a   a   **70**   b

71   c   **72**
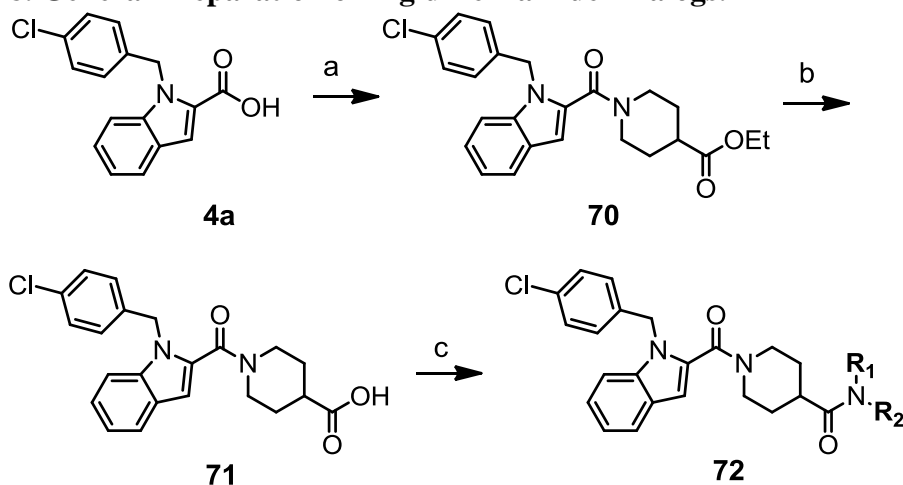
[a]Reagents and conditions: (a) **26**, EDC·HCl, HOBT, TEA, DCM, RT; (b) 10% aq. NaOH, EtOH, RT; (c) **HNR$_1$R$_2$**, EDC·HCl, HOBT, TEA, DCM, RT.

Scheme 19. Preparation of 2-(aminomethyl)indane 75.[a]

73   a   **74**   b   **75**
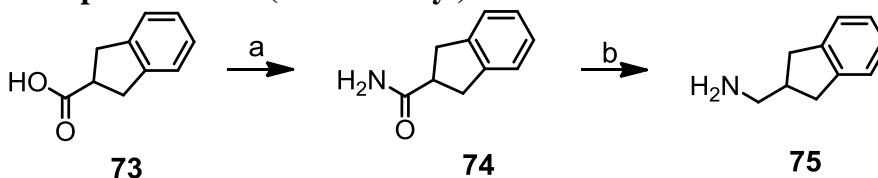
[a]Reagents and conditions: (a) HMDS, HATU, DIPEA, DMF, rt, 20 h; (b) LiAlH$_4$, THF, RT, 8 h.

2-(Aminomethyl)indane **75** (**Scheme 19**, for the synthesis of **CCG-206382**) was accessed first through amidation with the 2-indane carboxylic acid **73** and HMDS, after which aqueous workup conditions resulted in hydrolytic degradation of the silyl functionality to reveal the primary carboxamide **74**. This procedure was found to be more efficient than direct aminolysis of the carboxylic acid. Reduction of the carboxamide with LAH provided the desired amine **75**, which was coupled with indole carboxylic acid **71** as described in **Scheme 18**. Racemic 2-(2-aminoethyl)indane **78** was prepared first

through double displacement of dibromoxylene **76** with acetylacetate followed by reductive amination of the ketone **77** (**Scheme 20**).

Scheme 20. Preparation of Racemic 2-(2-Aminoethyl)indane.[a]

| | **76** | | **77** | | **78** |
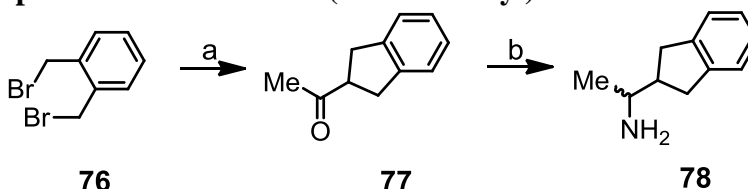
[a]Reagents and conditions: (a) ACAC, NaH, DMF; (b) 7 M NH$_3$/MeOH, NaBH$_4$, RT, 20 h.

The 7- and 3-methyl indole analogs were generally prepared through classical Fischer indolization (**Scheme 21** and **Scheme 22**). The appropriate anilines (*o*-toluidine **79** and aniline **84**) were transformed into their respective diazonium salts and reduced to hydrazines through the action of tin(II) chloride.[138] The hydrazines were then allowed to undergo Fischer indolization with the necessary β-ketoester **80** or **85**, but only the 7-methyl indole required a transition metal Lewis acid catalyst to facilitate cyclization to indole **81**. Overall, this route was found to be more expedient than the reported two-step Japp-Klingemann/Fischer indolization approach[139] to the indole scaffold. Alkylation of the indole *N*1-position with 4-chlorobenzyl chloride **2a** was found to be slow for the 7-methyl analog **81**, presumably due to steric hindrance of the nucleophile. Subsequent saponification and amide coupling with benzylamine yielded the desired methyl indole analogs **CCG-205431** and **CCG-206447**.

Scheme 21. Preparation of 7-Methylindole CCG-206447.[a]
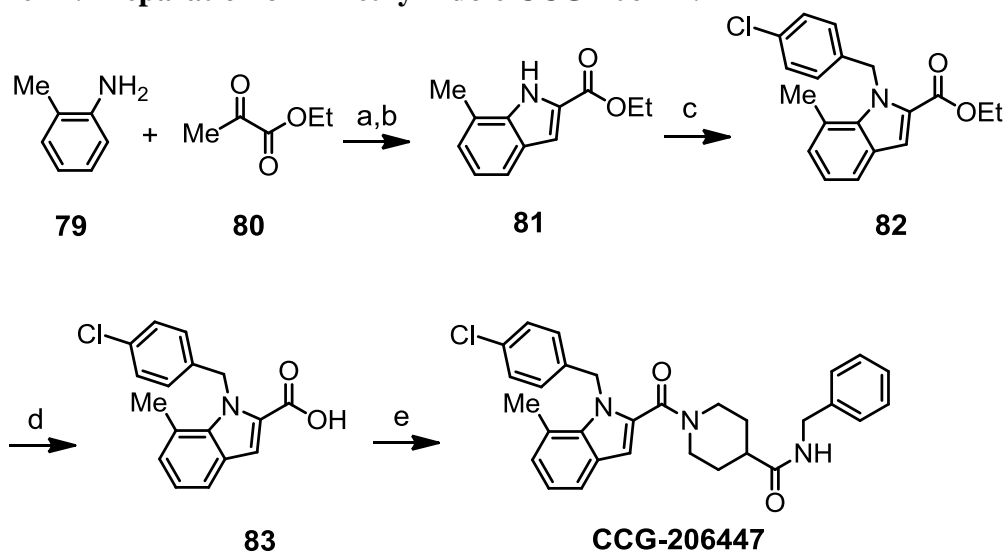
[a]Reagents and conditions: (a) **79**, NaNO$_2$, conc. HCl, 0 °C, 30 min; *then* SnCl$_2$, 4 °C, 11 h, 56%; (b) **80**, ZnCl$_2$, TsOH, EtOH, reflux, 8 h; (c) **2a**, K$_2$CO$_3$, DMF, 60 °C, 3 d, 46%; (d) 10% aq. KOH, EtOH, 70 °C, 4 h, 94%; (e) *N*-benzylpiperidine-4-carboxamide, EDC·HCl, HOBT, DIPEA, DMF, RT, 18 h, 47%.

Scheme 22. Preparation of 3-Methylindole CCG-205431.[a]
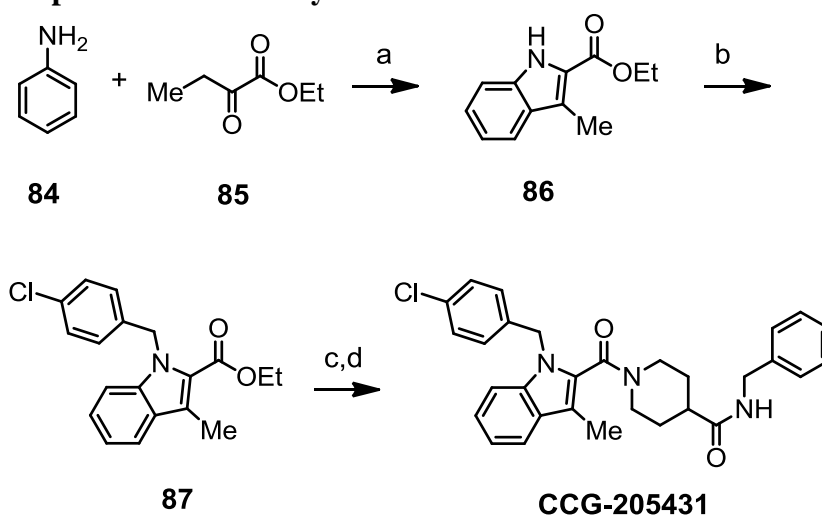
[a]Reagents and conditions: (a) PhNH$_2$, NaNO$_2$, conc. HCl, 0 °C, 15 min; *then* SnCl$_2$, 0 °C, 24 h; *then* **85**, EtOH, TsOH, reflux, 12 h, 65%; (b) **2a**, K$_2$CO$_3$, DMF, 60 °C, 12 h, 72%; (c) THF, 5 M aq. NaOH, 55 °C, 18 h, 96%; (d) *N*-benzylpiperidine-4-carboxamide hydrochloride, HATU, DIPEA, DMF, RT, 20 h, 51%.

The pyrrolopiperidine analogs **CCG-206485** and **CCG-212394** were accessed first through the partial reduction of the imide **88** to the amide **89** as reported by Dugar et al.[140] Presumably, selectivity arises from chelation-directed reduction of the carbonyl situated on the same side of the molecule as the azine nitrogen.[141] Complete reduction of the pyridine gave the piperidine **90**, and amidation with this and the indole carboxylic acid **4a** provided intermediate **91**, which was alkylated with the requisite electrophiles (**Scheme 23**).

Scheme 23. Preparation of pyrrolidopiperidine Analogs CCG-206485 and CCG-212394.[a]

88   89   90

91   R = Bn, CCG-206485
R = pyr, CCG-212394

"Bn"   "pyr"
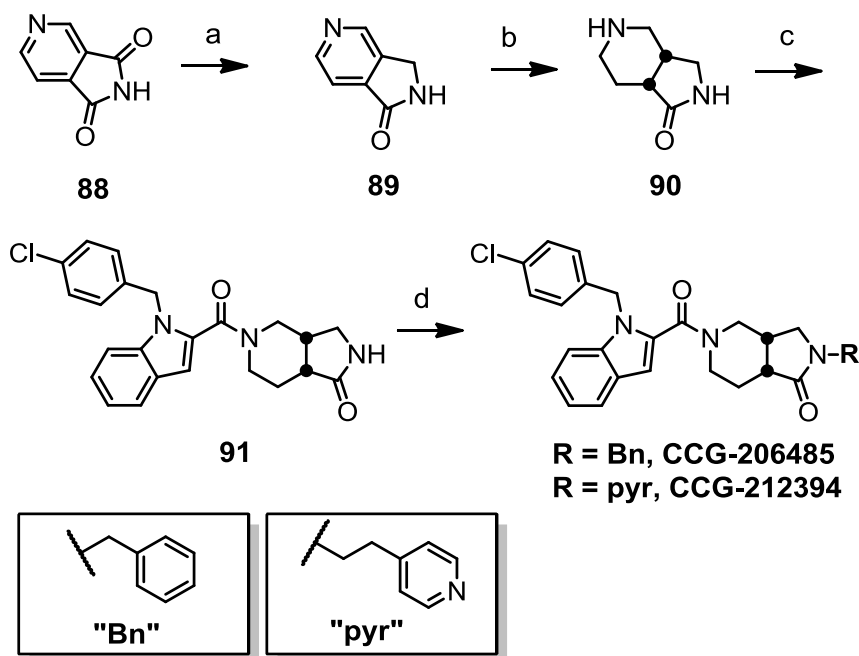
[a]Reagents and conditions: (a) Zn^0, AcOH, 6 h, 115 °C, 38%; (b) PtO_2, 50 psi H_2, AcOH, RT, 2 d; (c) **4a**, EDC·HCl, HOBT, DIPEA, DCM, RT, 12 h, 56%; (d) $Ar(CH_2)_nBr$, NaH, DMF, RT, 18 h, 72%.

Scheme 24. Preparation of Inverse Amide CCG-212052.[a]

CCG-212052
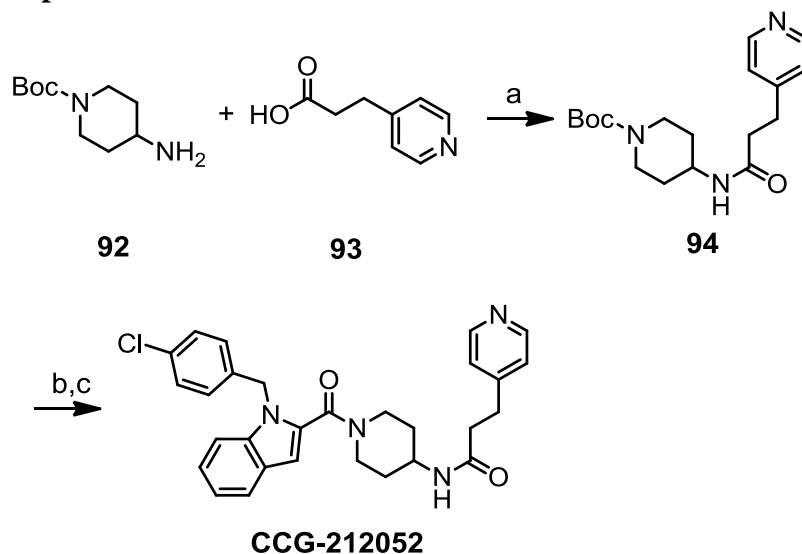
[a]Reagents and conditions: (a) EDC·HCl, HOBT, TEA, DCM, RT, 30 h, 71%; (b) HCl, dioxane, RT, 1 h, 100%; (c) **4a**, EDC·HCl, HOBT, TEA, DCM, RT, 22 h, 61%.

Scheme 25. Preparation of Shifted Amide CCG-224001.[a]

CCG-224001
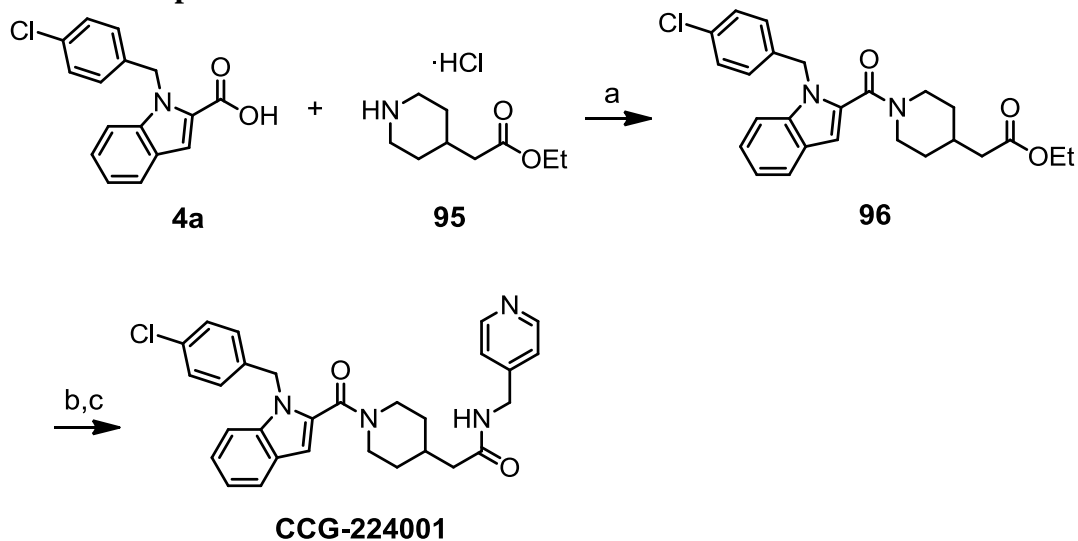
[a]Reagents and conditions: (a) EDC·HCl, HOBT, TEA, DCM, RT; (b) KOH, H₂O, EtOH, RT; (c) 4-(aminomethyl)pyridine, EDC·HCl, HOBT, TEA, DCM, RT.

The inverse amide **CCG-212052** was prepared through the amide coupling of *N*1-protected 4-aminopiperidine **92** and 3-(4-pyridyl)propanoic acid **93**. Subsequent

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deprotection and coupling with indole carboxylic acid **4a** furnished the desired inverse amide **CCG-212052** (**Scheme 24**).

To prepare the shifted amide analog **CCG-224001**, the commercially available amino ester **95** was first coupled with the indole carboxylic acid **4a**. The ester of the resulting intermediate was then hydrolyzed and coupled with 4-(aminomethyl)pyridine to provide the final compound (**Scheme 25**).

It was anticipated that a Wittig reaction would ultimately provide the key (Z)-alkene feature of analog **CCG-211829**, so the requisite phosphonium bromide **98** was prepared from 4-(3-hydroxypropyl)pyridine **97**.[142] This was then deprotonated by *n*BuLi to provide the ylide, which was subsequently permitted to react with the piperdinyl aldehyde **8** to provide the desired alkene **99a** with >50 Z:E selectivity. Deprotection and subsequent coupling with the indole carboxylic acid **4a** gave the (Z)-alkene analog **CCG-211829** (**Scheme 26**).

A large number of highly E-selective olefinations exist,[143] but because the phosphonium bromide salt was already in hand, the Schlosser modification of the Wittig reaction was employed in order to access (E)-alkene analog **CCG-211822** (**Scheme 26**). In this procedure, the intermediate oxaphosphetane is deprotonated by PhLi – uniquely suited to deprotontion of sterically shielded protons[144] – and allowed to equilibrate to the more thermodynamically favorable *trans*-betaine carbanion before being quenched with *t*BuOH. In this way, the desired E stereochemistry was quickly accessed to provide the (E)-alkene **99b**. Deprotection and amide coupling with indole carboxylic acid **4a** yielded the desired (E)-alkene analog **CCG-211822**.

Scheme 26. Preparation of (E)- and (Z)-alkene Analogs.[a]

97   **98**   **99a (Z)**

99b (E)

CCG-211829 (Z)

CCG-211822 (E)
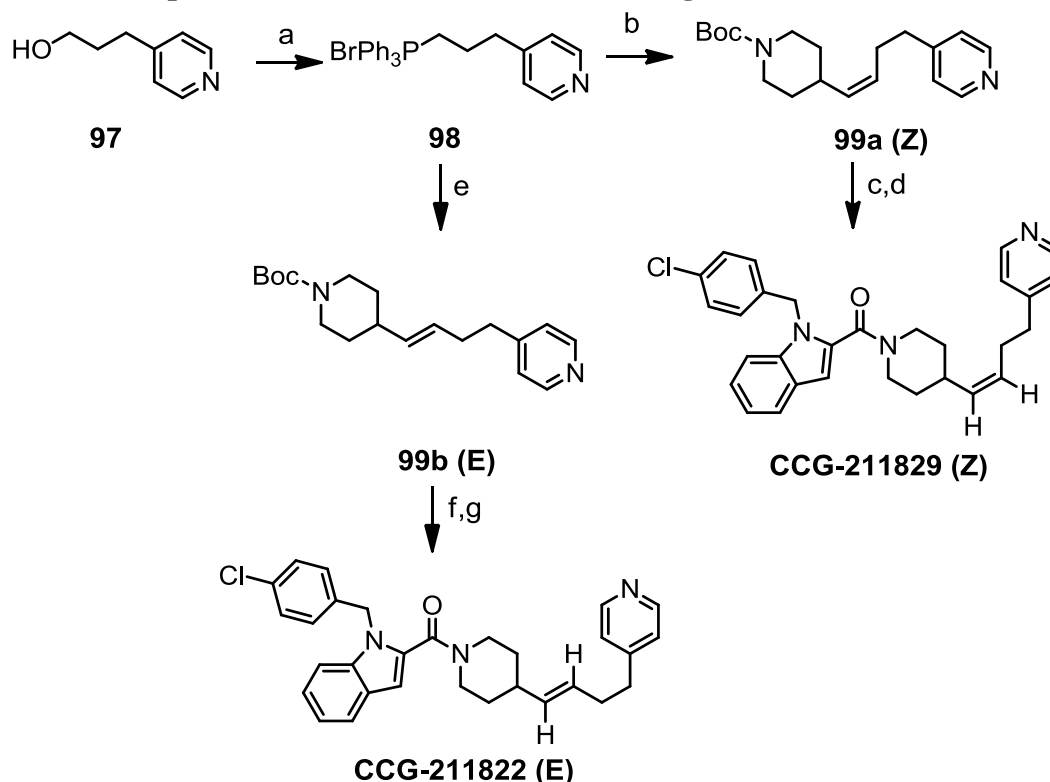
[a]Reagents and conditions: (a) Conc. HBr, PPh$_3$, tol, reflux, 11 h, 34%; (b) *n*-BuLi, THF, RT; *then* **8**, RT, 12 h, 52%; (c) 4 M HCl/1,4-dioxane, RT, 1 h; (d) 1-(4-chlorobenzyl)-1H-indole-2-carboxylic acid, EDC·HCl, HOBT, TEA, DCM, RT, 14 h, 56% (2 steps); (e) PhLi, LiBr, THF, RT, 20 min; *then* **8**, THF, -78 °C, 30 min; *then* PhLi, -78 °C, 30 min; *then t*-BuOH, THF, -78 °C–RT, 36 h, 41%; (f) 4 M HCl/1,4-dioxane, RT, 1 h; (g) 1-(4-chlorobenzyl)-1H-indole-2-carboxylic acid, EDC·HCl, HOBT, TEA, DCM, RT, 14 h, 60% (2 steps).

The piperazinyl urea analog **CCG-212390** was prepared by treating boc-piperazine **100** with phosgene and 4-(2-aminoethyl)pyridine **27** (**Scheme 27**). Order of addition was critical; if **27** was treated with phosgene, the intermediate *N*-acyl chloride would rapidly react with the pyridine nitrogen and no productive chemistry would arise. Thus, the boc-piperazine **100** was treated first with phosgene, and the pyridyl amine **27** was added last. Once the urea was in hand, deprotection and coupling with indole carboxylic acid **4a** provided the desired urea analog **CCG-212390**.

Scheme 27. Preparation of Urea Analog CCG-212390.[a]
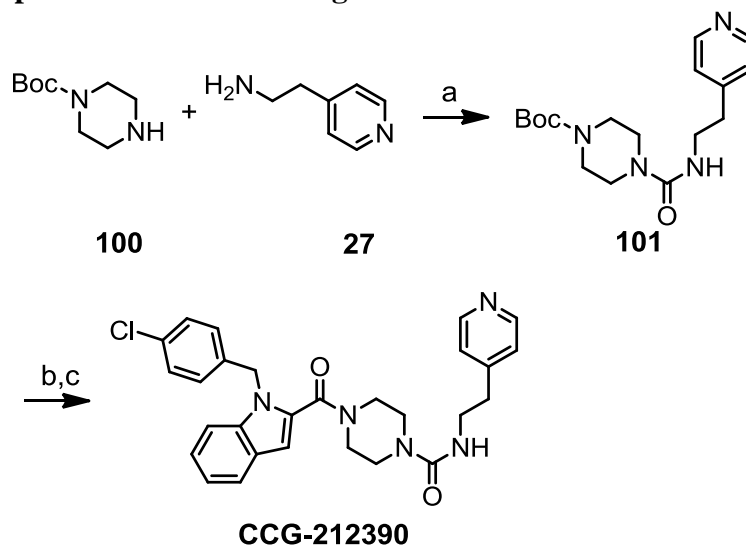
[a]Reagents and conditions: (a) COCl$_2$, TEA, tol, DCM, 0 °C, 1 h, 81%; (b) HCl, dioxane, RT, 1 h, 100%; (c) **4a**, EDC·HCl, HOBT, TEA, DCM, RT,

Initial attempts to access the tetrahydropyridine core of **CCG-211823** proceeded through the partial reduction of the formyl pyridinium salt of ethyl isonicotinate with NaBH$_4$,[145, 146] but this method was found to be poor yielding and complicated by side products. The selenoxide elimination was then identified as a viable alternative, shown in **Scheme 28**.[147, 148] Treatment of protected ethyl isonipecotate **102** with LDA and subsequent addition of phenylselenyl chloride provided the phenylselenide **103**. The compound could undergo the oxidation-elimination at this point, but it was found that carrying the selenyl functionality through the first amidation aided purification efforts. In this way, phenylselenide intermediate **105** was oxidized with *m*CPBA under cold conditions and basified to prevent Bronsted-acid catalyzed hydrolytic decomposition via a seleno-Pummerer pathway,[149] and the resulting tetrahydropyridine ethyl ester **106** was

saponified then amidated with 4-(2-aminoethyl)pyridine to give the desired analog **CCG-211823**.

Scheme 28. Preparation of Tetrahydropyridine Analog CCG-211823.[a]
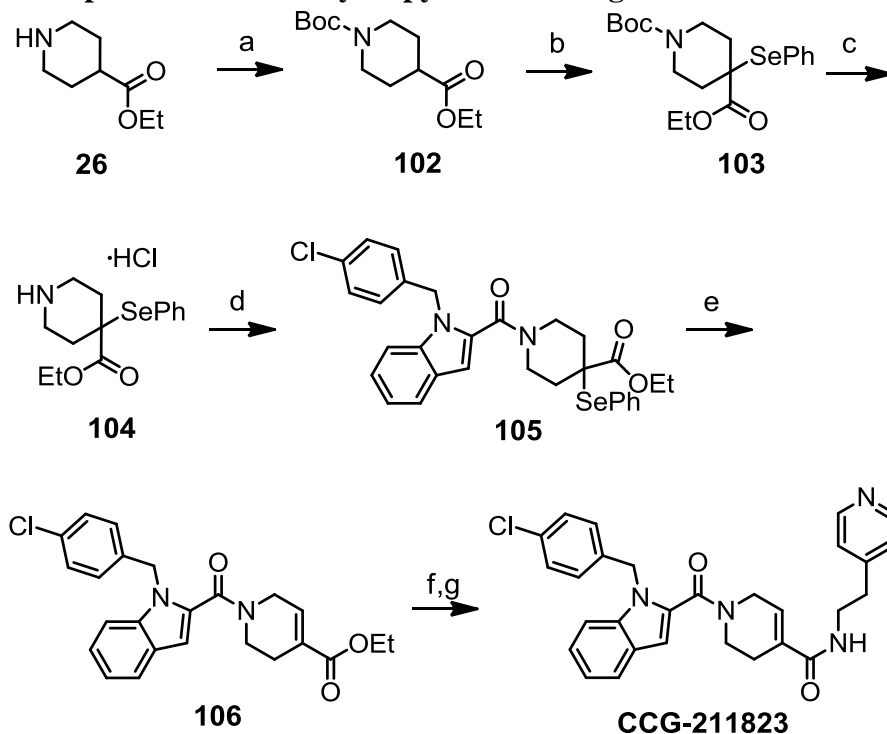
[a]Reagents and conditions: (a) Boc$_2$O, Na$_2$CO$_3$, THF, H$_2$O, reflux, 86%; (b) *n*-BuLi, DIPA, -78 °C, 15 min; *then* PhSeCl, THF, -78 °C–RT, 3 h, 58%; (c) 4 M HCl/1,4-dioxane, RT, 2 d, 100%; (d) **4a**, EDC·HCl, HOBT, TEA, DCM, RT, overnight, 63%; (e) *m*CPBA, DCM, -78 °C, 30 min; *then* TEA, DCM, -78 °C; *then* -78 °C–RT, 1 h, 82%; (f) 10% aq. NaOH, EtOH, RT, 95%; (g) **27**, EDC·HCl, HOBT, TEA, DCM, RT, 91%.

In a manner similar to the preparation of the tetrahydropyridine analog **CCG-211823**, Boc-protected ethyl isonipecotate **102** was alkylated with methyl iodide after treatment with LDA to provide the methylpiperidine **107**. This piperidine was deprotected and coupled with indole carboxylic acid **4a**, and the ethyl ester **109** was

saponified and coupled with **27** to give the desired methyl analog **CCG-211829** (**Scheme 29**).

Scheme 29. Preparation of Methylpiperidine Analog CCG-211829.[a]
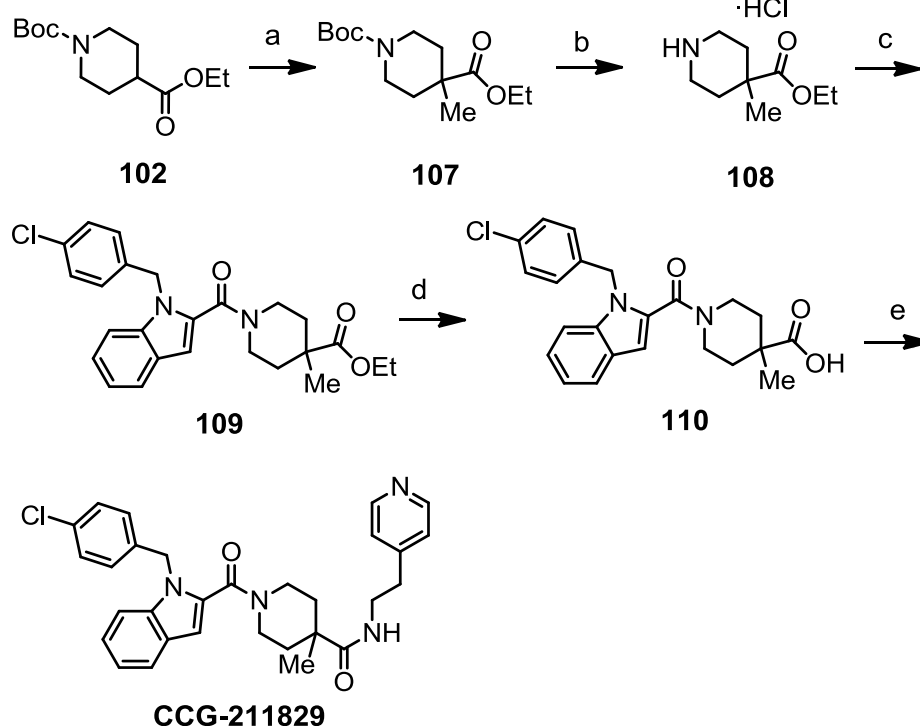
[a]Reagents and conditions: (a) *n*BuLi, DIPA, THF, -78 °C; *then* MeI, THF, -78 °C; (b) 4 M HCl, dioxane, RT, 1 h; (c) **4a**, EDC·HCl, HOBT, TEA, DCM, RT; (d) **27**, EDC·HCl, HOBT, TEA, DCM, RT.

The key cyclopropane of the exo-bicyclic analog **CCG-222980** was installed by a method previously reported.[150, 151] An electron-deficient carbenoid was generated from ethyl diazoacetate and treated with Cbz-protected 2,5-dihydropyrrole **111** over several days. This yielded a mixture of unreacted starting material and two diastereomers of the product **112**, enriched in the exo-isomer, which was taken forward after tedious separation and deprotected via hydrogenolysis and coupled with the indole carboxylic

acid **4a**. Treatment with KOTMS hydrolyzed the ester to the potassium salt, which was subsequently coupled with amine **27** to yield the final analog (**Scheme 30**).

Scheme 30. Preparation of Exo Bicyclic Analog CCG-222980.[a]

111   (endo)-**112**   (exo)-**112**

113   **CCG-222980**
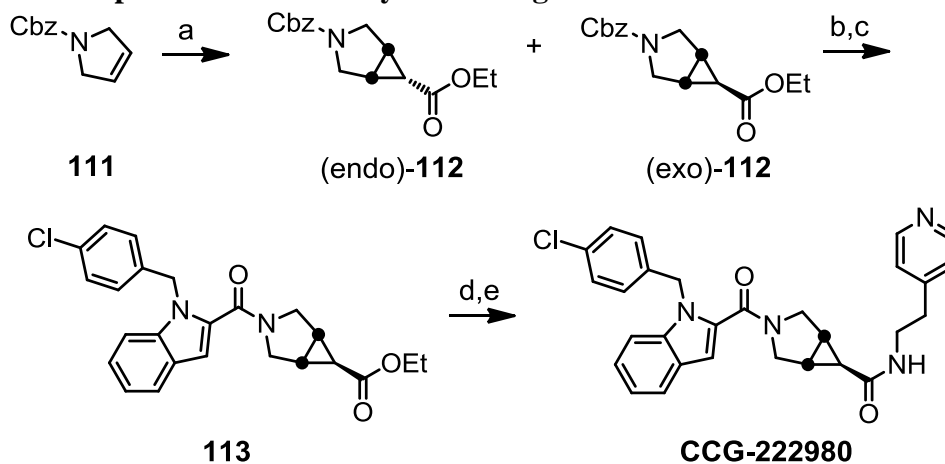
[a]Reagents and conditions: (a) N_2CHCO_2Et, $Rh_2(OAc)_4$, DCM, reflux, 72 h, 3:1 dr; (b) Pd/C, H_2, EtOH, HCl, RT; (c) **4a**, EDC·HCl, HOBT, TEA, DCM, RT; (d) KOTMS, THF, RT; (e) **27**, EDC·HCl, HOBT, TEA, DCM, RT.

Scheme 31. Preparation of Bridged Analog CCG-224000.[a]

114   **115**
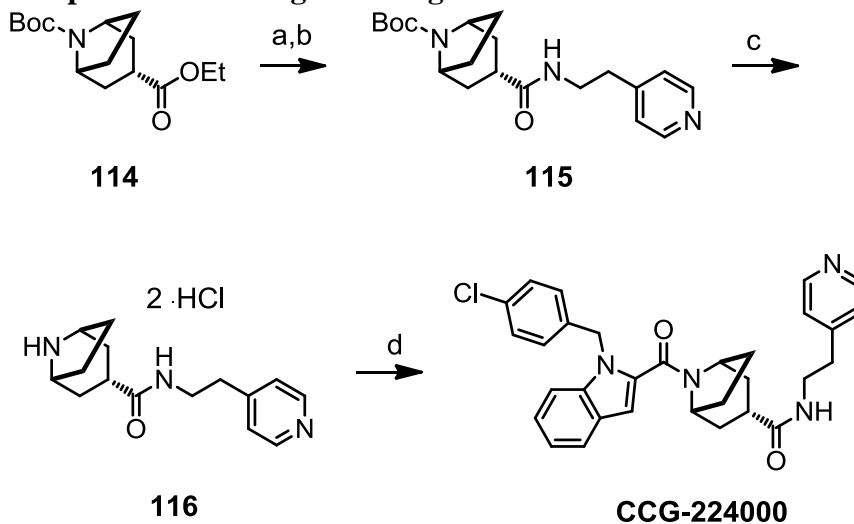
116   **CCG-224000**

[a]Reagents and conditions: (a) KOTMS, THF, RT, 12 h; (b) **27**, EDC·HCl, HOBT, TEA, DCM, RT; (c) HCl, dioxane, RT; (d) **4a**, EDC·HCl, HOBT, TEA, DCM, RT.

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The ethylene-bridged piperidine analog **CCG-224000** was first constructed by hydrolyzing the commercially available protected amino ester **114** and coupling the resulting acid with amine **27** to provide the latent right-hand half **115** of the analog. Deprotection under acidic conditions followed by amide coupling with the indole carboxylic acid **4a** provided the desired analog (**Scheme 31**).

Scheme 32. Preparation of the Spiro Analog CCG-224220.[a]
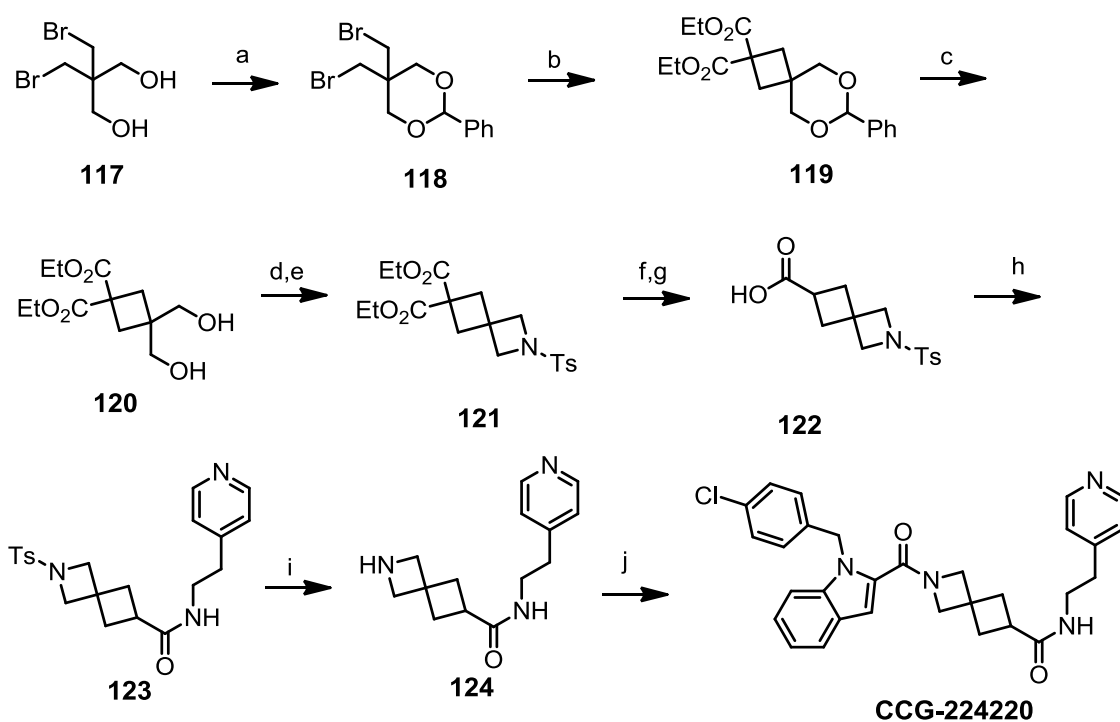
[a]Reagents and conditions: (a) PhCHO, tol, reflux, 2 h, 85%; (b) diethyl malonate, NaH, DMF, reflux, 12 h, 84%; (c) Pd/C, H_2, MeOH, RT, 6 d, 98%; (d) MsCl, TEA, DCM, RT, 3 h; (e) $TsNH_2$, K_2CO_3, DMSO, 90 °C, 5 h, 48% (2 steps); (f) NaOH, H_2O, EtOH, reflux, 2 h, 91%; (g) pyridine, reflux, 12 h, 92%; (h) 4-pyridylethylamine, EDC·HCl, HOBT, TEA, DCM, RT, 12 h, 80%; (i) Na^0, naphthalene, THF, RT, 5 min; (j) **4a**, EDC·HCl, HOBT, TEA, DCM, RT, 14 h, 61% (2 steps).

The spiro core of **CCG-224220** was assembled in manner similar to that reported (**Scheme 32**).[152, 153] The diol **117** was protected as an acetal,[154] and the bromides were

doubly displaced by diethylmalonate under basic conditions to give the diester **119**. The acetal was removed to reveal the alcohols of **120**, which were subsequently mesylated and displaced by tosylamide to give the cyclized spiro product **121**. Ester hydrolysis and thermal decarboxylation furnished the mono-carboxylic acid **122** in good yield, and this was coupled with amine **27** to give the latent right-hand half **123** of the desired final compound. Cleavage of the tosyl proved to be a substantial challenge, however. Various conditions with samarium (II) iodide either failed to remove the tosyl, or reduced and cleaved the amide preferentially.[155, 156] Other reported methods of tosyl deprotection were employed with no success[157, 158] until sodium napthalenide was used,[159] which quickly and cleanly cleaved the S-N bond to reveal the amine **124**, which was then coupled with the indole carboxylic acid **4a** to afford the desired analog **CCG-224220**.

Scheme 33. Preparation of Azetidine-Containing CCG-224002.[a]
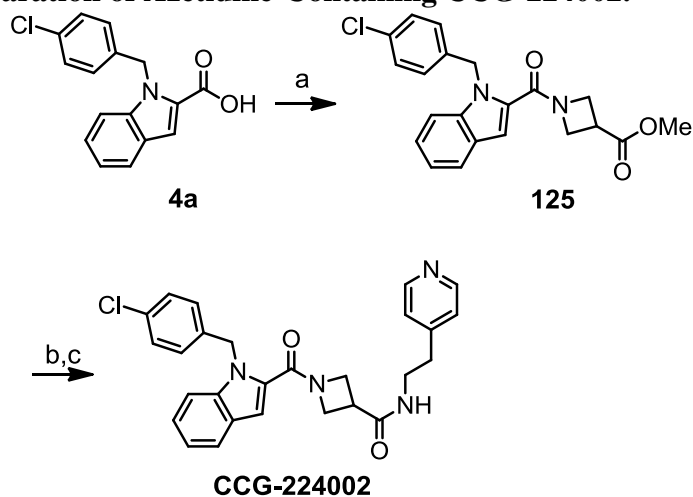
4a   **125**

CCG-224002

[a]Reagents and conditions: (a) methyl 4-azetidinecarboxylate hydrochloride, EDC, HOBT, DIPEA, DCM, rt; (b) 10% aq. NaOH, EtOH; (c) **27**, EDC, HOBT, TEA, DCM, rt.

The azetidine-containing analog **CCG-224002** (**Scheme 33**)

Scheme 33was assembled in a manner similar to that used for construction of the azetidine-containing pyrrole analogs. The indole carboxylic acid **4a** was coupled with the azetidine methyl ester, and the intermediate **125** was saponified and coupled with 4-(2-aminoethyl)pyridine to give the final analog.

Scheme 34. Preparation of Azapane Analog CCG-222661.[a]
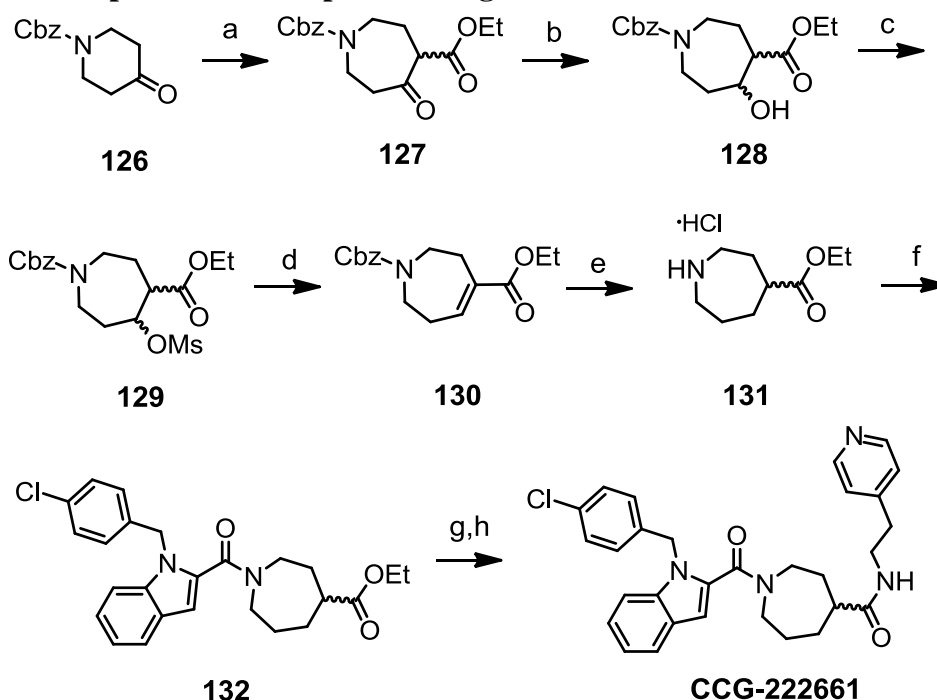
[a]Reagents and conditions: (a) N_2CHCO_2Et, $BF_3 \cdot OEt_2$, Et_2O, -78 °C–RT, 1h; (b) $NaBH_4$, EtOH, 0 °C–RT, 2h; (c) MsCl, TEA, DCM, 0 °C–RT, 6 h; (d) DBU, THF, 80 °C, 1 h; (e) Pd/C, 50 psi H_2, EtOH, HCl, RT, 24 h; (f)) **4a**, EDC·HCl, HOBT, TEA, DCM, RT, 16 h; (g) 10% aq. NaOH, EtOH; (h) **27**, EDC·HCl, HOBT, TEA, DCM, RT.

The core azapane of **CCG-222661** was first accessed by way of ring expansion of *N*-protected 4-piperidone **126** with ethyl diazoacetate and the strong Lewis acid BF_3 (**Scheme 34**).[160] The resulting racemic γ-ketoester **127** was reduced with $NaBH_4$ to the γ-

hydroxy ester and mesylated. The mesylate **129** was then easily eliminated under mild heating with DBU, and the ene ester **130** was exposed to heterogenous reduction conditions to both reduce the alkene and the Cbz protecting group. The product amine **131** was then coupled with indole carboxylic acid **4a**, saponified, and then coupled with amine **27** to give the azapane analog **CCG-222661**.

Conclusion

The primary goal of this chapter was the identification of the unknown inhibitor pharmacophore. To achieve this, conformationally-restricted analogs were prepared and evaluated in the WEEV replicon assay. While the majority of compounds possessed substantially reduced potency, the data pointed towards the importance of the distal amide carbonyl to activity and a potential equatorial-equatorial relationship between the 1,4-substituents of the core piperidine ring. Intriguingly, the azapane **CCG-222661** was identified as our most potent compound to-date, with an $IC_{50} = 0.49 \pm 0.09$ for the racemate, and with an expected IC_{50} up to two-fold better for the active enantiomer. It is hoped this data may be useful for the generation of a computational 3-D pharmacophore model that may be applied to the identification of new alphavirus inhibitor templates via *in silico* screening methods.

Chapter V. **Metabolic Stability**

Rationale

Metabolism is the enzymatic modification of compounds so that an organism may enhance clearance, and is a crucial detoxification mechanism for removing reactive or toxic xenobiotics.[161] Unfortunately, even drugs considered safe may undergo this process. Metabolism dictates the overall biological availability of a compound, and those compounds that undergo rapid metabolism may be depleted before reaching their intended biological target. Metabolic stability is one of the most crucial aspects of early drug development, and instability is a potential killer of lead series.[81]

The majority of metabolism occurs in the liver, where a battery of metabolic enzymes resides, although metabolism may occur elsewhere as well (blood, epithelial intestinal cells, etc.)[162, 163] These enzymes largely consist of a class called the cytochrome p450s, or CYPs, that are responsible for Phase I metabolism, whose purpose is the installation of polar functional groups to improve aqueous solubility to enhance clearance and to provide handles for further Phase II metabolic pathways.[79, 164]

Potential sites of metabolism can be easily predicted using computational models like that shown in **Figure 19**.[165-169] However, while likely, empirical methods are required to confirm such predictions because properties like atom accessibility and overall lipophilicity are important factors. To this end, it was necessary for us to develop

an in-house metabolic stability assay to facilitate our ability to rapidly identify labile

functionalities and design them out of future analogs.
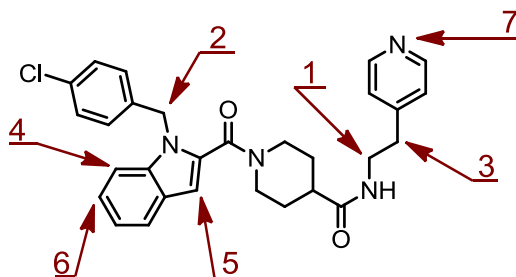
Figure 19. Predicted Sites of Metabolism in CCG-205432, Rank Ordered.
Predicted sites of metabolism are rank ordered where 1 = most likely, 2 = next most
likely, etc. Predictions calculated using smartCYP v.2.4.2.[165, 166]
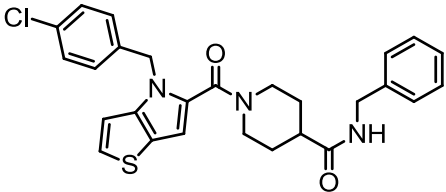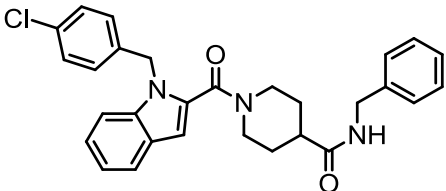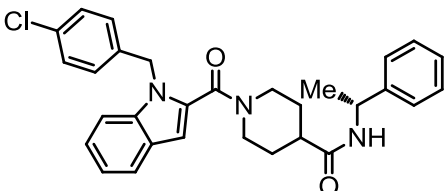
The assay utilized commercially-available mouse liver microsomes harvested

from Balb/C mice, though the line of mice used by our collaborators in the lab of Prof.

David Irani for *in vivo* testing were C57 BL/6, and we found that the results were

translatable. Compounds were incubated in the presence of microsomes over 15 minutes,

and aliquots were removed and quenched over this time at specific time points. We also

developed an LC/MS/MS method to maximize throughput and detection sensitivity so

that parent compound concentration could be quickly assessed at concentrations as low as

single-digit nanomolar, or less than one percent of starting incubation concentration.

SAR

As discussed in **Chapter I**, avoidance of predicted metabolic instability led to the

replacement of the early thienopyrrole scaffold with indole. However, it remained

necessary to demonstrate empirically that we had gained stability through this substitution, so we established a mouse liver microsome-based metabolic stability assay and a corresponding LC/MS/MS analytical method for our lab.

Table 15. MLM Metabolic Stabilities for Thienopyrrole and Indole Leads.

CCG No.	Structure	IC_{50} $(\mu M)^a$	CC_{50} $(\mu M)^b$	MLM $T_{1/2}$ $(min)^c$	ClogP
203881		11.1 ± 1.3	>100	1.7 ± 1.7	4.4
102516†		15.6 ± 1.8	>100	7.3 ± 3.9	4.5
203926†		6.8 ± 1.0	>100	31 ± 7.1	4.9

[a] Inhibition of luciferase expression in WEEV replicon assay. [b] Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. [c] Half-life in BALB/c mouse liver microsome incubation. Values are half-lives calculated from the equation $T_{1/2}=\ln(2)/slope$, where the slope was ln(% detected) against time, plotted from the mean of at least n=3 independent experiments. Values are mean $\pm$ SEM of n = 3 independent experiments. †Synthesized by Janice Sindac.
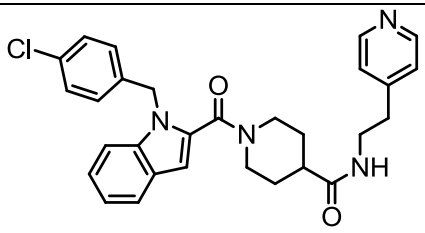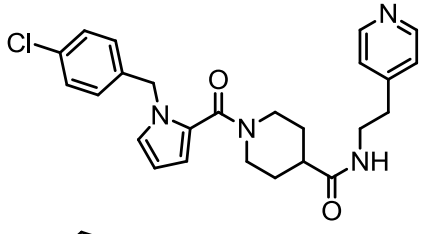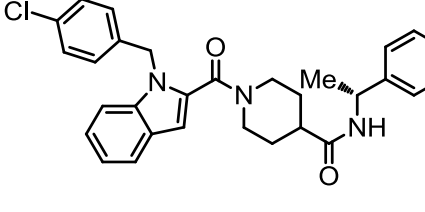
Our newly developed protocol allowed for convenient in-house metabolic stability analysis and was able to differentiate the short half-lives of our leads (**Table 15**). Key to this was optimizing the concentration of microsomes, as the compounds were so

unstable as to be not differentiable under the conditions originally employed. Ultimately, we found that quartering the amount relative to established protocols worked well. In the assay, we discovered that the indole **CCG-102516** had a half-life of 7.3 ± 3.9 minutes, whereas the thienopyrrole **CCG-203881** had a half-life of only 1.7 ± 1.7 minutes, supporting our original stability hypothesis and validating our thienopyrrole/indole switch. Despite this, **CCG-102516** was still quite metabolically unstable. Interestingly, **CCG-203926**, which differs by only a methyl group from **CCG-102516**, had dramatically improved stability, with a half-life of 31 ± 7.1 minutes. This was presumably due to steric hindrance of the potentially labile benzylic site.

Table 16 shows the activities of the lead indole compound **CCG-205432**, its **pyrrole-substituted analog CCG-206381, and the best compound from Table 15, CCG-203926**. Note that the stability of **CCG-203926** (2.3 ± 0.1 minutes) does not match its stability in **Table 15** (31 ± 7.1 minutes); this was mostly likely due to the fresher, more active microsomes used in the former study. However, the relative stabilities are not affected, and we can confidently conclude that **CCG-206381** (3.7 ± 1.3 minutes) and **CCG-205432** (16.7 ± 1.7 minutes) are substantially more stable than **CCG-203926** in the assay. This information suggests that the right-hand benzyl functionality may be a major site of metabolism, since stability is significantly improved by replacement of the benzyl amide with pyridylethyl substitution, although overall reduction in ClogP might also be a factor. The data also indicate that pyrroles are far more metabolically labile than indoles in this template. Detail regarding the application of these data appears in **Chapter III**.

Table 16. MLM Metabolic Stabilities For Indole and Pyrrole Analogs.

CCG No.	Structure	IC$_{50}$ (μM)[a]	CC$_{50}$ (μM)[b]	MLM T$_{1/2}$ (min)[c]	ClogP
205432†		0.53 ± 0.1	65	16.7 ± 1.7	3.1
206381		0.68 ± 0.04	>100	3.7 ± 1.3	1.7
203926†		6.8 ± 1.0	>100	2.3 ± 0.1	4.9

[a]Inhibition of luciferase expression in WEEV replicon assay. [b]Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. [c]Half-life in BALB/c mouse liver microsome incubation. Values are half-lives calculated from the equation T$_{1/2}$=ln(2)/slope, where the slope was ln(% detected) against time, plotted from the mean of at least n=3 independent experiments. Values are mean ± SEM of n = 3 independent experiments. †Synthesized by Janice Sindac.

With the metabolic stability knowledge gained from **Table 15** and **Table 16**, a variety of proposed metabolically stable compounds were prepared and evaluated. Pyridine **CCG-212090** was over ten-fold more stable towards metabolism than **CCG-203926** (**Table 17**). ClogP is highly correlated with CYP metabolism,[170, 171] thus **CCG-212090**'s stability may be attributed to its ten-fold lower ClogP relative to **CCG-203926**. Alternatively, this could suggest that the benzyl ring is a potential site of metabolism, as pyridines are known to be less susceptible due to their electron-deficiency and

polarity.[172, 173] While imidazoles are expected to be more stable than pyrroles by virtue of the electron-withdrawing property of the azine nitrogen, the improved half-life of **CCG-206582** is more likely indicative of CYP inhibition, as the imidazole moiety is known to bind to heme iron, a crucial component of CYP enzyme function.[174-176]

Table 17. MLM Stability and WEEV Replicon Data for Select Analogs.
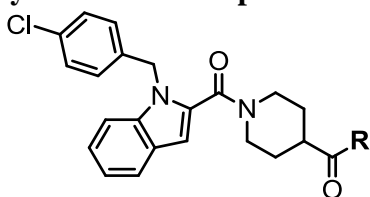
CCG No.	R	IC_{50} $(\mu M)^a$	CC_{50} $(\mu M)^b$	MLM $T_{1/2}$ $(min)^c$	ClogP
205432†		0.53 ± 0.1	65	11.5 ± 0.7	3.1
203926†		6.8 ± 1.0	>100	1.8 ± 0.5	4.9
206582†		4.9 ± 2.6	>100	17.0 ± 2.7	2.7
212090†		0.6 ± 0.3	92	23.4 ± 4.0	3.4

[a]Inhibition of luciferase expression in WEEV replicon assay. [b]Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. [c]Half-life in BALB/c mouse liver microsome incubation. Values are half-lives calculated from the equation $T_{1/2}=\ln(2)/slope$, where the slope was ln(% detected) against time, plotted from the mean of at least n=2 independent experiments. Values are mean $\pm$ SEM of n = 2 independent experiments. †Synthesized by Janice Sindac.

Table 18. MLM Stability and WEEV Replicon Data for Select Analogs.
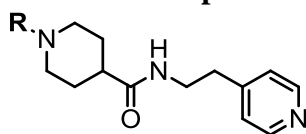
CCG No.	R	IC$_{50}$ (μM)^a	CC$_{50}$ (μM)^b	MLM T$_{1/2}$ (min)^c	ClogP
205432†		0.53 ± 0.1	65	11.5 ± 0.7	3.1
206381		0.68 ± 0.04	>100	2.9 ± 0.1	1.7
211751		4.4 ± 0.5	>100	9.1 ± 3.0	1.9
209023 †		1.6 ± 0.5	46	77.9 ± 20.4	2.4
206565†		4.1 ± 0.1	75	43.0 ± 1.5	2.3

^{a}Inhibition of luciferase expression in WEEV replicon assay. ^{b}Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. ^{c}Half-life in BALB/c mouse liver microsome incubation. Values are half-lives calculated from the equation T$_{1/2}$=ln(2)/slope, where the slope was ln(% detected) against time, plotted from the mean of at least n=2 independent experiments. Values are mean ± SEM of n = 2 independent experiments. †Synthesized by Janice Sindac.

Azaindole **CCG-211751** was prepared in an effort to silence possible sites of metabolism in the indole phenyl ring, but its lack of improved stability indicates that the phenyl portion of the indole may only be a minor site of metabolism (**Table 18**).

Conversely, benzimidazole **CCG-206565** was markedly more stable than the corresponding indole **CCG-205432**, due either to stabilization of the pyrrole ring, or simply reduced ClogP. Intriguingly, replacement of chlorine with hydrogen upon the *N*-benzyl substituent of the template (**CCG-209023**) resulted in a dramatic half-life improvement. This may simply be the result of lower ClogP, as compounds with high logP are known to be susceptible to CYP450 enzymes. Furthermore, it suggests that metabolism of the indole *N*-benzyl is not a major pathway. Despite having less activity than the lead **CCG-205432**, this information allowed us to move **CCG-209023** into mouse studies, on the basis that enhanced metabolic stability would translate into improved bioavailability. Gratifyingly, a ten mouse preliminary study demonstrated a 40% survival rate when infected mice were treated with the compound, marking **CCG-209023** as our most efficacious compound to-date.

Several fluorinated analogs (fluoropyrroles **CCG-209020** and **CCG-209021**, and Janice Sindac's fluoroindoles **CCG-208918** and **CCG-208919**) were synthesized to probe potential metabolic soft spots around the pyrrole and indole rings (**Table 19**). Incorporation of fluorine into compounds is a well-known and powerful tactic for improving stability towards Phase I metabolism as fluorine is similar in size to hydrogen (classical isostere) but is less reactive due to the strength of the C-F bond.[177-179] Interestingly, these analogs, while active in the WEEV replicon assay, were also surprisingly toxic and were therefore not promoted to MLM stability studies.

Table 19. WEEV Replicon Data for Fluorinated Analogs.
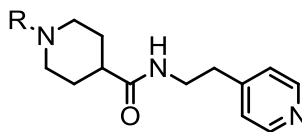
CCG No.	R	IC$_{50}$ (μM)^a	CC$_{50}$ (μM)^b
205432†		0.53 ± 0.1	65
206381		0.68 ± 0.04	>100
209021		0.93 ± 0.02	8.0
208918†		0.73 ± 0.05	24
208919†		0.92 ± 0.05	52

^{a}Inhibition of luciferase expression in WEEV replicon assay. ^{b}Cell viability determined by the MTT reduction assay. Values are mean of at least n=3 independent experiments. †Synthesized by Janice Sindac.

Synthesis

Scheme 35. Preparation of 4-Fluoropyrrole Analogs.[a]

R = pyr, CCG- 209021
R = ind, CCG-209020

"pyr"   "ind"
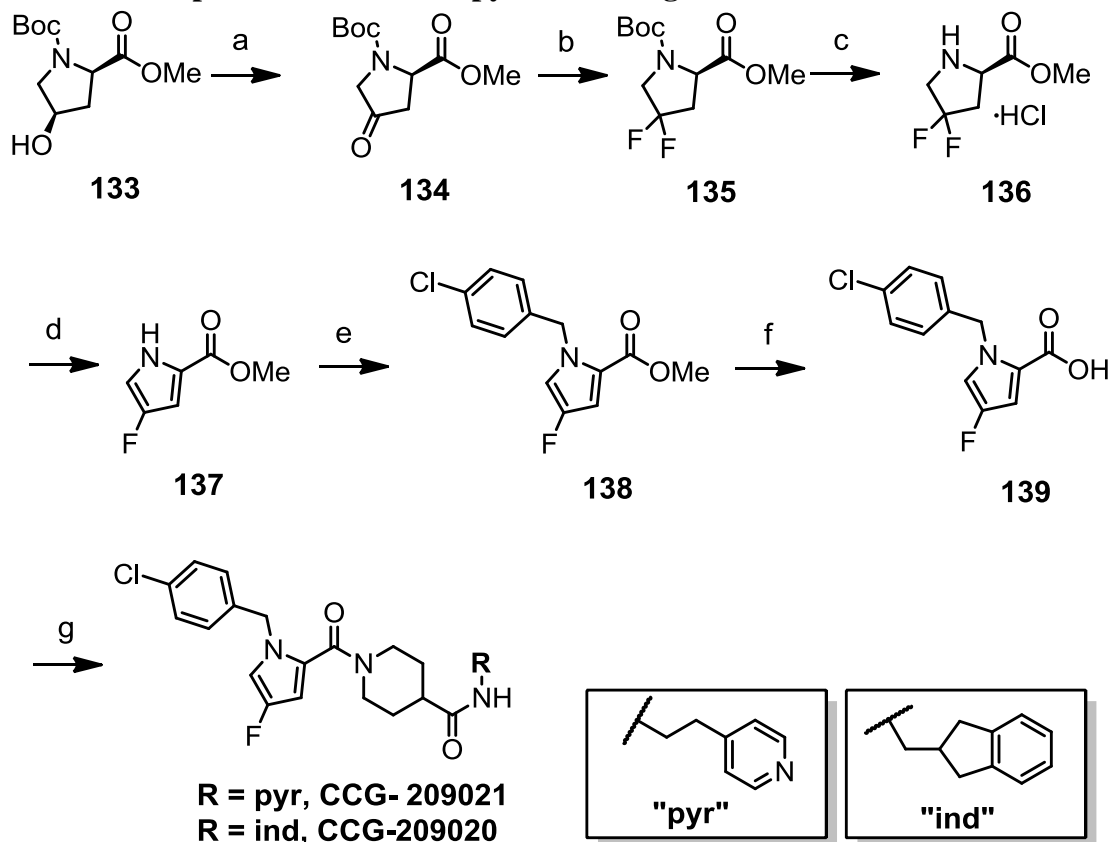
[a]Reagents and conditions: (a) TEMPO, TCCA, DCM, 0 °C–RT, 20 min, 99%; (b) DAST, DCM, -78 °C–RT, 12, h, 86%; (c) 4 M HCl/1,4-dioxane, THF, RT, 4 h, 100%; (d) MnO$_2$, THF, reflux, 3 h, 20%; (e) 4-chlorobenzyl chloride, K$_2$CO$_3$, DMF, 60 °C, 30 h, 83%; (f) 10% aq. NaOH, EtOH, RT, 18 h, 78%; (g)) **NH$_2$R$_1$**, EDC·HCl, HOBT, TEA, DCM, rt, 15 h, 52%.

Fluoropyrroles **CCG-209020** and **CCG-209021** were prepared in a manner similar to that previously described (**Scheme 35**).[180] The 4-hydroxyproline **133** was oxidized to 4-ketoproline **134** under TEMPO catalysis, and this was subsequently fluorinated with DAST to provide the germinal fluoroproline **135**. After acid-mediated deprotection, the resulting 1*H*-proline was aromatized to the 4-fluoropyrrole **127** with

MnO$_2$.[181] Alkylation with 4-chlorobenzyl choride gave the *N*-benzyl intermediate, which was saponified and coupled with the appropriate amine.

Scheme 36. Preparation of 6-Azaindole Analog CCG-211751.[a]

CCG-211751
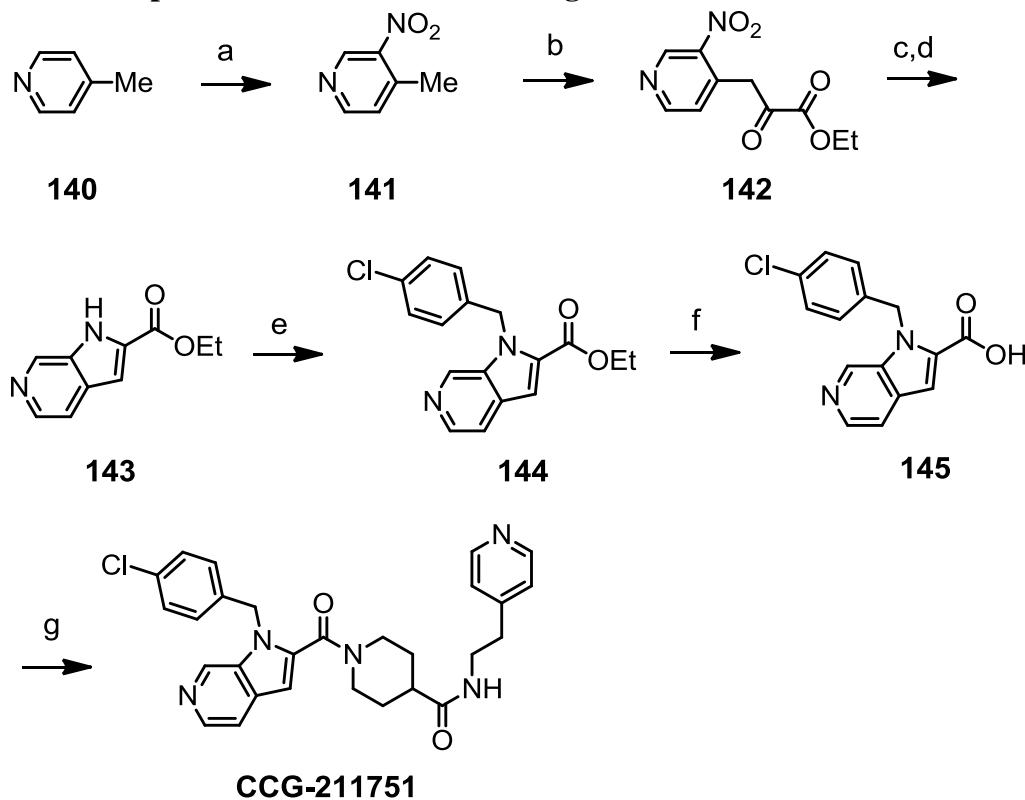
[a]Reagents and conditions: (a) TFAA, 0 °C, 2 h; *then* Fum. HNO$_3$, RT, 12 h; *then* Na$_2$S$_2$O$_5$, H$_2$O, RT, 24 h, 9%; (b) Na^0, diethyl oxalate, EtOH, tol, RT, overnight, 40%; (c) Pt/C, 1 ATM H$_2$, EtOH, AcOH, RT, 24 h; (d) EtOH, K$_2$CO$_3$, RT, 18 h, 51% (2 steps); (e) **2a**, *t*-BuOK, DMF, RT, 14 h, 65%; (f) 10% aq. NaOH, EtOH, RT, 24 h, 84%; (g) **36**, EDC·HCl, HOBT, TEA, DCM, RT, 20 h, 55%.

Azaindole analog **CCG-211751** was prepared using largely known procedures (**Scheme 36**).[182] First, 4-picoline was nitrated under inefficient but mild conditions as reported by Katritzky.[183] The resulting 3-nitro-4-picoline **141** was then treated with sodium in EtOH to form the anion, which was subsequently treated with diethyloxalate to

give the β-ketoester **142**. The nitro was then fully reduced to the amine under heterogeneous catalysis, and, upon introduction of basic conditions, this cyclized via addition/elimination to yield the desired 6-azaindole core **143**. Alkylation with **2a** was accomplished by fully deprotonating the azole nitrogren with *t*BuOK before addition of the electrophile. Weaker bases, such as alkali metal carbonates (*e.g.* Cs_2CO_3) returned mostly pyridinium salts. Saponification of the ester **144** and amidation with the amine dihydrochloride **36** provided the desired azaindole analog **CCG-211751**.

Conclusion

The primary goal of this chapter was the development of a mouse-liver microsome assay and LC/MS/MS protocol suitable for prioritizing compounds based upon their stability towards Phase I metabolism. To this end, we designed a relatively high-throughput MLM method that was successful in assessing and identifying compound stabilities, and that correlated well with *in vivo* pharmacokinetic data. Moreover, it facilitated identification of important modifications (*e.g.* pyrrole-to-aniline substitution and chlorine-to-hydrogen substitution) that significantly improved stability, leading to the discovery of our most stable compound to-date (**CCG-209023**).

Chapter VI. **Future Directions**

Overall, our design and development of inhibitors of neurotropic alphavirus replication has been successful in identifying and optimizing potent compounds both *in vitro* and *in vivo* against a broad spectrum of alphaviruses. Furthermore, we have successfully improved physicochemical properties both predictive and demonstrable of enhanced bioavailability and BBB-penetration. This is a significant step towards the development of a successful antiviral for neurotropic alphaviruses, as there are no known antivirals that possess CNS-permeability that are also therapeutically useful in the treatment of these viruses.[15, 40]

Furthermore, the general mechanism of action is unique as there are few known antivirals that target host machinery. Classically, such targets were avoided due to assumed toxicity issues, but the low toxicity of our compounds places them firmly within a small promising class of new host-targeting antivirals, and the only ones specific for alphaviruses.[184-187] The advantages of such a compound include enhanced broad-spectrum activity and – more importantly – significantly reduced incidence of resistance. Due to relatively rapid viral replication and poor fidelity, resistance-conferring mutations in viral proteins may arise after treatment with viral-targeting drugs, which places a large emphasis on the development of new drugs. On the other hand, due to the relatively low turnover of host cells, resistance-conferring mutations are expected to occur much more slowly, if at all, with host-targeting antivirals.

However, an important aspect of medicinal chemistry is the establishment of a pharmacophore model – the collection of structural and electronic features requisite for engaging with binding site contacts and maximizing activity. Importantly, such a model facilitates the design and development of future analogs, including those based on fundamentally different scaffolds. Over the course of the WEEV inhibitor project, we identified multiple attributes critical to activity, such as the potency-length dependence (**Chapter II**) and the disposition of the right-hand secondary amide (**Chapter IV**). With the aid of our collaborator Paul Kirchhoff, we have attempted to computationally develop a three-dimensional pharmacophore model of our compounds and apply such a model to an *in silico* high-throughput screen to discover new chemical templates. Unfortunately, we have been unable to generate a predictive pharmacophore model; models built from diverse WEEV inhibitor training sets could not discriminate between known active and inactive compounds. Moving forward, we would like to direct studies toward identification of the pharmacophore through the design, synthesis, and evaluation of new analogs.

Related to pharmacophore identification, we have also noted conformation-dependent recognition by MDR1 in this project. Despite the magnitude of on-going work in the field of medicinal chemistry, this observation has been noted only rarely in the literature. Historically, tremendous efforts have been expended by medicinal chemists in identifying *structure*-activity relationships for small molecule interactions with MDR1, but the SAR is still largely undefined due to the promiscuity of the transporter. Therefore, research regarding *conformation*-activity relationships could significantly enhance the design of transporter-evasive compounds.

Another important aspect of drug development is the identification of the biological target, and by extension, the mechanism of action through which the compounds exert their pharmacological effect. Our collaborators have made tremendous strides towards this end,[104] and Janice Sindac has developed several photo-affinity probes to assist in target identification. In contrast to the phenotype-based approach to analog design employed throughout the project, such information could facilitate a target-based avenue to future drug development. In this way, knowledge of the target could lead to models of the binding site (*e.g.* homology) that could allow us to predict and incorporate structural features tailored specifically to engage with potential binding site contacts. Also, because toxicity is often a product of off-target binding to structurally similar biological macromolecules, this could also allow us to improve the toxicity profile of our analogs, especially in light of their host-based mechanism of action. Therefore, the continued design and development of probes – no trivial endeavor – is an important aspect of the project's future.

Chapter VII. **Experimental Data**

All reagents were used as received from commercial sources unless otherwise noted. [1]H and ^{13}C spectra were obtained in DMSO-d_6 or CDCl$_3$ at room temperature, unless otherwise noted, on a Varian Inova 400 MHz, Varian Inova 500 MHz, or Bruker Avance DRX 500 instruments. Chemical shifts for the ^{1}H NMR and ^{13}C NMR spectra were recorded in parts per million (ppm) on the δ scale from an internal standard of residual tetramethylsilane (0 ppm). Rotamers are described as a ratio of rotamer A and B if possible. Otherwise, if the rotamers cannot be distinguished, the NMR peaks are described as multiplets. Mass spectroscopy data were obtained on a Waters Corporation LCT. HPLC retention times were recorded in minutes (min) using an Agilent 1100 series with an Agilent Zorbax Eclipse Plus–C18 column with the gradient 10% ACN/water (1 min), 10–90% ACN/water (6 min), and 90% ACN/water (2 min).

Reagent abbreviations and acronyms: ACAC (acetylacetone), AcOH (acetic acid), Ac$_2$O (acetic anhydride), AlCl$_3$ (aluminum chloride), Ar (argon), BnBr (benzyl bromide), Boc$_2$O (di-*tert*-butyl dicarbonate), Cs$_2$CO$_3$ (cesium carbonate), Cu[0] (elemental copper), Cu$_2$O (copper (I) oxide), DAST (diethylaminosulfur trifluoride), DCM (dichloromethane), DIPA (diisopropylamine), DIPEA (*N,N*-diisopropylethylamine, Hünig's base), DMF (*N,N*-dimethylformamide), DPPP (1,3-

bis(diphenylphosphino)propane, EDC·HCl (1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride), EGDME (ethylene glycol dimethyl ether), Et$_2$O (diethyl ether), EtOAc (ethyl acetate), EtOH (ethanol), H$_2$ (hydrogen), HATU (1-[bis(dimethylamino)methylene]-1H-1,2,3-triazolo[4,5-b]pyridinium 3-oxid hexafluorophosphate), H$_2$O (water), HBr (hydrobromic acid), HCl (hydrogen chloride or hydrochloric acid), Hex (hexanes), HMDS (hexamethyldisilazane), HOBT (1-hydroxybenzotriazole), ind (indane), IPA (isopropanol), K$_2$CO$_3$ (potassium carbonate), KOH (potassium hydroxide), LiAlH$_4$ (lithium aluminum hydride), LiBr (lithium bromide), m-CPBA (*meta*-chloroperoxybenzoic acid), MeCN (acetonitrile), MeI (methyl iodide), MeOH (methanol), MgCl$_2$ (magnesium chloride), MgSO$_4$ (magnesium sulfate), MnO$_2$ (manganese (IV) oxide), N$_2$ (nitrogen), Na^0 (elemental sodium), NaBH$_4$ (sodium borohydride), NaBH$_3$CN (sodium cyanoborohydride), NaBH(OAc)$_3$ (triacetoxy sodium borohydride), Na$_2$CO$_3$ (sodium carbonate), Na$_2$SO$_4$ (sodium sulfate), NAPDH (nicotinamide adenine dinucleotide phosphate), NaH (sodium hydride), NaHCO$_3$ (sodium bicarbonate), NaNO$_2$ (sodium nitrite), NaOH (sodium hydroxide), Na$_2$S$_2$O$_5$ (sodium metabisulfite), NH$_3$ (ammonia), n-BuLi (n-butyl lithium), Pd/C (palladium on carbon), PhCl (chlorobenzene), PhLi (phenyllithium), PhNH$_2$ (aniline), PhN(Tf)$_2$ (*N,N*-bis(trifluoromethylsulfonyl)anilie, PhSeCl (phenylselenyl chloride), PPh$_3$ (triphenylphosphine), Pt/C (platinum on carbon), PtO$_2$ (platinum (IV) oxide), pyr (pyridine), s-BuLi (*sec*-butyl lithium), SmI$_2$ (samarium (II) iodide), SnCl$_2$ (tin (II) chloride), t-BuOH (*tert*-butanol), t-BuOK (potassium *tert*-butoxide), TCCA (trichloroisocyanuric acid), TEA (triethylamine), TEMPO ((2,2,6,6-tetramethylpiperidin-1-yl)oxy), TFA (trifluoracetic acid), TFE (trifluoroethanol), TFFA (trifluoroacetic

anhydride), TFE (2,2,2-trifluoroethanol); THF (tetrahydrofuran), tol (toluene), TsOH (toluenesulfonic acid), Zn^0 (elemental zinc), $ZnCl_2$ (zinc chloride).

Condition abbreviations and acronyms: ATM (atmosphere), aq. (aqueous), conc. (concentrated), d (days), fum. (fuming), g (grams), h (hours), M (molar), mg (milligrams), mL (milliliters), µL (microliters), mM (millimolar), min (minutes), psi (pressure per square inch), RT (room temperature), wt. (weight).

Biology

Mouse Microsomal Metabolic Stability Procedure. Balb-C mouse liver microsomes were purchased from Invitrogen. $MgCl_2$ and NADPH were obtained from Sigma. Solvents were of HPLC grade or better. The HPLC-MS/MS system consisted of a ThermoElectron Finnigan TSQ Quantum Ultra AM.

Microsomal incubations were done in triplicate. Incubation mixtures contained 2 µL of 20mg/mL microsomes (approximately 0.04 mg microsomal protein), 4 µL of 100 µM DMSO-dissolved substrate (1.0 µM final), in 475 µL potassium phosphate buffer (0.1 M, pH 7.5, containing 3.3 mM $MgCl_2$.) The incubation mixture was allowed to shake at 37 °C for 5 min, and a T=0 aliquot was removed. Reaction was initiated by the addition of 20 µL of NaDPH (22 mM under the same buffer conditions) and the mixture was allowed to shake at 37 °C until the final aliquot was removed. 30 µL aliquots were taken after 0 minutes, 5 minutes, and 15 minutes. All aliquots were quenched by dilution in 90 µL MeCN containing an internal standard and the precipitate was pelleted via centrifugation. 20 µL of supernatant was injected onto the HPLC-MS/MS. Mobile phase

A was 95:5 H_2O:MeCN with 0.1% formic acid, and mobile phase B was MeCN with 0.1% formic acid. Column used was a Luna C18(2) 4.6 X 30 mm column with 3μM particle size. The flow rate was 2 mL/min and a gradient mobile phase composition was used: isocratic hold for 1 min at 90% A (10% B), 1 min gradient to 10% A (90% B), 1 min gradient back to 90% A (10% B), and 1 min isocratic hold at 90% A (10% B.) Ionization method consisted of positive electrospray. Source parameters were optimized for each individual substrate. Substrates and internal standards were followed by selected reaction monitoring. Substrate/internal standard area ratios were determined and converted into percent substrate remaining. The natural log of the percent remaining was plotted against time, the slope of the linear regression was determined, and the equation $T_{1/2} = -\ln(2)/k$ was used to calculate half-life.

Chemistry

Ethyl 1-(4-chlorobenzyl)-1H-indole-2-carboxylate (**3a**). Ethyl 1*H*-indole-2-carboxylate **1** (4.50 g, 23.78 mmol) was dissolved in anhydrous DMF (60 mL) under N_2 at RT, then cooled to -10 °C. Granular *t*-BuOK (2.94 g, 26.20 mmol) was added to the solution, eliciting a slight yellow color. 4-Chlorobenzyl chloride **2a** (4.02 g, 24.97 mmol) – pre-dissolved in anhydrous DMF (5 mL) – was added via syringe pump at 0.1 mL/min at -10 °C. After this time, was allowed to stir at RT for 12 h. The reaction was then diluted with 1:1 EtOAc:Hex and quenched by the addition of sat. aq. $NaHCO_3$. The organic phase was then washed with H_2O (10 X) and brine (1X), dried ($MgSO_4$), and concentrated to give the pure compound as a white solid that was subsequently washed thoroughly with hexanes. Yield: 6.80 g, 21.67 mmol, 91%. ^{1}H NMR (500 MHz,

Chloroform-d) δ 7.78 (d, *J* = 8.0 Hz, 1H), 7.48 (d, *J* = 1.6 Hz, 1H), 7.38 – 7.34 (m, 2H), 7.25 (ddd, *J* = 15.1, 8.4, 2.8 Hz, 3H), 7.07 – 7.01 (m, 2H), 5.83 (s, 2H), 4.39 (qd, *J* = 7.1, 1.6 Hz, 2H), 1.42 (td, *J* = 7.1, 1.6 Hz, 3H).

Ethyl   1-(4-fluorobenzyl)-1H-indole-2-carboxylate **(3b)**.   Ethyl   1*H*-indole-2-carboxylate **1** (500 mg, 2.64 mmol) was dissolved in anhydrous DMF (8 ml), followed by 1-(chloromethyl)-4-fluorobenzene **2b** (0.380 ml, 3.17 mmol) and the addition of solid K_2CO_3 (730 mg, 5.29 mmol). This was stirred at 60 °C for 20 h, at which time the reaction was allowed to cool to room temperature. Precipitate formed upon the addition of water (40 mL), was isolated over a filter, and dried under high vacuum overnight to give 550 mg of the title compound as a white solid which was recrystallized from Et_2O. Yield: 550 mg, 1.850 mmol, 70%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.72 (d, *J* = 7.9 Hz, 1H), 7.40 (s, 1H), 7.37 – 7.31 (m, 2H), 7.18 (ddd, *J* = 7.7, 6.5, 1.3 Hz, 1H), 7.05 (dd, *J* = 8.4, 5.3 Hz, 2H), 6.94 (t, *J* = 8.5 Hz, 2H), 5.81 (s, 2H), 4.34 (q, *J* = 7.1 Hz, 2H), 1.38 (t, *J* = 7.1 Hz, 3H).

1-(4-Chlorobenzyl)-1H-indole-2-carboxylic acid **(4a)**. Ethyl 1-(4-chlorobenzyl)-1*H*-indole-2-carboxylate **3a** (5.95 g, 18.96 mmol) was added to a mixture of 10% aq. NaOH (13.7 mL, 37.9 mmol), EtOH (15 mL), and THF (15 mL), but did not completely dissolve. This suspension stirred at RT for 14 h, over which time all material dissolved and the reaction proceeded to completion. Solvent was stripped off *in vacuo*, the residue was taken up in H_2O and acidified to pH<1 with conc. HCl at 0 °C. The resulting precipitate was collected over a filter and washed with cold 1 M HCl, air-dried, then

dried thoroughly under high vacuum to give a white powder. No further purification necessary. Yield: 5.20 g, 18.20 mmol, 96%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.76 (d, J = 8.1 Hz, 1H), 7.56 (s, 1H), 7.40 − 7.31 (m, 2H), 7.27 − 7.16 (m, 3H), 7.00 (d, J = 8.2 Hz, 2H), 5.81 (s, 2H).

1-(4-Fluorobenzyl)-1H-indole-2-carboxylic acid (**4b**). Ethyl 1-(4-fluorobenzyl)-1H-indole-2-carboxylate **3b** (409 mg, 1.376 mmol) was suspended in ethanol (5 mL) and 7M NaOH (5 mL) and stirred at 50 °C for 3 h. Ethanol content was reduced *in vacuo*. The solution was then acidified to pH 2 and the precipitate was collected over a filter, washed with a small amount of ice cold-water, air dried on the filter, and then finally vacuum dried to give the title compound as a fine, white powder. Yield: 360 mg, 1.337 mmol, 97%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.41 (d, J = 5.8 Hz, 1H), 7.81 − 7.71 (m, 4H), 7.70 − 7.60 (m, 3H), 7.18 (d, J = 5.7 Hz, 1H), 5.90 (d, J = 5.5 Hz, 2H).

Tert-Butyl 4-(hydroxymethyl)piperidine-1-carboxylate (**7**). Piperidine-4-methanol **6** (3.74 g, 32.5 mmol) and Boc_2O (8.5 g, 38.9 mmol) were added to THF (50 mL) at RT, followed by Na_2CO_3 (6.88 g, 64.9 mmol) and H_2O (30 mL). Addition was mildly exothermic. The reaction was refluxed at 100 °C for 2 h, after which it was allowed to cool to RT. The reaction was then extracted with EtOAc, and the extract was washed with brine (1X), dried ($MgSO_4$), and concentrated. The residue was purified by silica flash chromatography (150 g silica, 50% EtOAc:Hex). Concentration provided a white solid. Yield: 6.09 g, 28.3 mmol, 87%. ^{1}H NMR (500 MHz, Chloroform-d) δ 4.10 (s, 2H),

3.47 (t, J = 5.2 Hz, 1H), 2.69 (s, 2H), 1.70 (d, J = 13.2 Hz, 2H), 1.62 (ddp, J = 15.2, 6.7, 4.0 Hz, 1H), 1.44 (s, 9H), 1.12 (qd, J = 12.5, 4.4 Hz, 2H).

Tert-Butyl   4-formylpiperidine-1-carboxylate   **(8)**.   Tert-butyl   4-(hydroxymethyl)piperidine-1-carboxylate **7** (1.50 g, 6.97 mmol) and anhydrous DMSO (1.24 mL, 17.42 mmol) were dissolved in DCM (100 mL) at RT, then cooled to -78 °C. $(COCl)_2$ (1.22 mL, 13.93 mmol) was then added dropwise under N_2 and the reaction was allowed to stir for 30 min at -78 °C. Still at this temperature, TEA (4.86 mL, 34.8 mmol) was added dropwise and a slight yellow color change was observed. The reaction was allowed to stir for 30 min at -78 °C, then 3 h at 0 °C. After this time, the reaction was quenched by the addition of 0.1 M HCl and material was extracted with EtOAc. The extract was washed with 0.1 M HCl (2X), H_2O (3X), and brine (1X), then dried ($MgSO_4$) and concentrated *in vacuo*. This was immediately purified via silica flash chromatography (100 g silica, 40% EtOAc:Hex) to provide the desired product as a white crystal after removal of solvent. Yield: 1.32 g, 6.19 mmol, 89%. ^{1}H NMR (400 MHz, Chloroform-d) δ 9.58 (s, 1H), 3.99 – 3.82 (m, 2H), 2.85 (t, J = 11.9 Hz, 2H), 2.35 (dt, J = 10.5, 6.0 Hz, 1H), 1.82 (d, J = 12.4 Hz, 2H), 1.52 – 1.44 (m, 2H), 1.37 (s, 9H).

Tert-Butyl 4-(piperidin-1-ylmethyl)piperidine-1-carboxylate **(9)**. Piperidine (1.2 mL, 11.73 mmol) was dissolved in TFE (10 mL) over 3 Å MS, then a drop of glacial AcOH (~ 50 µL) was added. *Tert*-butyl 4-formylpiperidine-1-carboxylate **8** (500 mg, 2.35 mmol) was added slowly with stirring at RT. $NaCNBH_3$ (192 mg, 4.69 mmol) was added and the reaction was stirred at RT for 19 h. After this time, the reaction was diluted

with EtOAc and washed with 10% aq. Na_2CO_3 (3X), dried ($MgSO_4$), concentrated under high vacuum. The residue was then purified by silica flash chromatography (60 g silica, 70% to 100% EtOAc:Hex). Yield: 477 mg, 1.69 mmol, 72%. ^{1}H NMR (500 MHz, Chloroform-d) δ 4.03 (bs, 2H), 2.64 (bs, 2H), 2.28 (bs, 3H), 2.08 (d, J = 6.4 Hz, 2H), 1.71 – 1.45 (m, 10H), 1.42 (s, 9H), 1.03 (m, 2H).

Tert-Butyl 4-(cyclohexyl(hydroxy)methyl)piperidine-1-carboxylate **(10)**. *Tert*-butyl 4-formylpiperidine-1-carboxylate **8** (250 mg, 1.172 mmol) was dissolved in anhydrous Et_2O (20 mL) and a 2 M solution of cyclohexylmagnesium chloride (0.62 mL, 1.23 mmol) in Et_2O was added dropwise to this under N_2. The reaction was allowed to stir at RT for 2 h, after which time the reaction was quenched by the addition of EtOH then sat. aq. $NaHCO_3$. Material was extracted with EtOAc, and the extract was dried ($MgSO_4$) and concentrated to give the desired alcohol as a solid, which was used directly in the subsequent reaction without purification. Yield: 296 mg, 0.20 mmol, 85%.

Tert-Butyl 4-(dibenzylamino)piperidine-1-carboxylate **(13)**. *Tert*-butyl 4-aminopiperidine-1-carboxylate **12** (200 mg, 1.00 mmol) was dissolved in DCM (3 mL), followed by the addition of H_2O (1.5 mL) and granular Na_2CO_3 (318 mg, 3.00 mmol), and finally benzyl bromide (0.30 mL, 2.50 mmol). The reaction was stirred at reflux (55 °C) for 3 h, after which H_2O (10 mL) was added and the material was extracted with EtOAc. The extract was dried ($MgSO_4$) and concentrated, and the resulting residue was purified by silica flash chromatography (20 g silica, 5% EtOAc:Hex) to give the desired amine as a white solid. Yield: 330 mg, 0.87 mmol, 87%. ^{1}H NMR (500 MHz,

Chloroform-d) δ 7.40 (d, J = 7.2 Hz, 4H), 7.33 (t, J = 8.2, Hz, 4H), 7.29 − 7.23 (m, 2H), 4.18 (s, 2H), 3.67 (s, 4H), 2.69 (tt, J = 11.8, 3.5 Hz, 1H), 2.59 (bs, 2H), 1.84 (d, J = 12.4 Hz, 2H), 1.63 − 1.52 (m, 2H), 1.52 − 1.46 (m, 9H).

4-(3-Bromopropyl)pyridine (**16**). 3-(Pyridin-4-yl)propan-1-ol (500 mg, 3.64 mmol) was dissolved in conc. HBr (1 mL) in a pressure vessel stirred at 130 °C for 2h, after which the reaction was allowed to cool. The reaction was then basified to pH>14 with 10% aq. NaOH at 0 °C and extracted with EtOAc (3X). The combined extracts were dried (MgSO$_4$) and concentrated. The residue was then purified on a silica plug (20 g silica, 100% EtOAc) to give a crude oil that was taken into the next reaction without further purification. Yield: 292 mg, 1.46 mmol, 40%.). TOF ES+ MS: (M + H, Br^{79}) 200.0, (M + H, Br^{81}) 202.0.

Tert-Butyl 4-(3-(pyridin-4-yl)propyl)piperazine-1-carboxylate (**17**). *t*-Butyl piperazine-1-carboxylate (186 mg, 1.00 mmol) was dissolved in anhydrous DMF (10 mL) under N$_2$, and a 60% oil suspension of NaH (120 mg, 3.00 mmol) was added. After stirring at RT under N$_2$ for 1 h, 4-(3-bromopropyl)pyridine (200 mg, 1.00 mmol) was added and the reaction was allowed to stir for 19 h. After this time, the reaction was quenched with sat. aq. NaHCO$_3$ then diluted with EtOAc and washed with H$_2$O (3X), 10% aq. Na$_2$CO$_3$ (3X), and brine (1X), then dried (MgSO$_4$) and concentrated. The residue was purified by silica flash chromatography (25 g silica, 1% to 10% methanolic ammonia:EtOAc). Yield: 201 mg, 0.660 mmol, 66%. ^{1}H NMR (500 MHz, Chloroform-d)

δ 8.49 (d, *J* = 5.9 Hz, 2H), 7.11 (d, *J* = 5.8 Hz, 2H), 3.45 − 3.39 (m, 4H), 2.65 (t, *J* = 7.7 Hz, 2H), 2.42 − 2.31 (m, 6H), 1.83 (p, *J* = 7.6 Hz, 2H), 1.46 (s, 9H).

(1-(4-Fluorobenzyl)-1H-indol-2-yl)(4-(hydroxymethyl)piperidin-1-yl)methanone **(CCG-206462)**. The following was added sequentially to DMF (10 mL): 1-(4-fluorobenzyl)-1*H*-indole-2-carboxylic acid (1.175 g, 4.36 mmol), DIPEA (2.29 mL, 13.09 mmol), EDC·HCl (1.00 g, 5.24 mmol), and HOBT (802 mg, 5.24 mmol). This solution was allowed to stir at RT for 1.5 h, at which time 4-piperidinemethanol (754 mg, 6.55 mmol) was added. Stirring continued for 22 h at RT, at which time the solution was partitioned between H_2O and 1:1 solution of EtOAc:Et$_2$O. The organic extract was then washed with saturated aq. Na_2CO_3, dried with $MgSO_4$, and concentrated *in vacuo*. Purification was accomplished via silica flash chromatography (100 g silica, 80% EtOAc/Hexanes.) Resulting residue was crystallized via Et$_2$O:Hex trituration to give the title compound as a white solid. Yield: 1.51 g, 4.11 mmol, 94%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.63 (d, *J* = 7.9 Hz, 1H), 7.37 (d, *J* = 8.3 Hz, 1H), 7.29 − 7.22 (m, 1H), 7.14 (t, *J* = 7.5 Hz, 1H), 7.10 − 7.04 (m, 2H), 6.91 (t, *J* = 8.2 Hz, 2H), 6.60 (s, 1H), 5.46 (s, 2H), 4.64 (bs, 1H), 4.14 (bs, 1H), 3.42 (t, *J* = 5.1 Hz, 2H), 2.76 (s, 2H), 1.44 (t, *J* = 5.0 Hz, 1H), 1.05 (bs, 1H), 0.71 (bs, 1H).

1-(1-(4-Fluorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carbaldehyde **(20)**. (1-(4-fluorobenzyl)-1*H*-indol-2-yl)(4-(hydroxymethyl)piperidin-1-yl)methanone (762 mg, 2.08 mmol) was dissolved in DCM (80 mL) in dry glassware under N_2. The solution was chilled to -78 °C and anhydrous DMSO (0.37 mL, 5.20 mmol) and (COCl)$_2$ (0.36 mL,

4.16 mmol) were added. The reaction was allowed to stir for 30 min, after which TEA (1.45 mL, 10.40 mmol) was added dropwise and stirring continued for 1 h, after which the temperature was elevated to 0 °C and stirring continued for 30 min. At this time, the solution was taken up in EtOAc while still cold and immediately washed with 0.1 M HCl (2X), dried (MgSO$_4$), and concentrated to give a white solid. Yield: 527 mg, 1.46 mmol, 70%. ^{1}H NMR (500 MHz, Chloroform-d) δ 9.61 (s, 1H), 7.66 (d, J = 7.9 Hz, 1H), 7.40 (d, J = 8.2 Hz, 1H), 7.32 – 7.26 (m, 1H), 7.19 – 7.14 (m, 1H), 7.12 – 7.06 (m, 2H), 6.93 (t, J = 8.6 Hz, 2H), 6.63 (s, 1H), 5.48 (s, 2H), 4.55 (bs, 1H), 4.07 (s, 1H), 2.50 – 2.41 (m, 1H), 2.16 – 1.93 (m, 2H), 1.84 – 1.47 (m, 4H).

Methyl 1-(4-chlorobenzyl)-1H-pyrrole-2-carboxylate (**23a**). Methyl 1*H*-pyrrole-2-carboxylate (2.77 g, 22.14 mmol) was dissolved in anhydrous DMF (15 mL). Granular K$_2$CO$_3$ (1.94 g, 26.6 mmol) was added, followed by the addition of 4-chlorobenzyl chloride (1.80 mL, 26.6 mmol). The reaction was permitted to stir at 60 °C for 24 h, then again 90 °C for 48 h. Cs$_2$CO$_3$ (21.6 g, 66.4 mmol) was then added, and the reaction was allowed to stir at 90 °C for 24 h, then NaI (332 mg, 2.214 mmol) was added and stirring continued at 90 °C for 24 h. At this time, the reaction was allowed to cool to RT, diluted with EtOAc, and washed with brine (3×). The organic phase was then dried (MgSO$_4$), concentrated *in vacuo,* and purified by silica flash chromatography (150 g silica, 5% EtOAc:Hex) to provide methyl 1-(4-chlorobenzyl)-1*H*-pyrrole-2-carboxylate as colorless crystals. Yield: 4.91 g (89%). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.32 (d, J = 4.7 Hz, 1H), 7.26 (d, J = 3.5 Hz, 1H), 7.05–6.98 (m, 3H), 6.92–6.86 (m, 1H), 6.20 (dd, J = 3.9, 2.6 Hz, 1H), 5.52 (s, 2H), 3.76 (s, 3H).

Ethyl 1-(4-chlorobenzyl)-1H-imidazole-2-carboxylate **(23b)**. Ethyl 1*H*-imidazole-2-carboxylate (4 g, 28.5 mmol), 4-chlorobenzyl chloride (4.38 mL, 34.3 mmol), and Na_2CO_3 (3.63 g, 34.3 mmol), was dissolved in anhydrous DMF (8 mL). The solution was stirred at RT for 24 h, at which time H_2O was added and material was extracted with EtOAc. The organic phase was collected, dried over $MgSO_4$, and decanted. Purification accomplished via silica flash chromatography (150 g silica, 10% EtOAc/Hexanes to 80% EtOAc/Hex.) The title compound was obtained as a clear, yellow-tinted oil. Yield: 7.28 g, 27.5 mmol, 96%. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.29 (d, J = 8.4 Hz, 2H), 7.18 (d, J = 0.9 Hz, 1H), 7.09 (d, J = 8.4 Hz, 2H), 7.06 (d, J = 0.9 Hz, 1H), 5.59 (s, 2H), 4.37 (q, J = 7.2 Hz, 2H), 1.39 (t, J = 7.1 Hz, 3H).

Ethyl 1-(1-(4-chlorobenzyl)-1H-pyrrole-2-carbonyl)piperidine-4-carboxylate **(24a)**. To methyl 1-(4-chlorobenzyl)-1*H*-pyrrole-2-carboxylate (4.5 g, 18.02 mmol) was added to EtOH (50 mL) and 10% aq. NaOH (20 mL) and stirred overnight at RT, then again overnight at 50 °C to give completion. The EtOH was stripped by rotary evaporation until material began to precipitate, at which point the aqueous mixture was cooled in an ice bath and acidified with conc. HCl, which elicited further precipitation. The precipitate was collected over a filter and washed with a small amount of cold 1 M HCl and Hex. The material was then dried under high vacuum to provide the carboxylic acid as a white powder. Yield: 4.20 g, (99%). ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.34 (d, J = 8.2 Hz, 2H), 7.19 (s, 1H), 7.06 (d, J = 8.2 Hz, 2H), 6.82 (s, 1H), 6.16–6.08 (m, 1H), 5.53 (s, 2H).

The following were added sequentially to anhydrous DMF (20 mL), 1-(4-chlorobenzyl)-1H-pyrrole-2-carboxylic acid (2.0 g, 8.5 mmol), DIEA (4.45 mL, 25.5 mmol), EDCI (1.79 g, 9.34 mmol), and HOBt (1.43 g, 9.34 mmol), and this solution was allowed to stir at RT for 30 min. Finally, ethyl isonipecotate (1.44 mL, 9.34 mmol) was added and the reaction was permitted to stir at RT for 24 h. At this time, DCM was removed in vacuo and the residue was dissolved in ethyl acetate, which was subsequently washed with 1 M HCl (3X), H_2O (2X), 10% aq. Na_2CO_3 (2X), and at last brine (1X). The organic layer was then dried ($MgSO_4$) and concentrated *in vacuo*. The residue was crystallized from EtOAc:Hex via the slow evaporation of solvent at RT to give ethyl 1-(1-(4-chlorobenzyl)-1H-pyrrole-2-carbonyl)piperidine-4-carboxylate as large translucent crystals. Yield: 2.16 g (68%). ^{1}HNMR (400 MHz, CDCl3) δ 7.24 (d, J = 8.4 Hz, 2H), 7.03 (d, J = 8.4 Hz, 2H), 6.84−6.74 (m, 1H), 6.31 (dd, J = 3.7, 1.5 Hz, 1H), 6.18−6.06 (m, 1H), 5.27 (s, 2H), 4.21 (d, J = 11.5 Hz, 2H), 4.14 (q, J = 7.1 Hz, 2H), 2.94 (t, J = 11.5 Hz, 2H), 2.46 (tt, J = 10.7, 4.0 Hz, 1H), 1.79 (d, J = 12.7 Hz, 2H), 1.56−1.30 (m, 2H), 1.25 (t, J = 7.1 Hz, 3H).

1-(1-(4-Fluorobenzyl)-1H-pyrrole-2-carbonyl)piperidine-4-carboxylic acid **(25a)**. Ethyl 1-(1-(4-chlorobenzyl)-1H-pyrrole-2-carbonyl)piperidine-4-carboxylate (700 mg, 1.867 mmol) was dissolved in EtOH (60 mL) and H_2O (20 mL). Granular LiOH (134 mg, 5.60 mmol) was added to this, and the reaction was allowed to stir at RT for 24 h. After this time, most of the solvent was removed in vacuo, and the remaining aqueous solution was chilled to 0 °C and acidified by concentrated HCl, which elicited a thick, sticky precipitate that adhered to the flask. The aqueous solution was decanted, and a small

amount of 1 N HCl was added, and the material sonicated. The aqueous solution was again decanted, and a small amount of ice-cold water was added, mixed, and decanted. Remaining water and HCl was then removed by rotary evaporation, followed by high-vacuum drying to afford the title compound as a white powder. Yield: 620 mg (96%).
^{1}HNMR (400 MHz, DMSO-d6) δ 7.33 (dd, J = 9.0, 2.7 Hz, 2H), 7.14−6.97 (m, 2H), 6.27 (dd, J = 3.6, 1.5 Hz, 1H), 6.10−5.94 (m, 1H), 5.25 (s, 2H), 3.98 (d, J = 11.0 Hz, 2H), 3.03−2.80 (m, 2H), 2.35−2.19 (m, 1H), 1.64 (d, J = 11.1 Hz, 2H), 1.24 (d, J = 11.5 Hz, 2H).

Methyl 1-(1-(4-chlorobenzyl)-1H-pyrrole-2-carbonyl)azetidine-3-carboxylate **(29).** 1-(4-chlorobenzyl)-1H-pyrrole-2-carboxylic acid (500 mg, 2.12 mmol) was added to DCM (20 mL), followed sequentially by DIPEA (1.1 mL, 6.36 mmol), EDC·HCl (447 mg, 2.33 mmol), HOBT (357 mg, 2.33 mmol), and methyl azetidine-3-carboxylate hydrochloride (387 mg, 2.55 mmol). The reaction was allowed to stir at rt for 18 h, at which time the DCM was removed *in vacuo* and the residue was taken up in 2:1 EtOAc:Et$_2$O and washed with 1 M aq. HCl (5X), H$_2$O (5X), 10% aq. Na$_2$CO$_3$ (5X), and brine (1X), then dried (MgSO$_4$) and concentrated. The resulting residue was purified by silica flash chromatography (25 g silica, 5% to 30% EtOAc/Hex) to give the desired compound as a clear, slightly yellow oil. Yield: 508 mg, 1.53 mmol, 72%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.25 (d, *J* = 8.3 Hz, 2H), 7.04 (d, *J* = 8.2 Hz, 2H), 6.81 (s, 1H), 6.58 − 6.44 (m, 1H), 6.20 − 6.13 (m, 1H), 5.52 (s, 2H), 4.36 (bs, 4H), 3.75 (s, 3H), 3.43 (p, *J* = 7.5 Hz, 1H).

Ethyl 1-((4-chlorobenzyl)carbamoyl)piperidine-4-carboxylate **(31a)**. (4-chlorobenzyl)isocyanate (0.30 mL, 2.26 mmol) was dissolved in DCM (10 mL) at RT, then ethyl isonipecotate **26** (0.38 mL, 2.49 mmol) was added dropwise with stirring, resulting in precipitation and discoloration. This slurry was allowed to stir at RT for 1 h, at which time the precipitate was collected over a filter and dried via aspirator to afford an off-white granular solid. No further purification was necessary. Yield: 690 mg, 2.13 mmol, 94%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.30 – 7.25 (m, 2H), 7.25 – 7.20 (m, 2H), 4.84 (s, 1H), 4.37 (d, *J* = 5.6 Hz, 2H), 4.14 (q, *J* = 7.1 Hz, 2H), 3.87 (dt, *J* = 13.3, 3.7 Hz, 2H), 2.96 – 2.87 (m, 2H), 2.46 (ddd, *J* = 14.6, 10.7, 3.8 Hz, 1H), 1.91 (dd, *J* = 13.4, 3.1 Hz, 2H), 1.71 – 1.62 (m, 2H), 1.25 (t, *J* = 7.2 Hz, 3H).

Ethyl 1-((4-chlorophenethyl)carbamoyl)piperidine-4-carboxylate **(31b).** 1-Chloro-4-(2-isocyanatoethyl)benzene 19 (0.45 mL, 2.94 mmol) was dissolved in DCM (10 mL) at RT. Ethyl isonipecotate **26** was then added dropwise, which elicited precipitation. The slurry was allowed to stir at RT for 1 h, at which time the precipitate was collected and dried over a filter to afford ethyl 1-((4-chlorophenethyl)-carbamoyl)piperidine-4-carboxylate as an off-white granular solid. This material was taken into the next reaction in this crude form. Yield: 916 mg (92%). ^{1}H NMR (500 MHz, Chloroform-d) δ 7.26 (d, *J* = 8.2 Hz, 2H), 7.11 (d, *J* = 8.3 Hz, 2H), 4.52 (t, *J* = 5.3 Hz, 1H), 4.13 (q, *J* = 7.1 Hz, 2H), 3.79 (dt, *J* = 13.4, 3.8 Hz, 2H), 3.44 (q, *J* = 6.7 Hz, 2H), 2.90 – 2.80 (m, 2H), 2.78 (t, *J* = 6.9 Hz, 2H), 2.44 (tt, *J* = 10.8, 3.9 Hz, 1H), 1.92 – 1.83 (m, 2H), 1.62 (qd, *J* = 11.2, 4.1 Hz, 2H), 1.25 (t, *J* = 7.1 Hz, 3H).

Ethyl 1-(2-(4-chlorophenyl)acetyl)piperidine-4-carboxylate (**33a**). 4-chlorophenylacetic acid (900 mg, 5.28 mmol) was dissolved in DCM (20 mL), followed by the sequential addition of TEA (2.21 mL, 15.83 mmol), EDC·HCl (1110 mg, 5.80 mmol), HOBT (889 mg, 5.80 mmol), and ethyl isonipecotate (0.894 mL, 5.80 mmol). The reaction was stirred at RT for 28 h, after which time the DCM was evaporated off. The residue was taken up in EtOAc and washed with 1M HCl (3X), H_2O (5X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated. The residue was then purified through a plug of silica (30 g silica, 5% EtOAc:Hex). Yield: 1.31 g, 4.22 mmol, 80%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.29 (d, *J* = 8.3 Hz, 2H), 7.17 (d, *J* = 8.2 Hz, 2H), 4.40 (dt, *J* = 12.5, 4.0 Hz, 1H), 4.13 (q, *J* = 7.1 Hz, 2H), 3.79 (dt, *J* = 13.5, 3.7 Hz, 1H), 3.69 (s, 2H), 3.12 − 3.05 (m, 1H), 2.89 − 2.82 (m, 1H), 2.52 − 2.45 (m, 1H), 1.96 − 1.87 (m, 2H), 1.83 − 1.77 (m, 1H), 1.61 − 1.57 (m, 1H), 1.49 (ddd, *J* = 24.1, 10.9, 3.8 Hz, 1H), 1.25 (t, *J* = 7.1 Hz, 3H).

Ethyl 1-(3-(4-chlorophenyl)propanoyl)piperidine-4-carboxylate (**33b**). 3-(4-chlorophenyl)propionic acid (0.90 g, 4.87 mmol) was dissolved in DCM (20 mL), followed by TEA (2.04 mL, 14.62 mmol), EDC·HCl (1.03 g, 5.36 mmol), HOBT (0.82 g, 5.36 mmol), and ethyl isonipecotate (0.83 mL, 5.36 mmol). The solution was stirred at RT for 28 h, after which time the DCM was removed *in vacuo* and the residue was dissolved in 2:1 EtOAc:Et$_2$O and washed with 1 M aq. HCl (3X), H_2O (2X), 10% aq. Na_2SO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated. The residue was then purified by silica flash chromatography (100 g silica, 5% to 50% EtOAc:Hex). ^{1}H NMR (500 MHz, Chloroform-d) δ 7.20 (d, *J* = 8.2 Hz, 2H), 7.11 (d, *J* = 8.2 Hz, 2H), 4.41

– 4.34 (m, 1H), 4.10 (q, J = 7.1 Hz, 2H), 3.71 (d, J = 13.7 Hz, 1H), 3.04 – 2.96 (m, 1H), 2.89 (t, J = 7.8 Hz, 2H), 2.80 – 2.72 (m, 1H), 2.58 – 2.53 (m, 2H), 2.46 (tt, J = 10.7, 4.0 Hz, 1H), 1.90 – 1.80 (m, 2H), 1.54 (ddd, J = 24.1, 13.3, 3.5 Hz, 2H), 1.22 (t, J = 7.1 Hz, 3H).

1-Tert-Butyl 4-ethyl piperidine-1,4-dicarboxylate **(34)**. Ethyl isonipecotate **26** (3.53 mL, 22.91 mmol), Boc$_2$O (6.0 g, 27.50 mmol), and Na$_2$CO$_3$ (4.86 g, 45.80 mmol) were added to a mixture of H$_2$O (8 mL) and THF (20 mL) at RT (mild exotherms). This was subsequently refluxed at 80 °C for 2 h, over which time the reaction proceeded from a white suspension to a clear biphasic solution. The reaction was allowed to cool and was extracted with EtOAc (2X) with no dilution of the aqueous phase. The organic extracts were combined, dried (MgSO$_4$), and concentrated *in vacuo*. The clear residue was then purified by silica flash chromatography (150 g, 10% EtOAc:Hex) to give a thin, clear, colorless liquid. Yield: 5.07 g, 19.70 mmol, 86%. ^{1}H NMR (500 MHz, Chloroform-d) δ 4.12 (q, J = 7.1 Hz, 2H), 3.99 (s, 2H), 2.81 (t, J = 11.4 Hz, 2H), 2.41 (tt, J = 11.0, 3.6 Hz, 1H), 1.85 (d, J = 12.3 Hz, 2H), 1.60 (qd, J = 12.6, 12.1, 4.4 Hz, 2H), 1.43 (s, 9H), 1.23 (t, J = 6.8 Hz, 3H).

Tert-Butyl 4-((2-(pyridin-4-yl)ethyl)carbamoyl)piperidine-1-carboxylate **(35)**. The boc-protected ethyl isonipecotate **34** (3.00 g, 11.66 mmol) was dissolved in EtOH (20 mL) and 10% aq. NaOH (10 mL) and stirred at RT for 4 h, after which time the EtOH was stripped off *in vacuo* and the solution neutralized with conc. HCl at 0 ºC *as quickly as possible*, and the resulting precipitate was collected over a filter and washed with cold

H₂O, then dried via aspirator, and finally high vacuum to provide the desired carboxylic acid as a white powder. Yield: 2.43 g, 10.60 mmol, 91%.

The following was added to DCM: 1-(tert-butoxycarbonyl)piperidine-4-carboxylic acid (600 mg, 2.62 mmol), DIPEA (1.37 mL, 7.85 mmol), EDC (552 mg, 2.88 mL), HOBT (441 mg, 2.88 mmol), and pyridylethylamine (0.34 mL, 2.88 mL). The solution was stirred at RT for 17 h, at which time the DCM was stripped off and 10% aq. sodium carbonate was added. Material was extracted out with EtOAc (3X). The organic extractions were pooled, dried over magnesium sulfate, and concentrated *in vacuo*. The residue was taken up in a small amount of EtOAc and diethyl ether was added. The precipitate was collected over a filter and washed with diethyl ether to give the title compound as an off-white solid. Yield: 634 mg, 1.9 mmol, 73%. ^{1}H NMR (400 MHz, Chloroform-d) δ 8.49 (d, J = 5.9 Hz, 2H), 7.09 (d, J = 5.9 Hz, 2H), 5.59 (t, J = 5.3 Hz, 1H), 4.22 – 3.95 (m, 2H), 3.52 (q, J = 6.8 Hz, 2H), 2.81 (t, J = 6.9 Hz, 2H), 2.68 (t, J = 12.5 Hz, 2H), 2.14 (tt, J = 11.6, 3.8 Hz, 1H), 1.77 – 1.65 (m, 2H), 1.56 (qd, J = 12.1, 4.3 Hz, 2H), 1.43 (s, 9H).

N-(2-(Pyridin-4-yl)ethyl)piperidine-4-carboxamide dihydrochloride (**36**). The Boc-protected **35** (575 mg, 1.72 mmol) was suspended in Et₂O at RT, and 4M HCl in dioxane (6 mL, 24 mmol) was added. The mixture was stirred at RT for 30 min, at which time the organic solution was decanted off and the solid material collected and dried *in vacuo*. The title compound was thus obtained as a tan powder. Yield: 448 mg, 1.6 mmol, 97%. ^{1}H NMR (400 MHz, DMSO-d6) δ 9.22 (bs, 1H), 8.91 (bs, 1H), 8.79 (d, J = 6.4 Hz, 2H), 8.18 (t, J = 5.6 Hz, 1H), 7.89 (d, J = 6.3 Hz, 2H), 3.40 (q, J = 6.2 Hz, 2H), 3.15 (d, J

= 12.6 Hz, 2H), 3.00 (t, *J* = 6.4 Hz, 2H), 2.83 – 2.69 (m, 2H), 2.39 – 2.26 (m, 1H), 1.80 –

1.59 (m, 4H). TOF ES+ MS: (M + H) 234.1, (M + Na) 256.1.

(R)-1-(4-Chlorobenzyl)pyrrolidine-2-carboxylic acid **(R-38)**. *L*-proline (500 mg,

4.34 mmol) was dissolved in IPA (20 mL) in a dry flask, followed by the addition of

KOH pellets (730 mg, 13.03 mmol). Reaction was stirred at 40 °C while an IPA (5 mL)

solution of 4-chlorobenzyl chloride (770 mg, 4.78 mmol) was added via syringe pump

over 3 h. Reaction was allowed to stir an additional 6 h, after which time the reaction was

allowed to cool to RT. The solution was then acidified to pH 5 and DCM was added and

the reaction stirred for 16 hours at RT. After this time, the precipitate was filtered off and

washed with DCM (2X), and the combined organic filtrate extracts were concentrated to

provide a light yellow solid. Dried under high vacuum. Yield: 956 mg, 3.77 mmol, 87%.

^{1}H NMR (400 MHz, Chloroform-d) δ 10.24 (s, 1H), 7.89 (d, *J* = 9.0 Hz, 1H), 7.35 (s,

4H), 6.62 (s, 2H), 4.75 (s, 4H), 1.58 (s, 2H). TOF ES– MS: (M – H) 238.1.

(S)-1-(4-Chlorobenzyl)pyrrolidine-2-carboxylic acid **(S-38)**. *D*-proline (500 mg,

4.34 mmol) was dissolved in IPA (20 mL) in a dry flask, followed by the addition of

KOH pellets (730 mg, 13.03 mmol). Reaction was stirred at 40 °C while an IPA (5 mL)

solution of 4-chlorobenzyl chloride (770 mg, 4.78 mmol) was added via syringe pump

over 3 h. Reaction was allowed to stir an additional 6 h, after which time the reaction was

allowed to cool to RT. The solution was then acidified to pH 5 and DCM was added and

the reaction stirred for 16 hours at RT. After this time, the precipitate was filtered off and

washed with DCM (2X), and the combined organic filtrate extracts were concentrated to

provide a light yellow solid. Taken directly into the subsequent step without further characterization. Yield: 923 mg, 3.64 mmol, 84%. TOF ES– MS: (M – H) 238.1.

(R)-1-(4-Chlorobenzoyl)pyrrolidine-2-carboxylic acid **(R-39)**. *L*-proline (800 mg, 6.95 mmol) was dissolved in IPA (30 mL) in a dry flask, followed by the addition of KOH pellets (1.17 g, 20.85 mmol). Reaction was stirred at RT while an IPA solution of 4-chlorobenzoyl chloride (1.34 g, 7.64 mmol) was added via syringe pump over 1 h. Reaction was allowed to stir an additional 1 h, after which time the reaction was cooled to 0°C. The solution was then acidified to pH 5, DCM was added, and the reaction stirred for 12 hours at RT. After this time, the precipitate was filtered off and washed with DCM (2X), and the combined organic filtrate extracts were concentrated to provide an off-white solid without further purification. Taken directly into the subsequent step without further characterization. Yield: 1.58 g, 6.23 mmol, 90%. TOF ES– MS: (M – H) 252.0.

(S)-1-(4-Chlorobenzoyl)pyrrolidine-2-carboxylic acid **(S-39)**. *D*-proline (800 mg, 6.95 mmol) was dissolved in IPA (30 mL) in a dry flask, followed by the addition of KOH pellets (1.17 g, 20.85 mmol). Reaction was stirred at RT while an IPA solution of 4-chlorobenzoyl chloride (1.34 g, 7.64 mmol) was added via syringe pump over 1 h. Reaction was allowed to stir an additional 1 h, after which time the reaction was cooled to 0°C. The solution was then acidified to pH 5, DCM was added, and the reaction stirred for 13 hours at RT. After this time, the precipitate was filtered off and washed with DCM (2X), and the combined organic filtrate extracts were concentrated to provide an off-

white solid without further purification. Taken directly into the subsequent step without further characterization. Yield: 1.62 g, 6.39 mmol, 92%. TOF ES– MS: (M – H) 252.0.

(R)-Ethyl 1-(1-(4-chlorobenzoyl)pyrrolidine-2-carbonyl)piperidine-4-carboxylate
(R-40). (R)-1-(4-chlorobenzoyl)pyrrolidine-2-carboxylic acid (430 mg, 1.79 mmol) and TEA (750 µL, 5.38 mmol) were dissolved in DCM (10 mL) followed by HATU (720 mg, 1.88 mmol), ethyl isonipecotate (290 µL, 1.88 mmol), and 3Å MS. The reaction was allowed to stir at RT for 17 h, at which time the DCM was removed and the residue taken up in a 1:1 solution of EtOAc:Et$_2$O and washed with 0.5 M aq. HCl (2X), H$_2$O (1X), 10% aq. Na$_2$CO$_3$ (3X), and brine (1X). The solution was then dried (MgSO$_4$) and concentrated, and the residue was purified via silica flash chromatography (100 g silica, 50% to 100% EtOAc:Hex) to give the desired product as a clear oil. Yield: 420 mg, 1.11 mmol, 62%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.52 (d, *J* = 8.1 Hz, 2H), 7.35 – 7.30 (m, 2H), 5.04 (dt, *J* = 14.6, 7.4 Hz, 1H), 4.50 – 4.44 (m, 1H), 4.40 (m, 0.5H), 4.27 (dd, *J* = 9.6, 3.9 Hz, 0.5H), 4.17 – 4.09 (m, 2H), 4.04 (d, *J* = 13.6 Hz, 0.5H), 3.94 (d, *J* = 13.7 Hz, 0.5H), 3.80 (tt, *J* = 11.5, 5.9 Hz, 0.5H), 3.67 (q, *J* = 7.8 Hz, 1H), 3.50 (ddd, *J* = 10.5, 7.2, 4.1 Hz, 1H), 3.41 – 3.30 (m, 0.5H), 3.17 (dt, *J* = 13.8, 6.9 Hz, 0.5H), 3.05 – 2.95 (m, 0.5H), 2.90 – 2.76 (m, 0.5H), 2.62 – 2.48 (m, 1H), 2.22 (ddt, *J* = 22.4, 15.3, 7.9 Hz, 1H), 2.06 – 1.81 (m, 7H), 1.64 (ddd, *J* = 27.7, 15.2, 8.6 Hz, 2H), 1.27 – 1.21 (m, 3H).

(S)-Ethyl 1-(1-(4-chlorobenzoyl)pyrrolidine-2-carbonyl)piperidine-4-carboxylate
(S-40). (S)-1-(4-chlorobenzoyl)pyrrolidine-2-carboxylic acid (490 mg, 2.05 mmol) and TEA (860 µL, 6.15 mmol) were dissolved in DCM (10 mL) followed by HATU (820 mg,

2.15 mmol), ethyl isonipecotate (330 μL, 2.15 mmol), and 3Å MS. The reaction was allowed to stir at RT for 16 h, at which time the DCM was removed and the residue taken up in a 1:1 solution of EtOAc:Et$_2$O and washed with 0.5 M aq. HCl (2X), H$_2$O (1X), 10% aq. Na$_2$CO$_3$ (3X), and brine (1X). The solution was then dried (MgSO$_4$) and concentrated, and the residue was purified via silica flash chromatography (100 g silica, 50% to 100% EtOAc:Hex) to give the desired product as a clear oil. Yield: 340 mg, 0.91 mmol, 44%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.52 (d, J = 8.1 Hz, 2H), 7.33 (m, 2H), 5.04 (dt, J = 14.6, 7.4 Hz, 1H), 4.49 – 4.40 (m, 1H), 4.27 (d, J = 9.6 Hz, 0.5H), 4.13 (dp, J = 13.9, 7.4 Hz, 2H), 4.04 (d, J = 13.6 Hz, 0.5H), 3.94 (d, J = 13.7 Hz, 0.5H), 3.80 (tt, J = 11.5, 5.9 Hz, 0.5H), 3.67 (q, J = 7.8 Hz, 1H), 3.50 (ddd, J = 10.5, 7.2, 4.1 Hz, 1H), 3.39 – 3.29 (m, 0.5H), 3.17 (dt, J = 13.8, 6.9 Hz, 0.5H), 3.05 – 2.94 (m, 0.5H), 2.88 – 2.80 (m, 0.5H), 2.61 – 2.48 (m, 1H), 2.22 (ddt, J = 22.4, 15.3, 7.9 Hz, 1H), 2.10 – 1.80 (m, 4H), 1.64 (m, 2H), 1.30 – 1.19 (m, 3H).

Methyl 2-(4-chlorophenoxy)benzoate **(42)**. Methyl salicylate (0.40 mL, 3.86 mmol), pyridine (0.75 mL, 9.26 mmol), (4-chlorophenyl)boronic acid (531 mg, 3.40 mmol), and Cu(OAc)$_2$ (617 mg, 3.40 mmol) were added to MeOH (10 mL) with 4Å MS and allowed to stir at RT for 5 d. After this time, the reaction mixture was filtered over celite and the filtrate was concentrated. The resulting residue was taken up in EtOAc and washed with 5% aq. NaOH (3X) to remove residual copper, then H$_2$O (3X), 1 M aq. HCl (3X), and brine (1X), then the organic phase as dried (MgSO$_4$) and concentrated, and the resulting residue was purified via silica flash chromatography (1% to 10% EtOAc:Hex) to provide the desired ether as colorless clear oil. Yield: 233 mg, 0.89 mmol, 23%. [1]H

NMR (500 MHz, Chloroform-d) δ 7.90 (dd, *J* = 7.8, 1.6 Hz, 1H), 7.46 (ddd, *J* = 9.0, 7.9, 1.7 Hz, 1H), 7.29 – 7.24 (m, 2H), 7.23 – 7.17 (m, 1H), 6.96 (d, *J* = 8.3 Hz, 1H), 6.92 – 6.84 (m, 2H), 3.94 (s, 3H).

Methyl 2-((4-chlorophenyl)amino)benzoate **(43a)**. Methyl salicylate (1.50 mL, 11.57 mmol), *N,N*-Bis(triflyl)aniline (4.76 g, 13.31 mmol), and DIPEA (6.06 mL, 34.70 mmol) were dissolved into anhydrous DMF (20 mL) in dry glassware under N_2 at RT. The solution was stirred for 18 h at RT, at which time the reaction was diluted with 2:1 EtOAc:Et_2O and washed with H_2O (5X) and brine (1X), then dried ($MgSO_4$) and concentrated. The residue was purified via silica flash chromatography (80 g silica, 5% EtOAc:Hex) to give methyl 2-((((trifluoromethyl)sulfonyl)oxy)benzoate as a clear, colorless oil. Yield: 2.76 g, 9.72 mmol, 84%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.09 (d, *J* = 7.8 Hz, 1H), 7.63 (t, *J* = 7.8 Hz, 1H), 7.48 (t, *J* = 7.6 Hz, 1H), 7.31 (d, *J* = 8.3 Hz, 1H), 3.97 (s, 3H).

A Schlenck flask was filled with toluene (60 mL, not anhydrous) and then charged sequentially with the following reagents: methyl 2-((((trifluoromethyl)sulfonyl)oxy)benzoate (900 mg, 3.17 mmol), 4-chloroaniline (485 mg, 3.80 mmol), Cs_2CO_3 (1444 mg, 4.43 mmol), Pd(OAc)$_2$ (36 mg, 0.16 mmol), and DPPP (104 mg, 0.25 mmol). The mixture was then thoroughly sparged with argon, and the flask was fitted with a condenser and then purged and filled with an argon atmosphere. The reaction was stirred at reflux under argon for 6 h, then allowed to cool to RT and stir for 12 h. At this time, the reaction was diluted with EtOAc and washed with 1M aq. HCl (3X) and brine (1X), dried ($MgSO_4$), and concentrated. The residue was purified via

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silica flash chromatography (40 g silica, 1% to 5% EtOAc:Hex gradient) to yield the desired compound as a clear, vaguely yellow oil. Yield: 710 mg, 2.71 mmol, 86%. ^{1}H NMR (500 MHz, Chloroform-d) δ 9.46 (s, 1H), 7.98 (d, J = 9.0 Hz, 1H), 7.37 − 7.26 (m, 3H), 7.24 − 7.16 (m, 3H), 6.77 (t, J = 7.6 Hz, 1H), 3.92 (d, J = 1.1 Hz, 3H).

Methyl 2-(phenylamino)benzoate **(43b)**. Methyl salicylate (1.50 mL, 11.57 mmol), *N,N*-Bis(triflyl)aniline (4.76 g, 13.31 mmol), and DIPEA (6.06 mL, 34.70 mmol) were dissolved into anhydrous DMF (20 mL) in dry glassware under N_2 at RT. The solution was stirred for 18 h at RT, at which time the reaction was diluted with 2:1 EtOAc:Et$_2$O and washed with H_2O (5X) and brine (1X), then dried (MgSO$_4$) and concentrated. The residue was purified via silica flash chromatography (80 g silica, 5% EtOAc:Hex) to give methyl 2-((((trifluoromethyl)sulfonyl)oxy)benzoate as a clear, colorless oil. Yield: 2.76 g, 9.72 mmol, 84%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.09 (d, J = 7.8 Hz, 1H), 7.63 (t, J = 7.8 Hz, 1H), 7.48 (t, J = 7.6 Hz, 1H), 7.31 (d, J = 8.3 Hz, 1H), 3.97 (s, 3H).

A Schlenck flask was filled with toluene (30 mL, not anhydrous) and then charged sequentially with the following reagents: methyl 2-((((trifluoromethyl)sulfonyl)oxy)benzoate (440 mg, 1.63 mmol), aniline (178 μL, 1.94 mmol), Cs$_2$CO$_3$ (743 mg, 2.28 mmol), Pd(OAc)$_2$ (18 mg, 0.08 mmol), and DPPP (67 mg, 0.16 mmol). The mixture was then thoroughly sparged with argon, and the flask was fitted with a condenser and then purged and filled with an argon atmosphere. The reaction was stirred at reflux under argon for 6 h, then allowed to cool to RT and stir for 12 h. At this time, the reaction was diluted with EtOAc and washed with 1M aq. HCl

(3X) and brine (1X), dried (MgSO$_4$), and concentrated. The residue was purified via silica flash chromatography (40 g silica, 1% to 5% EtOAc:Hex gradient) to yield the desired compound as a clear, somewhat yellow oil. Yield: 377 mg, 1.66 mmol, 57%. ^{1}H NMR (500 MHz, Chloroform-d) δ 9.47 (s, 1H), 7.97 (d, J = 8.0 Hz, 1H), 7.33 (d, J = 16.2, 7.3 Hz, 3H), 7.26 (d, J = 7.8 Hz, 3H), 7.10 (t, J = 7.3 Hz, 1H), 6.74 (t, J = 7.5 Hz, 1H), 3.91 (s, 3H).

Ethyl   1-(2-((4-chlorophenyl)amino)benzoyl)piperidine-4-carboxylate   (**44a**). Methyl 2-((4-chlorophenyl)amino)benzoate (650 mg, 2.48 mmol) was dissolved in 200 proof EtOH (15 mL) at RT and 10% aq. NaOH (6 mL) was added. The reaction was allowed to stir at RT for 5 h, at which time the solvent was stripped off *in vacuo* and 1M aq. HCl was added to the residue. The solution was then chilled in an ice bath and acidified to pH 1, and the resulting white precipitate was collected over a filter, washed with 1M aq. HCl, and dried via aspirator and high vacuum to yield a white powder. No further purification necessary. Yield: 581 mg, 2.35 mmol, 94%. ^{1}H NMR (500 MHz, Chloroform-d) δ 9.27 (s, 1H), 8.06 (dd, J = 8.1, 1.5 Hz, 1H), 7.38 (ddd, J = 8.6, 7.1, 1.6 Hz, 1H), 7.35 − 7.31 (m, 2H), 7.23 − 7.19 (m, 2H), 7.17 (d, J = 8.5 Hz, 1H), 6.79 (t, J = 7.5 Hz, 1H).

2-((4-chlorophenyl)amino)benzoic acid (370 mg, 1.49 mmol) was then dissolved in DCM (15 mL), followed by ethyl isonipecotate (253 μL, 1.64 mmol), TEA (625 μL, 4.48 mmol), EDC·HCl (315 mg, 1.64 mmol), and HOBT (252 mg, 1.64 mmol). The reaction was stirred at RT for 16 h, at which time the reaction was diluted with 2:1 EtOAc:Et$_2$O and washed with 1 M aq. HCl (2X), 10% aq. Na$_2$CO$_3$ (3X), and brine (1X),

dried (MgSO$_4$) and concentrated. The residue was purified via silica flash chromatography (12 g silica, 5% to 20% EtOAc:Hex) to provide the product as a yellow oil. Yield: 129 mg, 0.333 mmol, 22%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.32 (d, *J* = 8.2 Hz, 1H), 7.25 – 7.15 (m, 4H), 7.02 (d, *J* = 8.7 Hz, 2H), 6.89 (t, *J* = 7.4 Hz, 1H), 4.30 (bs, 2H), 4.15 (q, *J* = 7.1 Hz, 2H), 3.09 (t, *J* = 11.1 Hz, 2H), 2.56 (tt, *J* = 10.6, 4.0 Hz, 1H), 1.95 (bs, 2H), 1.75 – 1.67 (m, 2H), 1.25 (t, *J* = 7.1 Hz, 3H).

Methyl 2-((4-chlorophenyl)(methyl)amino)benzoate **(45)**. Methyl 2-((4-chlorophenyl)amino)benzoate (150 mg, 0.57 mmol) was dissolved in anhydrous DMF (8 mL) under N$_2$. The solution was then charged with granular K$_2$CO$_3$ (158 mg, 1.15 mmol). MeI (40 μL, 0.63 mmol), dissolved in 1 mL anhydrous DMF, was added dropwise to the solution at RT under N$_2$, and the reaction was allowed to stir for 12 h at RT. After this time, the reaction was diluted in 2:1 EtOAc:Et$_2$O and washed with H$_2$O (3X), 10% aq. Na$_2$S$_2$O$_3$ (1X), and brine (1X), then dried (MgSO$_4$) and concentrated. The residue was purified via silica flash chromatography (25 g silica, 1% to 10% EtOAc:Hex) to afford the desired compound as a colorless oil taken crude into the next reaction. Yield: 139 mg, 0.50 mmol, 88%. TOF ES+ MS: (M + H) 276.1

Methyl 2-((4-chlorobenzyl)amino)benzoate **(47a)**. Methyl 2-aminobenzoate (500 mg, 3.31 mmol) was dissolved in anhydrous DMF (15 mL) at RT under N$_2$. *Tert*-BuOK (408 mg, 3.64 mmol) was then added and the solution became yellow, and this was allowed to stir at RT for 30 min, at which point 4-chlorobenzyl chloride (0.45 mL, 3.47 mmol) was added, and the solution lost color. The reaction was allowed to stir for 18 h

under N_2, at which time the reaction was diluted in EtOAc and washed with 10% aq. $NaHCO_3$ (1X), H_2O (5X), and brine (1X), then dried ($MgSO_4$) and concentrated. The residue was purified by silica flash chromatography (40 g silica, 1% to 30% EtOAc:Hex) to provide the desired compound as a colorless oil. Yield: 1.37 g, 2.88 mmol, 87%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.20 (t, J = 5.7 Hz, 1H), 7.95 (d, J = 8.0 Hz, 1H), 7.31 (q, J = 7.8, 7.1 Hz, 5H), 6.63 (t, J = 7.6 Hz, 1H), 6.58 (d, J = 8.5 Hz, 1H), 4.43 (d, J = 5.7 Hz, 2H), 3.88 (s, 3H). TOF ES+ MS: (M + H) 276.1.

Methyl 2-(benzylamino)benzoate **(47b)**. To anhydrous DMF in dry glassware was added methyl 2-aminobenzoate (280 mg, 1.85 mmol), benzyl bromide (330 mg, 1.94 mmol), and Cs_2CO_3 (1.50 g, 4.62 mmol). The reaction was stirred at 85 °C for 5 days, after which time the reaction was cooled to RT and diluted with 1:1 EtOAc:Et_2O and washed with H_2O (3X) and brine (1X), then dried ($MgSO_4$) and concentrated. The residue was purified via silica flash chromatography (5% EtOAc:Hex isochratic) to separate product from unreacted starting material. Yield: 138 mg, 0.57 mmol, 31%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.18 (s, 1H), 7.94 (d, J = 8.0 Hz, 1H), 7.40 – 7.25 (m, 6H), 6.65 (d, J = 8.5 Hz, 1H), 6.61 (t, J = 7.6 Hz, 1H), 4.46 (d, J = 5.6 Hz, 2H), 3.87 (s, 3H).

2-((4-Chlorophenethyl)amino)benzoic acid **(49)**. 2-Bromobenzoate (200 mg, 1.00 mmol), 2-(4-chlorophenyl)ethanamine (290 mg, 1.86 mmol), K_2CO_3 (260 mg, 1.86 mmol), copper powder (12 mg, 0.19 mmol), and copper (I) oxide (13 mg, 0.09 mmol) were added to ethylene glycol dimethyl ether (5 mL) in a dry pressure tube. The mixture

was sparged and the headspace purged with argon, and the reaction was stirred at 130 °C for 24 h. After cooling to RT, the reaction was diluted with EtOAc and H_2O and filtered through celite. The filtrate was acidified with conc. HCl and material was extracted with EtOAc (3X). The combined extracts were dried ($MgSO_4$) and concentrated. Purification of the resulting residue was accomplished via silica flash chromatography (25 g silica, 10% EtOAc:Hex with 1% formic acid) to give a white powder. Yield: 120 mg, 0.44 mmol, 44%. ^1H NMR (500 MHz, Chloroform-d) δ 8.06 (d, $J = 8.4$ Hz, 1H), 8.00 (d, $J = 8.0$ Hz, 1H), 7.41 (t, $J = 7.8$ Hz, 1H), 7.32 (d, $J = 8.2$ Hz, 2H), 7.21 (d, $J = 8.2$ Hz, 2H), 6.71 (d, $J = 8.5$ Hz, 1H), 6.64 (t, $J = 7.5$ Hz, 1H), 3.47 (t, $J = 7.1$ Hz, 2H), 2.97 (t, $J = 7.0$ Hz, 2H).

2-(4-Chlorobenzoyl)benzoic acid **(51)**. Aluminum chloride (9.0 g, 67.5 mmol) was carefully added at RT to chlorobenzene (50 mL) in dry glassware with a water separator for dense solvents, followed by phthalic anhydride (5.0 g, 33.8 mmol). The reaction was then refluxed under N_2 for 1.5 h, over which time it took on a deep red color. *Note:* longer reaction times result in rapid accumulation of a second, internal acylation. The reaction was then allowed to cool to RT over 1 h and was quenched by the slow and careful addition of 1 M aq. HCl and allowed to stir another 1 h, over which time color was lost and a white precipitate formed. The reaction was then taken up in EtOAc and washed with 1M aq. HCl (1X), then the solvent was removed *in vacuo*. The residue was then dissolved in 10% aq. NaOH, the resulting precipitate was filtered off, and the filtrate was washed with EtOAc (2X) before being chilled in an ice bath and acidified to pH 1 with conc. HCl. The resulting precipitate was collected over a filter to give the

desired product as a white powder. No further purification necessary. Yield: 6.34 g, 24.31 mmol, 72%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.08 (d, *J* = 7.8 Hz, 1H), 7.70 − 7.61 (m, 3H), 7.58 (tt, *J* = 7.7, 1.2 Hz, 1H), 7.36 (dd, *J* = 12.2, 7.9 Hz, 3H). TOF ES− MS: (M − H) 259.0.

2-(4-Chlorobenzyl)benzoic acid (**52**). 2-(4-chlorobenzoyl)benzoic acid **51** (200 mg, 0.77 mmol) was added to 28% aq. ammonia (10 mL), followed by zinc (600 mg, 9.21 mmol) and $CuSO_4 \cdot 5H_2O$ (80 mg, 0.31 mmol). This mixture was stirred at reflux for 48 h, with 2 mL recharges of 28% aq. ammonia every 6 h, barring 12 h at night. After this time, the reaction was allowed to cool to RT and the solution was decanted off the solid precipitate and concentrated *in vacuo*. The residue was taken up in H_2O and acidified to pH < 1 by conc. HCl, then extracted 1X with EtOAc. The extract was dried ($MgSO_4$) and concentrated, and the residue was crystallized from EtOAc to give tan crystals that were used immediately in the subsequent reaction. Yield: 62 mg, 0.25 mmol, 33%. TOF ES− MS: (M − H) 245.0.

Methyl 6-chloro-9H-carbazole-1-carboxylate (**55**). 9H-carbazole-1-carboxylic acid **54** (200 mg, 0.95 mmol) − prepared in a manner similar to that reported[120] − was stirred in refluxing sat. methanolic HCl for 1 h. The reaction was then allowed to cool and was concentrated. The residue was dissolved in EtOAc and washed with sat. aq. Na_2CO_3 (1X), then dried ($MgSO_4$), and concentrated. The residue was then purified by silica flash chromatography (10 g silica, 10% EtOAc/Hex) to give a white powder. Yield: 196 mg, 0.87 mmol, 92%. ^{1}H NMR (500 MHz, Chloroform-d) δ 9.94 (s, 1H), 8.26 (d, *J* =

7.6 Hz, 1H), 8.14 – 8.04 (m, 2H), 7.57 – 7.46 (m, 2H), 7.34 – 7.28 (m, 1H), 7.26 (t, J =
7.7 Hz, 1H), 4.03 (s, 3H).

Methyl 9H-carbazole-1-carboxylate (90 mg, 0.40 mmol) was stirred in DCM (15
mL) at 0 °C under N_2 while SO_2Cl_2 was added dropwise. The reaction was allowed to stir
at 0 °C for 4 h. At this time, the reaction was diluted with DCM and washed with 10%
aq. Na_2CO_3 (2X) and brine (1X), then dried (Na_2SO4) and concentrated. The residue was
purified via silica flash chromatography (10 g silica, 10 to 30% EtOAc/Hex) to give a
white powder. Yield: 75 mg, 0.29 mmol, 72%. ^{1}H NMR (500 MHz, Chloroform-d) δ 9.93
(s, 1H), 8.21 (d, J = 7.7 Hz, 1H), 8.10 (d, J = 7.6 Hz, 1H), 8.04 (s, 1H), 7.44 – 7.41 (m,
2H), 7.27 (t, J = 7.7 Hz, 1H), 4.03 (s, 3H). ^{1}H NMR (500 MHz, Benzene-d6) δ 9.70 (s,
1H), 8.03 (d, J = 7.6 Hz, 1H), 7.84 (d, J = 1.9 Hz, 1H), 7.70 (d, J = 7.6 Hz, 1H), 7.17 (dd,
J = 8.6, 2.0 Hz, 1H), 6.92 (t, J = 7.7 Hz, 1H), 6.46 (d, J = 8.6 Hz, 1H), 3.48 (s, 3H).

Ethyl   1-(2-oxo-1,2-dihydroquinolin-4-yl)piperidine-4-carboxylate **(57)**.   4-
chloroquinolin-2(1H)-one   was   prepared   according   to   literature   precedent.[124, 125]   4-
chloroquinolin-2(1H)-one (500 mg, 2.78 mmol), ethyl isonipecotate **26** (0.52 mL), NaI
(420 mg, 2.78 mmol), and Na_2CO_3 (885 mg, 8.35 mmol) were added to DMF (10 mL) at
RT under N_2 and stirred at 80 °C for 48 h. The reaction was then cooled to RT, at which
time it was diluted in 1:1 EtOAc:Et_2O and washed with 1 M HCl (3X), H_2O (3X), 10%
aq. Na_2CO_3 (3X), and brine (1X). The organic phase was dried ($MgSO_4$) and
concentrated. The residue was purified by silica flash chromatography (45 g silica, 20 to
80% EtOAc/Hex) to give a white microcrystalline solid. Yield: 567 mg, 1.89 mmol, 68%.
^{1}H NMR (500 MHz, Chloroform-d) δ 12.43 (s, 1H), 7.69 (d, J = 8.2 Hz, 1H), 7.48 – 7.40

(m, 2H), 7.17 (t, J = 7.2 Hz, 1H), 6.13 (s, 1H), 4.18 (q, J = 7.1 Hz, 2H), 3.54 (d, J = 12.4 Hz, 2H), 2.82 (t, J = 10.8 Hz, 2H), 2.54 (ddd, J = 15.0, 10.8, 4.1 Hz, 1H), 2.13 – 2.07 (m, 2H), 2.06 – 1.98 (m, 2H), 1.28 (t, J = 7.1 Hz, 3H).

Ethyl 1-(1-(4-chlorophenyl)-2-oxo-1,2-dihydroquinolin-4-yl)piperidine-4-carboxylate (**58**). Ethyl 1-(2-oxo-1,2-dihydroquinolin-4-yl)piperidine-4-carboxylate **57** (490 mg, 1.64 mmol) was dissolved in DCM (10 mL) with 4 Å MS, followed by the addition of 4-chlorophenylboronic acid (510 mg, 3.27 mmol), pyridine (0.26 mL, 3.27 mmol), TEA (0.46 mL, 3.27 mmol), and Cu(OAc)$_2$ (590 mg, 3.27 mmol). The reaction was stirred at RT for 4 d, over which time the reaction became increasingly green with precipitate. After this time, the reaction was diluted with more DCM and washed with 10% aq. NaOH (3X), 10% aq. NaHCO$_3$ (1X), and brine (1X), then dried (MgSO$_4$) and concentrated. The resulting residue was purified by silica flash chromatography (25 g, 5 to 50% EtOAc/Hex) to give a white solid. Yield: 480 mg, 1.18 mmol, 72%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.79 (d, J = 8.0 Hz, 1H), 7.56 (d, J = 8.3 Hz, 2H), 7.32 (t, J = 7.8 Hz, 1H), 7.23 (d, J = 8.3 Hz, 2H), 7.19 (t, J = 7.6 Hz, 1H), 6.66 (d, J = 8.5 Hz, 1H), 6.21 (s, 1H), 4.21 (q, J = 7.1 Hz, 2H), 3.55 (d, J = 12.2 Hz, 2H), 2.84 (t, J = 11.5 Hz, 2H), 2.57 (ddd, J = 15.1, 10.9, 4.1 Hz, 1H), 2.16 – 2.09 (m, 2H), 2.09 – 1.96 (m, 2H), 1.31 (t, J = 7.1 Hz, 3H).

Ethyl 1-(1-(4-chlorobenzyl)-2-oxo-1,2-dihydroquinolin-4-yl)piperidine-4-carboxylate (**59**). Ethyl 1-(2-oxo-1,2-dihydroquinolin-4-yl)piperidine-4-carboxylate **58** (95 mg, 0.32 mmol) was dissolved in anhydrous DMF (5 mL) under nitrogen and cooled

to 0 °C. KO*t*Bu (43 mg, 0.38 mmol) was added, which elicited a slight color change. 4-chlorobenzyl chloride **4a** (49 µL, 0.38 mmol) was then added and the reaction was allowed to stir at RT for 15 h. At this time, the reaction was diluted with 2:1 EtOAc:Et$_2$O and washed with 1M aq. HCl (3X), H$_2$O (3X), 10% aq. Na$_2$CO$_3$ (3X), and brine (1X), then dried (MgSO$_4$) and concentrated. Residue was purified via silica flash chromatography (10 g silica, 1% to 30% EtOAc:Hex). Yield: 117 mg, 0.28 mmol, 87%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.79 (d, *J* = 7.9 Hz, 1H), 7.40 (t, *J* = 7.7 Hz, 1H), 7.28 − 7.23 (m, 2H), 7.17 (dd, *J* = 13.3, 8.2 Hz, 4H), 6.24 (s, 1H), 5.48 (s, 2H), 4.20 (q, *J* = 7.3 Hz, 2H), 3.52 (d, *J* = 12.4 Hz, 2H), 2.82 (t, *J* = 11.8 Hz, 2H), 2.55 (tt, *J* = 11.0, 8.8, 4.0 Hz, 1H), 2.06 (ddd, *J* = 31.9, 19.5, 6.8 Hz, 4H), 1.30 (t, *J* = 7.4 Hz, 3H).

Ethyl 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carboxylate **(70)**. The indole carboxylic acid **4a** (982 mg, 3.44 mmol) was added to DCM (15 mL), followed by ethyl isonipecotate **26** (0.80 mL, 5.16 mmol), DIPEA (1.80 mL, 10.31 mmol), EDC·HCl (800 mg, 4.12 mmol), and HOBT (630 mg, 4.12 mmol). The reaction was allowed to stir at RT for 24 h, after which time it was diluted with 1:1 EtOAc:Et$_2$O and washed with 1M HCl (5X), H$_2$O (5X), 10% aq. Na$_2$CO$_3$ (5X) and brine (1X), then dried (MgSO$_4$) and concentrated. The residue can then be crystallized from a two-phase organic solution of EtOAc and Hex to give crystals. Yield: 1.22 g, 2.88 mmol, 83%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.66 (d, *J* = 9.1 Hz, 1H), 7.36 (d, *J* = 8.3 Hz, 1H), 7.30 − 7.27 (m, 1H), 7.25 − 7.20 (m, 2H), 7.19 − 7.14 (m, 1H), 7.08 − 6.97 (m, 2H), 6.68 − 6.61 (m, 1H), 5.49 (d, *J* = 2.5 Hz, 2H), 4.49 (bs, 2H), 4.17 (qd, *J* = 7.1, 2.5 Hz, 2H), 3.08

– 2.95 (m, 2H), 2.51 (tq, *J* = 9.4, 3.6 Hz, 1H), 1.97 – 1.55 (m, 4H), 1.28 (td, *J* = 7.2, 2.5 Hz, 3H).

1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carboxylic acid **(71)**. The ethyl ester **70** (1.22 g, 2.88 mmol) was dissolved in EtOH (20 mL) and 5M aq. NaOH (20 mL) and stirred at 50 °C for 48 h. Most of the solvent was then removed *in vacuo* and the remaining aqueous solution was washed with Et$_2$O (1X), then acidified to pH<1 at 0 °C with conc. HCl. The material was extracted with 1:1 EtOAc:Et$_2$O (3X), and the combined extracts were dried (MgSO$_4$) and concentrated. The resulting residue was dissolved in EtOAc and triturated with Hex, collected over a filter, and dried to give the desired carboxylic acid as white, fluffy microcrystals. Yield: 1.03 g, 2.59 mmol, 90%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.59 (d, *J* = 7.9 Hz, 1H), 7.32 (d, *J* = 8.3 Hz, 1H), 7.28 – 7.23 (m, 1H), 7.19 – 7.11 (m, 3H), 6.99 (d, *J* = 8.0 Hz, 2H), 6.59 (s, 1H), 5.42 (s, 2H), 4.38 (bs, 1H), 4.08 (bs, 1H), 2.87 (bs, 2H), 2.38 (bs, *J* = 11.0 Hz, 1H), 1.90 – 1.37 (m, 4H).

2,3-Dihydro-1H-indene-2-carboxamide **(74)**. 2-indancarboxylic acid **73** (1.4 g, 8.63 mmol) was dissolved in anhydrous DMF (20 mL), and the following was added sequentially: DIPEA (3.02 mL, 17.26 mmol), HATU (3.94 g, 10.36 mmol), and HMDS (2.17 mL, 10.36 mmol) after 30 min of stirring at RT. After stirring for 20 h at RT, the solution was taken up in EtOAc and washed with 0.5 M HCl (3X), H$_2$O (3X), and 10% aq. Na$_2$CO$_3$ (3X). The organic phase was collected and dried over MgSO$_{4s}$, and concentrated *in vacuo*. The solid residue was recrystallized from EtOH to afford the title

compound as sharp, colorless crystals. Yield: 861 mg, 5.06 mmol, 59%. ^{1}H NMR (400 MHz, DMSO-d6) δ 7.40 (bs, 1H), 7.22 – 7.15 (m, 2H), 7.14 – 7.09 (m, 2H), 6.86 (bs, 1H), 3.19 – 3.10 (m, 1H), 3.06 – 3.02 (m, 4H).

(2,3-Dihydro-1H-inden-2-yl)methanamine **(75)**. 2,3-dihydro-1H-indene-2-carboxamide **74** (55 mg, 0.341 mmol) was added to anhydrous THF (8 mL), and cooled in an ice bath under nitrogen. LAH (0.34 mL, 0.341 mmol, 1M THF soln) was added under nitrogen, and the reaction was allowed to warm to rt and stir for 7 h. At this time, the reaction was quenched by the Fieser method and the precipitate removed over a filter. The filtrate was collected and concentrated *in vacuo*, and then taken up in a small amount of THF. 4M HCl/dioxane was added, the solvent removed *in vacuo*, and the residue sonicated in Et$_2$O to afford the easily handled hydrochloride salt. Yield: ^{1}H NMR (400 MHz, Chloroform-d) δ 7.22 – 7.16 (m, 2H), 7.16 – 7.10 (m, 2H), 3.08 (dd, *J* = 15.6, 8.0 Hz, 2H), 2.80 (d, *J* = 7.1 Hz, 2H), 2.66 (dd, *J* = 15.6, 7.0 Hz, 2H), 2.54 (p, *J* = 7.4 Hz, 1H). TOF ES+ MS: (M + H) 148.1, (M + Na) 170.1.

1-(2,3-Dihydro-1H-inden-2-yl)ethanone **(77).** Acetylacetate (97 µL, 0.95 mmol) was dissolved in anhydrous DMF under N$_2$, followed by the addition of 60% oil-suspended NaH (95 mg, 2.37 mmol). This was allowed to stir for 30 min, at which time 1,2-bis(bromoethyl)benzene **76** (250 mg, 0.95 mmol) was added. The reaction was allowed to stir for 1 h at RT, then 8 h at 100 °C, then allowed to cool to RT. The reaction was quenched with sat. aq. NH$_4$Cl, diluted with EtOAc, and washed with 10% aq. NaHCO$_3$ (3X) and brine (1X), then dried (MgSO$_4$) and concentrated. The residue was

then purified via silica flash chromatography (50 g silica, 1% to 5% EtOAc:Hex) to give a very greasy solid invisible to electrospray MS. Yield: ^{1}H NMR (400 MHz, Chloroform-d) δ 7.21 − 7.17 (m, 2H), 7.17 − 7.12 (m, 2H), 3.47 − 3.37 (m, 1H), 3.20 − 3.11 (m, 4H), 2.22 (d, J = 2.6 Hz, 3H).

1-(2,3-Dihydro-1H-inden-2-yl)ethanamine **(78)**. 1-(2,3-dihydro-1*H*-inden-2-yl)ethanone **77** (44 mg, 0.275 mmol) was dissolved in methanol (2 mL), and 7 M methanolic ammonia was added (1 mL, 7 mmol). This was allowed to stir at RT for 4 h, at which time NaBH$_4$ (31 mg, 0.824 mmol) and a catalytic drop of glacial AcOH was added. Stirring continued for 14 h at RT, after which time the solvent was removed in vacuo. The residue was taken up in EtOAc and washed with a small amount of 10% aq. Na$_2$CO$_3$, dried with MgSO$_4$, and concentrated. The residue was then taken on in subsequent reactions without further purification or characterization. TOF ES+ MS: (M + H) 162.2.

Ethyl 7-methyl-1H-indole-2-carboxylate **(81)**. To a 0 °C flask charged with crushed ice (~10 g) was added *o*-toluidine **79** (6.00 mL, 55.4 mmol), followed by conc. HCl (10 mL) which elicited the precipitation of the hydrochloride salt. Dissolved in H$_2$O (30 mL), NaNO$_2$ (3.82 g, 55.4 mmol) was added dropwise slowly to the flask, and more ice was added as needed. The reaction was allowed to stir at 0 °C for 30 min, after which ~15 g of crushed ice and a 0 °C slurry of SnCl$_2$ (32 g, 166.0 mmol) in conc. HCl (10 mL) was added. The reaction was then allowed to sit in a fridge (4 °C) for 11 h, after which time the precipitate was filtered off and collected, and washed with cold brine and

hexanes. The resulting precipitate was collected over a filter and washed with cold brine and 1:1 EtOAc:Hex. Unlike other reports, no attempt was made to basify, wash, and re-convert the hydrazine to the hydrazine hydrochloride salt (due to observed stability issues). The white solid was dried thoroughly under high vacuum. Yield: 4.92 g, 31.02 mmol, 56%.

Ethyl pyruvate **80** (1.17 mL, 10.51 mmol), *o*-tolylhydrazine hydrochloride (1.5 g, 9.46 mmol), $ZnCl_2$ (7.16 g, 52.55 mmol) and TsOH (9.05 g, 52.55 mmol) were dissolved in anhydrous EtOH (15 mL) at RT in dry glassware. The reaction was refluxed for 8 h, after which the reaction was allowed to cool to RT and the resulting precipitate was filtered off and washed with EtOH. The filtrate was concentrated to give a residue that was dissolved in 1:1 EtOAc:Et_2O and washed with 10% aq. Na_2CO_3 (2X), dried ($MgSO_4$), and concentrated. The residue was then purified by silica flash chromatography (100 g silica, 20% EtOAc:Hex) to give the desired indole as a foul-smelling oil. Yield: 1.37 g, 6.72 mmol, 71%. ^{1}H NMR (400 MHz, Chloroform-d) δ 8.88 (bs, 1H), 7.53 (d, *J* = 8.0 Hz, 1H), 7.27 − 7.21 (m, 1H), 7.14 − 7.02 (m, 2H), 4.42 (qd, *J* = 7.1, 1.3 Hz, 2H), 2.52 (s, 3H), 1.42 (td, *J* = 7.1, 1.3 Hz, 3H).

Ethyl 1-(4-chlorobenzyl)-7-methyl-1H-indole-2-carboxylate **(82)**. Ethyl 7-methyl-1*H*-indole-2-carboxylate **81** (500 mg, 2.46 mmol) was dissolved in anhydrous DMF (20 mL) and granular K_2CO_3 (425 mg, 3.08 mmol) and 4-chlorobenzyl chloride (0.39 mL, 3.08 mmol) were added. The mixture was stirred for 3 d at 60 °C, after which time the incomplete reaction was terminated by dilution with EtOAc and subsequent washing with brine (3X). The organic phase was dried over $MgSO_4$, concentrated, and purified vial

silica flash chromatography (80 g silica, 40% EtOAc:Hex) to provide the title compound as white crystals. Yield: 390 mg, 1.19 mmol, 46%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.59 – 7.53 (m, 1H), 7.43 – 7.40 (m, 1H), 7.20 (d, *J* = 8.4 Hz, 2H), 7.06 – 6.97 (m, 2H), 6.78 (d, *J* = 8.2 Hz, 2H), 6.08 (s, 2H), 4.28 (q, *J* = 7.1 Hz, 2H), 2.54 (s, 3H), 1.33 (t, *J* = 7.1 Hz, 3H).

1-(4-Chlorobenzyl)-7-methyl-1H-indole-2-carboxylic acid (**83**). Ethyl 1-(4-chlorobenzyl)-7-methyl-1*H*-indole-2-carboxylate **82** (212 mg, 0.647 mmol) was dissolved in EtOH (10 mL) and 10% aq. KOH (10 mL) was added. The reaction stirred for 4 h at 70 °C, after which time as much solvent was stripped off as possible *in vacuo*. The mostly aqueous mixture was cooled in an ice bath and acidified with conc. HCl to obtain a precipitate that was collected over a filter and washed with 1 M HCl and cold H_2O. No further purification necessary. Yield: 182 mg, 0.607 mmol, 94%. ^{1}H NMR (400 MHz, DMSO-d6) δ 12.94 (s, 1H), 7.36 – 7.24 (m, 4H), 7.01 – 6.95 (m, 2H), 6.76 (d, *J* = 8.1 Hz, 2H), 6.08 (s, 2H), 2.44 (s, 3H).

Ethyl 3-methyl-1H-indole-2-carboxylate (**86**). Phenylhydrazine was prepared in a manner similar to that reported.[138] Ethyl 2-oxobutanoate **85** (500 mg, 4.90 mmol), phenylhydrazine (0.44 mL, 4.41 mmol), and TsOH (2.3 g, 12.24 mmol) were dissolved in anhydrous EtOH (15 mL) at RT in dry glassware. The reaction was refluxed for 12 h, over which time it took on a dark hue. After the reaction time, the reaction was allowed to cool to RT and the resulting precipitate was filtered off and washed with EtOH. The combined filtrate was concentrated to give a residue that was dissolved in 1:1

EtOAc:Et$_2$O and washed with 10% aq. Na$_2$CO$_3$ (2X), dried (MgSO$_4$), and concentrated. The residue was then purified by silica flash chromatography (45 g silica, 20% EtOAc:Hex) to give the desired indole as a foul-smelling oil. Yield: 646 mg, 3.18 mmol, 65%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.83 (s, 1H), 7.69 (d, J = 8.1 Hz, 1H), 7.39 (d, J = 8.3 Hz, 1H), 7.35 (d, J = 7.8 Hz, 1H), 7.16 (t, J = 7.4 Hz, 1H), 4.45 (qd, J = 7.1, 1.1 Hz, 2H), 2.64 (d, J = 1.1 Hz, 3H), 1.46 (td, J = 7.2, 1.1 Hz, 3H).

Ethyl 1-(4-chlorobenzyl)-3-methyl-1H-indole-2-carboxylate **(87)**. Anhydrous DMF (12 mL) in dry glassware was charged with ethyl 3-methyl-1*H*-indole-2-carboxylate **86** (646 mg, 3.18 mmol), 4-chlorobenzyl chloride (0.50 mL, 3.97 mmol), and granular K$_2$CO$_3$, and the mixture was stirred at 60 °C for 14 h, allowed to cool for 1 h, then diluted in 1:1 EtOAc:Et$_2$O and washed with 10% aq. Na$_2$CO$_3$ (3X), dried (MgSO$_4$), and concentrated. The residue was purified by silica flash chromatography (40 g silica, 20% EtOAc:Hex) to give the desired compound as an oil. Yield: 750 mg, 2.29 mmol, 72%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.71 (dd, J = 8.1, 1.1 Hz, 1H), 7.33 − 7.23 (m, 2H), 7.22 − 7.12 (m, 3H), 6.99 − 6.90 (m, 2H), 5.73 (s, 2H), 4.34 (qd, J = 7.2, 1.1 Hz, 2H), 2.62 (d, J = 1.1 Hz, 3H), 1.35 (td, J = 7.1, 1.1 Hz, 3H).

5-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)octahydro-1H-pyrrolo[3,4-c]pyridin-1-one **(91)**. The following were added sequentially to DMF (10 mL): 1-(4-chlorobenzyl)-1*H*-indole-2-carboxylic acid **4a** (250 mg, 0.875 mmol), DIPEA (0.46 mL, 2.64 mmol), EDC·HCl (202 mg, 1.050 mmol), and HOBT (162 mg, 1.050 mmol). This solution was allowed to stir at RT for 1 h, at which time crude octahydro-1H-pyrrolo[3,4-

c]pyridin-1-one **90** (123 mg, 0.875 mmol) was added, having been prepared through the known route.[140] Stirring continued for 12 h at RT, at which time the solution was partitioned between water and 1:1 solution of EtOAc:Et$_2$O. The organic extract was then washed with water (1X) and saturated aq. Na$_2$CO$_3$ (1X), dried with MgSO$_4$, and concentrated *in vacuo*. Purification was accomplished via silica flash chromatography (100 g silica, 100% EtOAc.) Resulting residue was crystallized from the *in vacuo* removal of DCM to give the title compound as a slightly pink solid. Yield: 200 mg, 0.49 mmol, 56%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.72 (d, *J* = 7.9 Hz, 1H), 7.37 − 7.24 (m, 4H), 7.19 (t, *J* = 7.4 Hz, 1H), 7.14 (d, *J* = 8.1 Hz, 2H), 6.93 (s, 1H), 6.31 (bs, 1H), 5.51 (s, 2H), 4.57 (d, *J* = 13.2 Hz, 1H), 4.18 (bs, 1H), 3.56 − 3.46 (m, 1H), 3.39 (dd, *J* = 13.7, 5.1 Hz, 1H), 3.05 (d, *J* = 8.6 Hz, 1H), 2.63 (s, 1H), 2.50 (s, 1H), 1.96 − 1.73 (m, 2H), 1.54 (bs, 1H).

Tert-Butyl 4-(3-(pyridin-4-yl)propanamido)piperidine-1-carboxylate **(94)**. *t*-Butyl 4-aminopiperidine-1-carboxylate (500 mg, 2.50 mmol), 3-(pyridin-4-yl)propanoic acid (377 mg, 2.50 mmol), TEA (1.04 mL, 7.49 mmol), EDC·HCl (574 mg, 3.00 mmol), and HOBT (459 mg, 3.00 mmol) were all dissolved in DCM (30 mL) and stirred at RT for 30 h, after which time the DCM was removed *in vacuo* and the residue was taken up in EtOAc and washed with H$_2$O (1X), 10% Na$_2$CO$_3$ (3X), and brine (1X), then dried (MgSO$_4$) and concentrated. The resulting residue was purified via silica flash chromatography (40 g silica, 1% to 10% methanolic ammonia:EtOAc) to provide a white solid. Yield: 591 mg, 1.773 mmol, 71%.

Ethyl 2-(1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)piperidin-4-yl)acetate **(96).**

Indole carboxylic acid **4a** (520 mg, 1.82 mmol), ethyl 2-(piperidin-4-yl)acetate **95** (345 mg, 1.65 mmol), TEA (1.15 mL, 8.27 mmol), EDC·HCl (380 mg, 1.98 mmol), and HOBT (304 mg, 1.98 mmol) were all dissolved in DCM (50 mL) and stirred at RT for 24 h, after which time the DCM was removed *in vacuo* and the residue was taken up in EtOAc and washed with 10% aq. citric acid (3X), H_2O (2X), 10% Na_2CO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated. The resulting residue was purified via silica flash chromatography (40 g silica, 10% to 50% EtOAc:Hex) to provide a white powder. Yield: 460 mg, 1.05 mmol, 63%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.67 − 7.62 (m, 1H), 7.36 (d, $J = 8.3$ Hz, 1H), 7.29 − 7.25 (m, 1H), 7.24 − 7.20 (m, 2H), 7.18 − 7.13 (m, 1H), 7.06 − 7.01 (m, 2H), 6.62 (s, 1H), 5.48 (s, 2H), 4.63 (bs, 1H), 4.13 (q, $J = 7.2$ Hz, 3H), 2.88 (bs, 1H), 2.74 (bs, 1H), 2.18 (d, $J = 7.0$ Hz, 2H), 1.98 (ttt, $J = 11.0, 7.1, 3.7$ Hz, 1H), 1.75 (bs, 1H), 1.57 (bs, 1H), 1.26 (t, $J = 7.1$ Hz, 3H), 1.11 (bs, 1H), 0.65 (bs, 1H).

4-(3-(Triphenylphosphoranyl)propyl)pyridine bromide **(98)**. 4-(3-hydroxypropyl)pyridine **97** (1.42 g, 10.35 mmol) and PPh_3 (4.07 g, 15.53 mmol) were dissolved in conc. HBr (8 mL) in a pressure vessel. The reaction was sealed and the suspension as stirred at 135 °C for 2 h, over which time a homogenous solution was achieved. After this time, the reaction was allowed to cool to RT, eliciting precipitation. The reaction was taken up in toluene and transferred to a flask equipped with a Dean-Stark apparatus and water condenser, and H_2O was removed with heating of 150 °C for 3 h, at which time more conc. HBr (8 mL) was added. H_2O was again removed at 150 °C

for 6 h. Once sufficient water had been removed, a precipitate began to appear and the reaction was allowed to cool to RT, at which point it was diluted with H_2O (50 mL) and washed with Et_2O (5X). The pH was raised to pH 7 by the addition of 10% aq. NaOH, and the resulting solution was washed with Et_2O to remove PPh_3 which caused significant precipitation of product. The aqueous slurry was then washed with Et_2O (3X) and cooled in a fridge for 2 h, after which time the precipitate was collected over a filter and washed with cold H_2O and Et_2O. The highly hygroscopic crystalline material was dried under high vacuum for 1 d and stored in a desiccator. No further purification necessary. Off-white crystalline solid. Yield: 1.601 g, 3.46 mmol, 34%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.41 (d, J = 5.9 Hz, 2H), 7.82 − 7.74 (m, 9H), 7.70 − 7.64 (m, 6H), 7.17 (d, J = 5.9 Hz, 2H), 3.98 − 3.90 (m, 2H), 3.11 (t, J = 7.4 Hz, 2H), 1.93 (dq, J = 15.7, 7.8 Hz, 2H). TOF ES+ MS: (M+) 382.2.

(Z)-tert-Butyl 4-(4-(pyridin-4-yl)but-1-en-1-yl)piperidine-1-carboxylate (**99a**). Due to the poor ethereal solubility of the phosphonium bromide, the reaction was conducted at RT. 4-(3-(bromotriphenylphosphoranyl)propyl)pyridine **98** (330 mg, 0.714 mmol) was added to anhydrous THF in a flame-dried flask under N_2 at RT. To the suspension, *n*-BuLi (2.5 M in hexane) was added dropwise at RT until a light yellow color persisted, at which point a full equivalent was added (285 µL, 0.714 mmol) to give a red solution. The reaction then stirred for 10 min at RT under N_2 to allow for ylide dissolution. Predissolved in anhydrous THF (10 mL), *t*-butyl 4-formylpiperidine-1-carboxylate **8** (138 mg, 0.649 mmol) was added dropwise at RT to the reaction, and the reaction was allowed to stir at RT under N_2 for 14 h, at which time the reaction was

quenched with sat. NH$_4$Cl and extracted with a 1:1 solution of EtOAc:Et$_2$O . This extract was then washed with 10% aq. Na$_2$CO$_3$ (1X) and brine (1X), dried (MgSO$_4$) and concentrated. The residue was purified via silica flash chromatography (12 g silica, isocratic 60% EtOAc:Hex) to give a clear, colorless oil, >50:1 Z:E. Yield: 117 mg, 0.370 mmol, 57%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.47 (d, *J* = 4.9 Hz, 2H), 7.08 (d, *J* = 5.1 Hz, 2H), 5.38 (t, *J* = 6.1 Hz, 1H), 5.36 (d, *J* = 5.9 Hz, 1H), 4.04 (s, 2H), 2.67 (q, *J* = 10.3, 7.7 Hz, 4H), 2.31 (q, *J* = 7.1 Hz, 2H), 2.08 − 1.97 (m, 1H), 1.64 − 1.55 (m, 2H), 1.45 (s, 9H), 1.19 (tt, *J* = 13.2, 5.9 Hz, 2H). TOF ES+ MS: (M + H, no Boc) 217.2.

(E)-tert-Butyl 4-(4-(pyridin-4-yl)but-1-en-1-yl)piperidine-1-carboxylate **(99b)**. LiBr (340 mg, 3.92 mmol) was dried by flame-heating a 2-neck flask under high vacuum until the molten salt ceased bubbling. The flask was then purged with N$_2$ and a stir bar was added (so the bar would not char during the flame-dry) and the flask was allowed to cool to RT. The flask was then charged with anhydrous THF (40 mL) and 4-(3-(triphenylphosphoranyl)propyl)pyridine bromide **98** (823 mg, 1.78 mmol) under N$_2$. Without cooling – due to the poor ethereal solubility of the phosphonium bromide – 2 M PhLi in (Bu)$_2$O was added at RT until a slight yellow color persisted, at which point a full 1.1 equivalent of PhLi (0.98 mL, 1.96 mmol) was added, and the suspension gave way to mostly homogenous red solution after 20 min of RT stirring. This solution was then chilled to -78 °C, and *t*-butyl 4-formylpiperidine-1-carboxylate **8** (380 mg, 1.78 mmol) – pre-dissolved in anhydrous THF (10 mL) – was added dropwise to the solution, causing a loss of color. The light yellow solution was allowed to stir at -78 °C for 30 min, at which time the second equivalent of PhLi (0.98 mL, 1.96 mmol) was added dropwise, resulting

in the solution becoming a very dark cherry-red color. The cooling bath was then removed, and the reaction was allowed to stir for 30 min as it approached RT. The reaction was then re-chilled to -78 °C and *t*-BuOH (0.204 mL, 2.14 mmol) − predissolved in anhydrous THF (5 mL) − was added dropwise, eliciting precipitation and decolorization. The reaction was allowed to warm to RT and stir for 36 h, after which time the reaction was quenched with a small amount of sat. aq. NH_4Cl and diluted with a 2:1 $EtOAc:Et_2O$ solution and washed with 10% aq. Na_2CO_3 (1X) and brine (1X), dried ($MgSO_4$), and concentrated *in vacuo*. Purification accomplished by silica flash chromatography (40 g silica, 50% to 80% EtOAc:Hex) to provide the desired olefin as a clear colorless oil, ~5:1 E:Z. Yield: 231 mg, 0.730 mmol, 41%. ^1H NMR (400 MHz, Chloroform-d) δ 8.43 (d, J = 4.4 Hz, 2H), 7.06 (d, J = 4.5 Hz, 2H), 5.25 (t, J = 8.9 Hz, 1H), 5.16 (t, J = 10.0 Hz, 1H), 3.98 (s, 2H), 2.67 − 2.55 (m, 4H), 2.34 (q, J = 7.3 Hz, 2H), 2.27 − 2.12 (m, 1H), 1.40 (s, 9H), 1.30 (s, 2H), 1.13 (d, J = 11.8 Hz, 2H). TOF ES+ MS: (M + H, no Boc) 217.2, (M + Na, no Boc) 239.2.

Tert-Butyl 4-((2-(pyridin-4-yl)ethyl)carbamoyl)piperazine-1-carboxylate **(101)**. *t*-Butyl piperazine-1-carboxylate (400 mg, 2.15 mmol) was dissolved in DCM (15 mL) at RT under N_2 and cooled to 0 °C. 15% wt. phosgene (360 µL, 0.50 mmol) in toluene was added to the solution dropwise, followed by the dropwise addition of TEA (0.90 mL, 6.44 mmol), and then the dropwise addition of 4-(2-aminoethyl)pyridine (565 µL, 4.72 mmol). The reaction was allowed to stir for 1 h at 0 °C, at which point it was diluted with EtOAc and washed with 10% aq. NaOH (1X), H_2O (3X), and brine (1X), dried ($MgSO_4$), and concentrated. The residue was then purified via silica flash chromatography (40 g silica,

1% to 20% methanolic ammonia:EtOAc) to give a white solid after concentration. Yield: 582 mg, 1.74 mmol, 81%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.44 (d, J = 5.6 Hz, 2H), 7.09 (d, J = 5.4 Hz, 2H), 4.92 (t, J = 5.8 Hz, 1H), 3.47 (q, J = 6.7 Hz, 2H), 3.39 – 3.34 (m, 5H), 3.30 – 3.27 (m, 3H), 2.81 (t, J = 7.0 Hz, 2H), 1.43 (d, J = 1.6 Hz, 12H).

1-Tert-Butyl 4-ethyl 4-(phenylselanyl)piperidine-1,4-dicarboxylate (**103**). DIPA (2.49 mL, 17.49 mmol) was dissolved into anhydrous THF (10 mL) in dry glassware under N_2, then chilled to -78 °C. Then, 2.5 M *n*-BuLi (5.60 mL, 13.99 mmol) was added dropwise, and the reaction was allowed to stir for 10 min. 1-*Tert*-butyl 4-ethyl piperidine-1,4-dicarboxylate **102** (3.00 g, 11.66 mmol) – pre-dissolved in THF (10 mL) – was added dropwise to the reaction and the reaction was allowed to stir at -78 °C for 15 min. PhSeCl (2.68 g, 13.99 mmol) – pre-dissolved in anhydrous THF (10 mL) – was added dropwise to the reaction under the same conditions. The cooling bath was removed to allow for warming to RT, and the reaction was stirred for 3 h. After this time, the reaction was quenched by the addition of sat. aq. NH_4Cl and extracted with 1:1 EtOAc:Et_2O. The extract was subsequently washed with H_2O (2X) and brine (1X), dried ($MgSO_4$) and concentrated. The residue was purified by silica flash chromatography (80 g, 5% EtOAc:Hex) to give a clear, slightly yellow oil. Yield: 2.80 g, 6.78 mmol, 58%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.56 (d, J = 7.5 Hz, 2H), 7.39 (t, J = 7.4 Hz, 1H), 7.30 (t, J = 7.7 Hz, 2H), 4.10 (q, J = 7.1 Hz, 2H), 3.74 (s, 2H), 3.11 – 3.03 (m, 2H), 2.13 – 2.06 (m, 2H), 1.84 (s, 2H), 1.44 (s, 9H), 1.18 (t, J = 7.1 Hz, 3H).

Ethyl 4-(phenylselanyl)piperidine-4-carboxylate hydrochloride **(104)**. 1-*tert*-butyl 4-ethyl 4-(phenylselanyl)piperidine-1,4-dicarboxylate (1.70 g, 4.12 mmol) was dissolved in Et$_2$O (100 mL) at RT and 4 M HCl in 1,4-dioxane (15.46 mL, 61.80 mmol) was added dropwise. The reaction was stirred at RT for 4 d until complete by TLC. The solvent was removed *in vacuo* and Et$_2$O was added and stripped off five times (5X) to remove excess HCl and solidify the salt. Further removal of solvent and HCl via rotovap and, lastly, high vacuum furnished the hydrochloride salt. Yield, 1.44, g, 4.12 mmol, 100%. TOF ES+ MS: (M + H) 314.1, (M + Na) 336.0.

Ethyl 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)-4-(phenylselanyl)piperidine-4-carboxylate **(105)**. Ethyl 4-(phenylselanyl)piperidine-4-carboxylate hydrochloride (200 mg, 0.574 mmol) was dissolved in DCM (10 mL) and TEA (0.48 mL, 3.44 mmol), followed by 1-(4-chlorobenzyl)-1H-indole-2-carboxylic acid (164 mg, 0.574 mmol), EDC·HCl (121 mg, 0.631 mmol), and HOBT (97 mg, 0.631 mmol), and the reaction was allowed to stir at RT for 15 h. After this time, the reaction was diluted with a 1:1 solution of EtOAc:Et$_2$O and washed with 1M HCl (3X), H$_2$O (3X), 10% Na$_2$CO$_3$ (3X), and brine (1X), then dried (MgSO$_4$) and concentrated *in vacuo*. The resulting residue was purified via silica flash chromatography (12 g silica, 5% to 20% EtOAc:Hex) to yield an oil that may triturated to provide a fine white powder. Yield: 208 mg, 0.359 mmol, 63%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.64 (d, *J* = 7.9 Hz, 1H), 7.54 (d, *J* = 7.9 Hz, 2H), 7.43 – 7.36 (m, 2H), 7.30 (dt, *J* = 15.2, 7.8 Hz, 3H), 7.23 (d, *J* = 8.3 Hz, 2H), 7.16 (t, *J* = 7.5 Hz, 1H), 7.04 (d, *J* = 8.2 Hz, 2H), 6.61 (s, 1H), 5.48 (s, 2H), 4.12 (q, *J* = 7.1 Hz, 3H),

3.84 (s, 1H), 3.33 – 3.20 (m, 2H), 2.12 (s, 1H), 1.81 (s, 2H), 1.45 (s, 1H), 1.19 (t, *J* = 7.1 Hz, 3H).

Ethyl 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)-1,2,3,6-tetrahydropyridine-4-carboxylate **(106)**. Ethyl 1-(1-(4-chlorobenzyl)-1*H*-indole-2-carbonyl)-4-(phenylselanyl)piperidine-4-carboxylate (160 mg, 0.276 mmol) was dissolved in DCM (10 mL) at RT then cooled to -78 °C. Suspended in DCM (10 mL), ~60% *m*CPBA (80 mg, 0.276 mmol) was added to the solution dropwise and the reaction was allowed to stir for 30 min. Still at -78 °C, TEA (115 µL, 0.828 mmol) was added dropwise and the reaction was allowed to stir for another 30 min. At this time, the reaction was warmed to RT, then the reaction was then diluted with 2:1 EtOAc:Et$_2$O and washed with 10% aq. Na$_2$CO$_3$ (3X), H$_2$O (1X), 1 M HCl (2X), H$_2$O (1X) again, and sat. aq. Na$_2$CO$_3$ (1X). The organic phase was dried with MgSO$_4$ and concentrated. The resulting solid residue was taken up in Hex and the yellow supernatant was decanted off to remove selenoxides. Further purification was accomplished by silica flash chromatography (12 g silica, 5% to 20% EtOAc:Hex) to provide the desired unsaturated ester as a white solid. Yield: 96 mg, 0.227 mmol, 82%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.66 (d, *J* = 7.9 Hz, 1H), 7.37 (d, *J* = 8.3 Hz, 1H), 7.29 (t, *J* = 7.7 Hz, 1H), 7.22 – 7.11 (m, 3H), 7.00 (d, *J* = 7.5 Hz, 2H), 6.68 (s, 1H), 5.48 (s, 2H), 4.23 (q, *J* = 7.1 Hz, 4H), 3.68 (s, 2H), 2.32 (s, 2H), 1.31 (t, *J* = 7.1 Hz, 3H).

1-Tert-Butyl 4-ethyl 4-methylpiperidine-1,4-dicarboxylate **(107).** DIPA (0.62 mL, 4.37 mmol) was dissolved into anhydrous THF (10 mL) in dry glassware under N$_2$, then

chilled to -78 °C. Then, 2.5 M *n*-BuLi (1.40 mL, 3.50 mmol) was added dropwise, and the reaction was allowed to stir for 15 min. Boc-ethyl isonipecotate **102** (750 mg, 2.91 mmol) – pre-dissolved in THF (5 mL) – was added dropwise to the reaction and the reaction was allowed to stir at -78 °C for 15 min. MeI (220 μL, 3.50 mmol) – pre-dissolved in anhydrous THF (3 mL) – was added dropwise to the reaction under the same conditions. The cooling bath was removed to allow for warming to RT, and the reaction was stirred for 8 h. After this time, the reaction was quenched by the addition of sat. aq. NH$_4$Cl and diluted with 1:1 EtOAc:Et$_2$O, then washed with H$_2$O (3X) and brine (1X), dried (MgSO$_4$) and concentrated. The residue was purified by silica flash chromatography (40 g, 5 to 20% EtOAc:Hex) to give a clear colorless oil. ^{1}H NMR (500 MHz, Chloroform-d) δ 4.14 (q, *J* = 7.1 Hz, 2H), 3.74 (s, 2H), 2.96 (s, 2H), 2.04 (d, *J* = 13.3 Hz, 2H), 1.42 (s, 9H), 1.32 (t, *J* = 12.5 Hz, 2H), 1.24 (t, *J* = 7.5 Hz, 3H), 1.17 (s, 3H).

Ethyl 4-methylpiperidine-1,4-dicarboxylate hydrochloride **(108).** The Boc-protected amino ester **107** (500 mg, 1.84 mmol) was dissolved into 4M HCl dioxane (4 mL) and stirred for 15 min. After this time, the dioxane was removed *in vacuo* and the material was sonicated in Et$_2$O to give a tan solid. The Et$_2$O was decanted off and the precipitate was dried via aspirator, then high vacuum, to afford the desired amine hydrochloride as a tan, hygroscopic solid. No further purification necessary. Yield: 350 mg, 1.68 mmol, 100%. TOF ES+ MS: (M + H) 172.1, (M + Na) 194.1.

Ethyl   1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)-4-methylpiperidine-4-carboxylate **(109).** The amine hydrochloride **108** (240 mg, 1.16 mmol), indole carboxylic acid **4a** (300 mg, 1.05 mmol), TEA (585 µL, 4.20 mmol), EDC·HCl (242 mg, 1.26 mmol), and HOBT (193 mg, 1.26 mmol) were dissolved in DCM (30 mL) and stirred at RT for 22 h. After this time, the reaction as diluted with EtOAc and washed with 1M HCl (3X), H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic extract was dried ($MgSO_4$) and concentrated, and the residue purified via silica flash chromatography (40 g silica, 30% to 90% EtOAc:Hex) to give a white solid. Yield: 281 mg, 0.64 mmol, 61%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.65 (d, *J* = 7.9 Hz, 1H), 7.38 (d, *J* = 8.3 Hz, 1H), 7.31 − 7.26 (m, 1H), 7.21 (d, *J* = 7.3 Hz, 2H), 7.16 (t, *J* = 7.5 Hz, 1H), 7.03 (d, *J* = 7.8 Hz, 2H), 6.61 (s, 1H), 5.48 (s, 2H), 4.29 (bs, 1H), 4.18 (q, *J* = 7.4, 7.0 Hz, 2H), 3.86 (bs, 1H), 3.10 (bs, 1H), 2.98 (bs, 1H), 2.12 (bs, 1H), 1.85 (bs, 1H), 1.31 − 1.20 (m, 4H), 1.14 (s, 3H), 0.77 (s, 2H).

(1R,5S,6r)-Ethyl   3-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)-3-azabicyclo[3.1.0]hexane -6-carboxylate **(113)**. The amine *exo*-**112** (170 mg, 0.89 mmol) – prepared in a manner similar to that reported[150, 151] – indole carboxylic acid **4a** (304 mg, 1.06 mmol), TEA (495 µL, 3.55 mmol), EDC·HCl (204 mg, 1.06 mmol), and HOBT (163 mg, 1.06 mmol) were dissolved in DCM (10 mL) and stirred at RT for 18 h. After this time, the reaction as diluted with EtOAc:Et$_2$O (1:1) and washed with 1M HCl (1X), H_2O (2X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic extract was dried ($MgSO_4$) and concentrated, and the residue purified via silica flash chromatography (25 g silica, 5% to 20% EtOAc:Hex) to give a white solid. Yield: 244 mg, 0.58 mmol, 78%. ^{1}H NMR (500

MHz, Chloroform-d) δ 7.66 (d, J = 7.9 Hz, 1H), 7.37 (d, J = 8.4 Hz, 1H), 7.32 − 7.28 (m, 1H), 7.26 − 7.22 (m, 2H), 7.17 (dd, J = 8.2, 6.7 Hz, 1H), 6.99 (d, J = 8.1 Hz, 2H), 6.74 (s, 1H), 5.56 (d, J = 49.7 Hz, 2H), 4.22 (bd, J = 12.4 Hz, 1H), 4.15 (q, J = 7.2 Hz, 2H), 3.77 (bd, J = 13.0 Hz, 2H), 3.52 (s, 1H), 2.18 − 2.04 (m, 2H), 1.29 (td, J = 7.1, 1.2 Hz, 3H), 1.06 (t, J = 3.1 Hz, 1H).

(1R,3s,5S)-Tert-Butyl 3-((2-(pyridin-4-yl)ethyl)carbamoyl)-8-azabicyclo[3.2.1]octane-8-carboxylate (**115**). The Boc-protected amino ester **114** (175 mg, 0.65 mmol) was dissolved in anhydrous THF (10 mL) with KOTMS (83 mg, 0.65 mmol) and stirred under dry conditions under N_2 for 18 h, at which time a precipitate was observed and the solvent was removed *in vacuo* to give the potassium salt as a white powder. No further purification necessary. Yield: 190 mg, 0.65 mmol, 100%.

The potassium salt (191 mg, 0.65 mmol), amine **27** (93 μL, 0.78 mmol), EDC·HCl (150 mg, 0.78 mmol), HOBT (119 mg, 0.78 mmol), and TEA (362 μL, 2.60 mmol) were dissolved in DCM (10 mL) and stirred at RT for 24 h, at which time the DCM was removed *in vacuo* and the residue was taken up in EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic extract was dried (MgSO$_4$) and concentrated, and the residue purified via silica flash chromatography (25 g silica, 1% to 20% methanolic ammonia:EtOAc) to give a tan powder. Yield: 41 mg, 0.083 mmol, 57%. Yield: 146 mg, 0.41 mmol, 63%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.49 (d, J = 5.8 Hz, 2H), 7.10 (d, J = 5.7 Hz, 2H), 5.66 (t, J = 5.9 Hz, 1H), 4.23 (d, J = 40.3 Hz, 2H), 3.54 − 3.48 (m, 2H), 2.81 (t, J = 7.0 Hz, 2H), 2.58 (tt, J = 11.8, 5.3 Hz, 1H), 1.97 − 1.84 (m, 5H), 1.61 − 1.56 (m, 3H), 1.46 (d, J = 1.1 Hz, 9H).

N-(2-(Pyridin-4-yl)ethyl)-2-tosyl-2-azaspiro[3.3]heptane-6-carboxamide **(123).**

The tosyl amide carboxylic acid **122** (2.0 g, 6.77 mmol) – prepared in a manner similar to that reported[152, 153] – amine **27** (0.97 mL, 8.13 mmol), HATU (309 g, 8.13 mmol), and TEA (2.83 mL, 20.31 mmol) were dissolved in DMF (10 mL) and stirred at RT for 18 h, at which time the reaction was diluted with EtOAc and washed with H_2O (5X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic extract was dried ($MgSO_4$) and concentrated, and the residue purified via silica flash chromatography (80 g silica, 1% to 20% methanolic ammonia:EtOAc) to give an extremely thick, clear, yellow-tinged oil. Yield: 1.95 g, 4.88 mmol, 72%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.31 (d, J = 5.4 Hz, 2H), 7.59 (d, J = 8.0 Hz, 2H), 7.29 (d, J = 8.0 Hz, 2H), 6.99 (d, J = 5.4 Hz, 2H), 6.30 (t, J = 5.8 Hz, 1H), 3.61 (d, J = 5.5 Hz, 4H), 3.37 (q, J = 6.8 Hz, 2H), 2.69 (t, J = 7.1 Hz, 2H), 2.62 (p, J = 7.8 Hz, 1H), 2.37 (s, 3H), 2.12 (dd, J = 11.1, 8.7 Hz, 2H), 2.05 – 1.98 (m, 2H). TOF ES+ MS: (M + H) 400.1, (M + Na) 422.1.

Methyl 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)azetidine-3-carboxylate **(125).** The following reagents were dissolved in DCM (10 mL) at RT: indole carboxylic acid **4a** (150 mg, 0.53 mmol), methyl 4-azetidinecarboxylate hydrochloride (88 mg, 0.58 mmol), TEA (366 µL, 2.26 mmol), EDC·HCl (121 mg, 0.63 mmol), and HOBT (96 mg, 0.63 mmol). The reaction was allowed to stir for 26 h at RT, after which time the solvent was remove *in vacuo* and the residue was dissolved in EtOAc and washed with 1M HCl (3X), H_2O (2X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated. The residue was purified by silica flash chromatography (25 g silica, 5 to

40% EtOAc:Hex). Yield: 117 mg, 0.30 mmol, 58%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.67 (d, J = 8.0 Hz, 1H), 7.34 – 7.27 (m, 2H), 7.22 – 7.14 (m, 3H), 7.04 – 6.99 (m, 2H), 6.83 (d, J = 1.2 Hz, 1H), 5.73 (s, 2H), 4.53 (bd, J = 36.9 Hz, 2H), 4.32 (bd, J = 26.0 Hz, 2H), 3.78 (d, J = 1.4 Hz, 3H), 3.47 (p, J = 7.1 Hz, 1H).

Ethyl 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)azepane-4-carboxylate **(132).** The carboxylic acid **4a** (757 mg, 2.65 mmol), azapane ester hydrochloride **131** (550 mg, 2.65 mmol) – prepared in a manner similar to that reported[160] – EDC·HCl (558 mg, 2.91 mmol), HOBT (446 mg, 2.91 mmol), and TEA (1.48 mL, 10.59 mmol) were dissolved in DCM (20 mL) and stirred at RT for 18 h, at which time the DCM was removed *in vacuo* and the residue was taken up in EtOAc and washed with 1M HCl (2X), H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic extract was dried ($MgSO_4$) and concentrated, and the residue purified via silica flash chromatography (45 g silica, 1% to 20% methanolic ammonia:EtOAc) to give a colorless oil. Yield: 756 mg, 1.72 mmol, 65%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.09 (d, J = 8.4 Hz, 1H), 7.88 (d, J = 8.2 Hz, 1H), 7.74 (d, J = 8.1 Hz, 0.5H), 7.66 (d, J = 8.0 Hz, 0.5H), 7.59 (ddd, J = 8.2, 7.0, 1.1 Hz, 1H), 7.47 (ddd, J = 8.2, 7.0, 1.1 Hz, 1H), 7.36 (t, J = 7.6 Hz, 1H), 7.30 – 7.25 (m, 0.5H), 7.23 – 7.16 (m, 1.5H), 7.04 (d, J = 8.0 Hz, 1H), 6.99 (d, J = 8.1 Hz, 0.5H), 6.68 (d, J = 11.9 Hz, 0.5H), 6.37 (d, J = 1.2 Hz, 2H), 5.59 – 5.42 (m, 1H), 4.16 (ddt, J = 19.2, 14.0, 7.2 Hz, 2H), 3.98 – 3.60 (m, 2.5H), 3.51 (d, J = 12.2 Hz, 1H), 3.42 – 3.25 (m, 1H), 2.79 (s, 0.5H), 2.53 (d, J = 47.4 Hz, 0.5H), 2.41 (s, 0.5H), 2.20 – 2.04 (m, 1H), 1.94 (bs, 1.5H), 1.77 (bd, J = 12.1 Hz, 1H), 1.63 (m, 0.5H), 1.51 (bs, 0.5H), 1.38 (m, J = 10.0 Hz, 0.5H), 1.33 – 1.01 (m, 3H).

Methyl 1-(4-chlorobenzyl)-4-fluoro-1H-pyrrole-2-carboxylate **(138).** Methyl 4-fluoro-1*H*-pyrrole-2-carboxylate **137** (130 mg, 0.91 mmol) was dissolved in anhydrous DMF at room temperature, followed by the addition of potassium carbonate (151 mg, 1.09 mmol) and **2a** (0.14 mL, 1.09 mmol). The reaction was then stirred for 30 h at 60 °C, after which time it was allowed to cool to room temperature. The reaction was diluted with a 1:1 solution of ethyl acetate/diethyl ether, washed with water (3×) and brine (1×), dried with magnesium sulfate, and concentrated in vacuo. The resulting brown residue was purified via flash chromatography (60 g silica, 10% ethyl acetate/hexanes) to afford methyl 1-(4-chlorobenzyl)-4-fluoro-1*H*-pyrrole-2-carboxylate as a light-yellow oil. Yield: 203 mg (83%). ^{1}H NMR (400 MHz, CDCl3) δ 7.27 (d, J = 8.3 Hz, 2H), 7.03 (d, J = 8.3 Hz, 2H), 6.69−6.59 (m, 2H), 5.44 (s, 2H), 3.75 (s, 3H).

1-(4-Chlorobenzyl)-4-fluoro-1H-pyrrole-2-carboxylic acid **(139).** Methyl 1-(4-chlorobenzyl)-4-fluoro-1*H*-pyrrole-2-carboxylate **138** (150 mg, 0.56 mmol) was dissolved in ethanol (10 mL) at room temperature. Then 10% aqueous sodium hydroxide (2 mL) was added, and the reaction was stirred for 18 h at room temperature. At this time, solvent was stripped off in vacuo until material began to precipitate. Additional water was added (2 mL), and the solution was cooled in an ice bath, then acidified with concentrated HCl. The resulting precipitate was collected over a filter and washed with cold 1 M HCl, then dried under high vacuum to afford the desired carboxylic acid as a white powder. Yield: 111 mg (78%). ^{1}H NMR (400 MHz, CDCl3) δ 7.29 (d, J = 8.3 Hz, 2H), 7.04 (d, J = 8.3 Hz, 2H), 6.79 (d, J = 2.0 Hz, 1H), 6.71−6.65 (m, 1H), 5.43 (s, 2H).

Ethyl 1-(4-chlorobenzyl)-1H-pyrrolo[2,3-c]pyridine-2-carboxylate **(144).** Ethyl 1-(4-chlorobenzyl)-1*H*-pyrrolo[2,3-*c*]pyridine-2-carboxylate **143** (300 mg, 0.953 mmol) was dissolved into ethanol (8 mL) at RT, followed by the addition of 10% aqueous NaOH (8 mL), which initially caused precipitation, but homogeneity was achieved over time. The reaction was allowed to stir at RT for 12 h, at which time the solvent was removed in vacuo. The residue was taken up in a small amount of water (5 mL), the pH was adjusted to ~4, and material was extracted with ethyl acetate (8×). The extracts were combined and the solvent removed again in vacuo to provide the title compound as a fine white powder. Yield: 226 mg, 83%. ^{1}H NMR (400 MHz, DMSO-d6) δ 9.64 (s, 1H), 8.40 (d, J = 6.3 Hz, 1H), 8.26 (d, J = 6.3 Hz, 1H), 7.61 (s, 1H), 7.35 (d, J = 8.5 Hz, 2H), 7.10 (d, J = 8.5 Hz, 2H), 6.06 (s, 2H).

(4-(Cyclohexylmethyl)piperazin-1-yl)(1-(4-fluorobenzyl)-1H-indol-2-yl)methanone **(CCG-203941).** The following reagents were dissolved in anhydrous DMF (1.5 mL) at RT: 1-(4-fluorobenzyl)-1H-indole-2-carboxylic acid **4b** (50 mg, 0.186 mmol), 1-(cyclohexylmethyl)piperazine (34 mg, 0.186 mmol), DIPEA (97 µL, 0.557 mmol), EDC·HCl (46 mg, 0.241 mmol), and HOBT (37 mg, 0.241 mmol). The reaction was allowed to stir for 24 h at RT, after which time it was diluted with 1:1 EtOAc:Et$_2$O and washed with 10% aq. Na$_2$CO$_3$ (2X), then dried (MgSO$_4$) and concentrated. The residue was purified by silica flash chromatography (20 g silica, 60% EtOAc:Hex). Yield: 45 mg, 0.10 mmol, 53%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.67 (d, *J* = 7.9 Hz, 1H), 7.41 (d, *J* = 8.1 Hz, 1H), 7.32 – 7.29 (m, 1H), 7.20 – 7.16 (m, 1H), 7.11 (dd, *J* =

8.6, 5.4 Hz, 2H), 6.95 (t, *J* = 8.7 Hz, 2H), 6.64 (s, 1H), 5.51 (s, 2H), 3.65 (bd, *J* = 74.0 Hz, 4H), 2.36 (s, 2H), 2.11 − 1.95 (m, 4H), 1.79 − 1.67 (m, 5H), 1.31 − 1.16 (m, 4H), 0.89 − 0.80 (m, 2H). HPLC t_R = 6.65 min, >95% purity.

(4-Benzylpiperazin-1-yl)(1-(4-fluorobenzyl)-1H-indol-2-yl)methanone **(CCG-203942).** The following was added sequentially to anhydrous DMF: 1-(4-fluorobenzyl)-1H-indole-2-carboxylic acid (50 mg, 0.186 mmol), DIPEA (65 µL, 0.371 mmol), EDC·HCl (46 mg, 0.241 mmol), HOBT (37 mg, 0.241 mmol), and 1-benzylpiperazine (33 mg, 0.186 mmol). The solution was allowed to stir at room temperature for 22 h. At this time, a 1:1 solution of EtOAc:Et$_2$O (5 mL) was added, and this was washed with aqueous 10% Na$_2$CO$_3$ (3X). The extract was then dried with anhydrous MgSO$_4$, filtered, and the filtrate was concentrated *in vacuo*. The crude residue was purified by column chromatography (20 g silica, 80% EtOAC/Hexanes) to provide the title compound. Yield: 57 mg, 0.13 mmol, 72 %. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.68 (d, *J* = 7.9 Hz, 1H), 7.43 (d, *J* = 8.3 Hz, 1H), 7.39 − 7.31 (m, 6H), 7.20 (t, *J* = 7.5 Hz, 1H), 7.12 (dd, *J* = 7.8, 5.6 Hz, 2H), 6.98 (t, *J* = 8.5 Hz, 2H), 6.66 (s, 1H), 5.53 (s, 2H), 3.68 (bd, *J* = 67.9 Hz, 4H), 3.49 (s, 2H), 2.21 (bd, 4H). HPLC t_R = 5.91 min, > 95%.

(4-Benzoylpiperidin-1-yl)(1-(4-fluorobenzyl)-1H-indol-2-yl)methanone **(CCG-203943).** The following reagents were dissolved in anhydrous DMF (2 mL) at RT: 1-(4-fluorobenzyl)-1H-indole-2-carboxylic acid (50 mg, 0.186 mmol), phenyl(piperidine-4-yl)methanone hydrochloride (42 mg, 0.186 mmol), DIPEA (72 µL, 0.557 mmol), EDC·HCl (46 mg, 0.241 mmol), and HOBT (37 mg, 0.241 mmol). The reaction was

allowed to stir for 24 h at RT, after which time it was diluted with 1:1 EtOAc:Et$_2$O and washed with 10% aq. Na$_2$CO$_3$ (3X), then dried (MgSO$_4$) and concentrated. The residue was purified by silica flash chromatography (20 g silica, 80% EtOAc:Hex). Yield: 68 mg, 0.154 mmol, 83%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.96 (dt, J = 8.4, 1.2 Hz, 2H), 7.72 – 7.65 (m, 1H), 7.64 – 7.58 (m, 1H), 7.54 – 7.49 (m, 2H), 7.41 (d, J = 8.3 Hz, 1H), 7.30 (ddd, J = 8.5, 7.1, 1.1 Hz, 1H), 7.19 (td, J = 7.4, 1.0 Hz, 1H), 7.16 – 7.11 (m, 2H), 7.02 – 6.93 (m, 2H), 6.69 (d, J = 0.9 Hz, 1H), 5.52 (s, 2H), 4.63 (bs, 1H), 4.20 (bs, 1H), 3.51 (tt, J = 10.7, 3.7 Hz, 1H), 3.06 (s, 2H), 2.05 – 1.48 (m, 4H). HPLC t_R = 8.32 min, >95%.

(4-(Benzyloxy)piperidin-1-yl)(1-(4-fluorobenzyl)-1H-indol-2-yl)methanone

(CCG-203944). The following reagents were dissolved in anhydrous DMF (3 mL) at RT: 1-(4-fluorobenzyl)-1H-indole-2-carboxylic acid (50 mg, 0.186 mmol), 4-(benzyloxy)piperidine hydrochloride (42 mg, 0.186 mmol), DIPEA (72 µL, 0.557 mmol), EDC·HCl (46 mg, 0.241 mmol), and HOBT (37 mg, 0.241 mmol). The reaction was allowed to stir for 24 h at RT, after which time it was diluted with 1:1 EtOAc:Et$_2$O and washed with 10% aq. Na$_2$CO$_3$ (3X), then dried (MgSO$_4$) and concentrated. The residue was purified by silica flash chromatography (20 g silica, 80% EtOAc:Hex). Yield: 29 mg, 0.066 mmol, 35%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.70 (d, J = 7.9 Hz, 1H), 7.45 – 7.37 (m, 5H), 7.36 – 7.28 (m, 2H), 7.21 (ddd, J = 7.9, 7.0, 0.9 Hz, 1H), 7.16 – 7.11 (m, 2H), 7.00 – 6.94 (m, 2H), 6.68 (d, J = 0.8 Hz, 1H), 5.53 (s, 2H), 4.58 (s, 2H), 3.92 (bd, J = 66.0 Hz, 2H), 3.66 (tt, J = 7.2, 3.5 Hz, 1H), 3.47 (bd, J = 48.5 Hz, 2H), 1.97 – 1.42 (m, 4H). HPLC t_R = 8.86 min, >95% purity.

(1-(4-Fluorobenzyl)-1H-indol-2-yl)(4-(piperidine-1-carbonyl)piperidin-1-yl)methanone **(CCG-203945).** The following was added sequentially to anhydrous DMF (2 mL): 1-(4-fluorobenzyl)-1*H*-indole-2-carboxylic acid (50 mg, 0.186 mmol), DIPEA (0.065 ml, 0.371 mmol), EDC·HCl (46.3 mg, 0.241 mmol), HOBT (37.0 mg, 0.241 mmol), and piperidin-1-yl(piperidin-4-yl)methanone (36.4 mg, 0.186 mmol). The solution was allowed to stir at RT for 24 h. At this time, a 1:1 solution of EtOAc:Et$_2$O (5 mL) was added, and this was washed with aqueous 10% Na$_2$CO$_3$ (3 x 2 mL). The extract was then dried with anhydrous MgSO$_4$, filtered, and the filtrate was concentrated *in vacuo.* The crude residue was purified by column chromatography (20 g silica, 80% EtOAc/Hex) to provide the title compound. Yield: 35 mg, 0.065 mmol, 35%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.70 − 7.64 (m, 1H), 7.37 (d, *J* = 8.4 Hz, 1H), 7.28 (dt, *J* = 5.9, 1.3 Hz, 1H), 7.17 (td, *J* = 7.5, 7.0, 1.0 Hz, 1H), 7.12 (dd, *J* = 8.5, 5.4 Hz, 2H), 6.99 − 6.93 (m, 2H), 6.67 (s, 1H), 5.49 (s, 2H), 4.61 (bs, 1H), 4.24 (bs, *J* = 8.8, 5.8 Hz, 1H), 3.58 (t, *J* = 5.5 Hz, 2H), 3.49 − 3.37 (m, 2H), 2.93 (d, *J* = 12.7 Hz, 2H), 2.75 (dq, *J* = 14.3, 7.7, 7.1 Hz, 1H), 1.77 − 1.58 (m, 8H), 1.31 − 1.25 (m, 1H), 0.94 − 0.81 (m, 1H). HPLC t_R = 7.55 min, >95% purity.

(4-(Dibenzylamino)piperidin-1-yl)(1-(4-fluorobenzyl)-1H-indol-2-yl)methanone **(CCG-204020).** *Tert*-butyl 4-(dibenzylamino)piperidine-1-carboxylate (330 mg, 0.867 mmol) was dissolved in DCM (5 mL), followed by the addition of trifluoroacetic acid (0.200 mL, 2.60 mmol). The reaction was allowed to stir for 18 h at RT, after which time

the solvent was removed *in vacuo* and the crude residue of *N,N*-dibenzylpiperidin-4-amine bis(trifluoroacetate) was used directly in the subsequent reaction

The following reagents were dissolved in anhydrous DMF (5 mL) at RT: 1-(4-fluorobenzyl)-1H-indole-2-carboxylic acid (233 mg, 0.867 mmol), N,N-dibenzylpiperidin-4-amine bis(trifluoroacetate) (243 mg, 0.867 mmol), DIPEA (757 μL, 4.34 mmol), EDC·HCl (183 mg, 0.954 mmol), and HOBT (146 mg, 0.954 mmol). The reaction was allowed to stir for 36 h at RT, after which time it was diluted with 1:1 EtOAc:Et$_2$O and washed with 10% aq. Na$_2$CO$_3$ (2X), then dried (MgSO$_4$) and concentrated. The residue was triturated with Et$_2$O to yield a light brown solid. Yield: 380 mg, 0.715 mmol, 82%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.69 (dd, J = 8.0, 3.9 Hz, 1H), 7.43 – 7.38 (m, 5H), 7.37 – 7.31 (m, 5H), 7.29 – 7.25 (m, 2H), 7.21 (t, J = 7.3, 3.4 Hz, 1H), 7.11 – 7.07 (m, 2H), 6.94 (t, J = 11.1, 8.7, 2.0 Hz, 2H), 6.65 (s, 1H), 5.53 (s, 2H), 4.78 (bs, 1H), 4.17 (bs, 1H), 3.59 (s, 4H), 2.85 – 2.67 (m, 2H), 2.58 (bs, 1H), 1.92 (bs, 1H), 1.67 (bs, 1H), 1.49 (bs, 1H), 1.04 (bs, J = 30.5 Hz, 1H). HPLC t_R = 6.48 min, >95%.

N-Benzyl-1-(1-(4-chlorobenzyl)-1H-pyrrole-2-carbonyl)piperidine-4-carboxamide (**CCG-204054**). The following was dissolved in anhydrous DMF (3 mL): 1-(1-(4-chlorobenzyl)-1H-pyrrole-2-carbonyl)piperidine-4-carboxylic acid (115 mg, 0.332 mmol), Hunig's Base (0.174 ml, 0.995 mmol), EDC·HCl (76 mg, 0.398 mmol), HOBt (60.9 mg, 0.398 mmol), and benzylamine (0.040 ml, 0.365 mmol). This was stirred at room temperature for 1 day with 3Å molecular sieves. At this time, a 1:1 solution of EtOAc:Et$_2$O (20 mL) was added and the organic layer was washed with 10% aq.

Na$_2$CO$_3$ (2 x 10 mL) and brine (1 x 10 mL). The organic solution was then dried (anhydrous MgSO$_4$) and concentrated *in vacuo*. Trituration in Et$_2$O provided 101 mg of the title compound as a fine, ruddy brown solid. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.39 – 7.34 (m, 2H), 7.33 – 7.26 (m, 5H), 7.06 (dd, J = 8.9, 2.5 Hz, 2H), 6.82 (dd, J = 2.7, 1.7 Hz, 1H), 6.35 (dd, J = 3.7, 1.6 Hz, 1H), 6.15 (dd, J = 3.7, 2.7 Hz, 1H), 5.76 (s, 1H), 5.30 (s, 2H), 4.47 (d, J = 5.6 Hz, 2H), 4.39 (bd, J = 13.3 Hz, 2H), 2.86 (t, J = 12.7 Hz, 2H), 2.33 (tt, J = 11.3, 3.8 Hz, 1H), 1.81 (d, J = 13.0 Hz, 2H), 1.51 (d, J = 12.9 Hz, 2H). HPLC t_R = 7.06 min, > 95%.

(1-(4-Fluorobenzyl)-1H-indol-2-yl)(4-(piperidin-1-ylmethyl)piperidin-1-yl)methanone **(CCG-204055).** *Tert*-butyl 4-(piperidin-1-ylmethyl)piperidine-1-carboxylate (100 mg, 0.469 mmol) was dissolved in DCM (5 mL), and TFA (1 mL) was added. The reaction was allowed to stir for 12 h, after which time the solvent was stripped off *in vacuo*, then thoroughly removed under high vacuum. The crude residue was then taken directly into the next step.

The following was added to anhydrous DMF (2 mL) sequentially: 1-(4-fluorobenzyl)-1H-indole-2-carboxylic acid (66.8 mg, 0.248 mmol), DIPEA (0.217 ml, 1.240 mmol), EDC·HCl (52.3 mg, 0.273 mmol), HOBT (41.8 mg, 0.273 mmol), and 1-(piperidin-4-ylmethyl)piperidine trifluoroacetate (45.2 mg, 0.248 mmol). This was stirred at room temperature for 24 h with 3Å MS. At this time, a 1:1 solution of EtOAc:Et$_2$O was added and the solution was washed with 10% aq. Na$_2$CO$_3$. The organic phase was dried with MgSO$_4$ and concentrated *in vacuo*. The residue was then purified by silica gel

chromatography (20g silica, 80% EtOAc/Hexanes) to give an oil. Yield: 58 mg , 0.134 mmol, 54%. HPLC t_R = 5.36 min, > 95%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.66 (d, J = 7.9 Hz, 1H), 7.40 (d, J = 8.3 Hz, 1H), 7.29 (ddd, J = 8.3, 6.9, 1.2 Hz, 1H), 7.18 (ddd, J = 8.0, 7.0, 0.9 Hz, 1H), 7.14 – 7.09 (m, 2H), 6.99 – 6.92 (m, 2H), 6.63 (d, J = 0.7 Hz, 1H), 5.50 (s, 2H), 4.66 (bs, 1H), 4.10 (bs, 1H), 2.77 (d, J = 64.4 Hz, 2H), 2.41 – 2.27 (m, 4H), 2.11 – 2.02 (m, 2H), 1.83 – 1.53 (m, 5H), 1.48 – 1.39 (m, 2H), 1.05 (bs, 1H), 0.66 (bs, 1H). HPLC t_R = 5.36 min, >95% purity.

(1-(4-Fluorobenzyl)-1H-indol-2-yl)(4-(hydroxy(phenyl)methyl)piperidin-1-yl)methanone **(CCG-204056).** The following was added sequentially to anhydrous DMF (5 mL): 1-(4-fluorobenzyl)-1H-indole-2-carboxylic acid (0.119 g, 0.443 mmol), Hunig'sBase (0.232 ml, 1.329 mmol), EDC·HCl (0.093 g, 0.487 mmol), HOBT (0.075 g, 0.487 mmol), and phenyl(piperidin-4-yl)methanol (0.085 g, 0.443 mmol). The solution was stirred at room temperature for 30 h, at which time a 1:1 solution of EtOAc:Et$_2$O was added and washed with 10% aq. Na$_2$CO$_3$. The organic solution was dried with MgSO$_4$ and concentrated *in vacuo* to give a residue that was purified by silica gel chromatography (45 g silica, 80% EtOAc/Hex). Yield: 67 mg, . ^{1}H NMR (500 MHz, Chloroform-d) δ 7.66 (d, J = 7.9 Hz, 1H), 7.38 (dd, J = 14.5, 7.8 Hz, 3H), 7.32 – 7.26 (m, 4H), 7.17 (t, J = 7.5 Hz, 1H), 7.09 (dd, J = 8.5, 5.3 Hz, 2H), 6.98 – 6.92 (m, 2H), 6.61 (s, 1H), 5.48 (d, J = 6.5 Hz, 2H), 4.67 (d, J = 54.5 Hz, 1H), 4.30 – 4.23 (m, 1H), 4.22 – 3.93 (m, 1H), 2.86 – 2.52 (m, 2H), 2.43 – 1.83 (m, 2H), 1.79 (dtd, J = 11.5, 7.7, 3.7 Hz, 1H), 1.28 – 0.94 (m, 2H). HPLC t_R = 7.85 min, > 95%.

(4-(Benzylamino)piperidin-1-yl)(1-(4-fluorobenzyl)-1H-indol-2-yl)methanone

(CCG-205420). (4-(Dibenzylamino)piperidin-1-yl)(1-(4-fluorobenzyl)-1*H*-indol-2-yl)methanone **CCG-204020** was dissolved in EtOH (10 mL) and THF (10 mL), along with a few drops of conc. HCl. This solution was sparged with N_2 and $Pd(OH)_2$ was added. The reaction was then shaken on a hydrogenator at 45 psi H_2 at RT for 1 h (longer reaction times cause rapid accumulation of complete debenzylation.) The reaction was filtered over celite and the filtrate was diluted with EtOAc and washed with sat. aq. Na_2CO_3 (1X), dried ($MgSO_4$) and concentrated to give the desired compound as an oil. Yield: 77%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.70 – 7.65 (m, 1H), 7.41 – 7.32 (m, 5H), 7.32 – 7.27 (m, 2H), 7.22 – 7.16 (m, 1H), 7.12 (dd, J = 8.3, 5.3 Hz, 2H), 6.96 (ddd, J = 9.7, 7.5, 1.1 Hz, 2H), 6.65 (s, 1H), 5.51 (s, 2H), 4.49 (bs, 1H), 4.07 (bs, 1H), 3.83 (s, 2H), 3.00 (bs, 2H), 2.77 (tt, J = 9.9, 4.0 Hz, 1H), 2.04 – 1.64 (m, 2H), 1.32 (m, 2H). TOF ES+ MS: 442.1 (M + H), 464.1 (M + Na). HPLC t_R = 5.83 min, > 90%.

(4-(Cyclohexyl(hydroxy)methyl)piperidin-1-yl)(1-(4-fluorobenzyl)-1H-indol-2-yl)methanone **(CCG-205421).** *Tert*-butyl 4-(cyclohexyl(hydroxy)methyl)piperidine-1-carboxylate (100 mg, 0.336 mmol) was dissolved in anhydrous THF (5 mL) and TFA (52 μL, 0.672 mmol) and stirred at RT for 15 h. After this time, the solvent was removed *in vacuo* via rotovap, then thoroughly removed under high vacuum. The resulting residue was taken directly into the next step.

4-(Cyclohexyl(hydroxy)methyl)piperidine trifluoroacetate (~105 mg, 0.336 mmol) was dissolved in anhydrous DMF (3 mL), followed by 1-(4-fluorobenzyl)-1*H*-indole-2-carboxylic acid (100 mg, 0.370 mmol), DIPEA (0.352 mL, 2.016 mmol),

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EDC·HCl (77 mg, 0.403 mmol), and HOBT (62 mg, 0.403 mmol). The reaction was allowed to stir at RT for 24 h, after which time the reaction was diluted with 1:1 EtOAc:Et$_2$O and washed with 10% aq. Na$_2$CO$_3$ (3X), dried (MgSO$_4$), and concentrated. The resulting residue was purified by silica flash chromatography (20 g silica, 80% EtOAc:Hex). Yield: 90 mg, 0.202 mmol, 60%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.66 (d, J = 7.9 Hz, 1H), 7.40 (d, J = 8.3 Hz, 1H), 7.31 − 7.27 (m, 1H), 7.18 (ddd, J = 7.9, 7.0, 1.0 Hz, 1H), 7.14 − 7.08 (m, 2H), 6.98 − 6.92 (m, 2H), 6.64 (d, J = 0.8 Hz, 1H), 5.51 (s, 2H), 4.75 (bs, 1H), 4.15 (bs, 1H), 3.05 (d, J = 5.6 Hz, 1H), 2.83 (bs, 1H), 2.68 (bs, 1H), 1.82 − 1.58 (m, 7H), 1.41 − 1.04 (m, 9H). ES+ TOF MS: 449.2 (M + H), 471.2 (M + Na). HPLC t_R = 8.72 min, > 90%.

N-Benzyl-1-(1-(4-chlorobenzyl)-3-methyl-1H-indole-2-carbonyl)piperidine-4-carboxamide (**CCG-205431**). Ethyl 1-(4-chlorobenzyl)-3-methyl-1*H*-indole-2-carboxylate (310 mg, 0.946 mmol) was dissolved in THF (20 mL) and 5 M aq. NaOH (20 mL) and stirred at 55 °C for 18 h. The reaction was then cooled to 0 °C and acidified to pH<1 with conc. HCl and material was extracted from the aqueous phase with 1:1 EtOAc:Et$_2$O. The extract was dried (MgSO$_4$) and concentrated, giving 1-(4-chlorobenzyl)-3-methyl-1*H*-indole-2-carboxylic acid as a yellow solid. Yield: 273 mg, 0.911 mmol, 96%.

The following reagents were dissolved in anhydrous DMF (3 mL) at RT: 1-(4-chlorobenzyl)-3-methyl-1*H*-indole-2-carboxylic acid (90 mg, 0.315 mmol), *N*-benzylpiperidine-4-carboxamide hydrochloride (70 mg, 0.315 mmol), DIPEA (170 μL, 0.945 mmol), and HATU (144 mg, 0.378 mmol). The reaction was allowed to stir for 20

h at RT, after which time it was diluted with 1:1 EtOAc:Et$_2$O and washed with 10% aq. Na$_2$CO$_3$ (3X), then dried (MgSO$_4$) and concentrated. The residue was purified by silica flash chromatography (20 g silica, 50% EtOAc:Hex). Yield: 77 mg, 0.153 mmol, 51%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.59 (d, *J* = 8.0 Hz, 1H), 7.33 (d, *J* = 8.3 Hz, 7H), 7.18 (t, *J* = 7.8 Hz, 3H), 7.03 (t, *J* = 8.2 Hz, 2H), 5.73 (s, 1H), 5.57 (s, 1H), 5.38 (d, *J* = 11.8 Hz, 2H), 4.77 (d, *J* = 10.9 Hz, 1H), 4.44 (d, *J* = 5.9 Hz, 2H), 3.76 (d, *J* = 14.8 Hz, 1H), 3.08 (d, *J* = 17.3 Hz, 1H), 2.80 (s, 3H), 2.29 (d, *J* = 21.5 Hz, 3H), 1.96 (d, *J* = 11.2 Hz, 1H), 1.76 (d, *J* = 17.2 Hz, 1H), 1.52 (s, 1H). ES+ TOF MS: 500.3 (M + H), 522.2 (M + Na). HPLC *t*$_R$ = 7.95 min, > 95%.

(1-(4-Chlorobenzyl)-1H-indol-2-yl)(4-(isoindoline-2-carbonyl)piperidin-1-yl)methanone. **(CCG-205470)** The following was added sequentially to DCM (2 mL): 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carboxylic acid (50 mg, 0.126 mmol), DIPEA (0.07 mL, 0.378 mmol), EDC·HCl (30 mg, 0.151 mmol), HOBT (24 mg, 0.151 mmol), and isoindoline (0.018 mL, 0.151 mmol). The mixture was stirred for 18 h at RT, at which time the solution diluted with a 1:1 solution of EtOAc:Et$_2$O and washed with 1M HCl (1X), 10% aq. Na$_2$CO$_3$ (1X) and brine (1X). The organic phase was dried with magnesium sulfate and concentrated *in vacuo*. The resulting residue was dissolved in EtOAc and precipitated out with Hex, and then collected over a filter and washed with a 10:1 solution of Hex:EtOAc. Yield: 22 mg, 0.044 mmol, 35%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.65 (d, *J* = 7.9 Hz, 1H), 7.40 − 7.21 (m, 8H), 7.15 (t, *J* = 7.4 Hz, 1H), 7.05 (d, *J* = 8.3 Hz, 2H), 6.68 (d, *J* = 1.6 Hz, 1H), 5.47 (s, 2H), 4.88 (s, 2H), 4.81 (s, 2H),

4.42 (bs, 4H), 2.97 (bs, 2H), 2.72 (td, *J* = 14.7, 12.7, 4.7 Hz, 1H), 1.78 (bs, 2H). ES+

TOF MS: 498.2 (M + H), 520.2 (M + Na). HPLC t_R = 5.85 min, > 95%.

1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-N-(2,3-dihydro-1H-inden-2-yl)piperidine-4-carboxamide. **(CCG-205471)** The following was added sequentially to DCM (2 mL): 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carboxylic acid (50 mg, 0.126 mmol), DIPEA (0.07 mL, 0.378 mmol), EDC·HCl (30 mg, 0.151 mmol), HOBT (24 mg, 0.151 mmol), and 2-aminoindane (0.020 mL, 0.151 mmol). The mixture was stirred for 36 h at rt, at which time the solution diluted with a 1:1 solution of EtOAc:Et$_2$O and washed with 1M HCl (1X), 10% aq. Na$_2$CO$_3$ (1X) and brine (1X), which resulted in a precipitate that failed to go into either phase. The precipitate was collected and washed with water and Et$_2$O to afford the title compound as a white solid. Yield: 52 mg, 0.102 mmol, 81%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.64 (d, *J* = 7.9 Hz, 1H), 7.35 − 7.29 (m, 1H), 7.25 − 7.11 (m, 8H), 7.03 (d, *J* = 8.1 Hz, 2H), 6.64 (s, 1H), 5.62 (d, *J* = 7.6 Hz, 1H), 5.46 (s, 2H), 4.83 − 4.69 (m, 1H), 4.32 (bs, 4H), 3.33 (dd, *J* = 16.3, 7.0 Hz, 2H), 2.89 − 2.72 (m, 4H), 2.27 − 2.14 (m, 1H), 1.74 (bs, 2H). ES+ TOF MS: 512.2 (M + H), 534.2 (M + Na). HPLC t_R = 5.96 min, > 95%.

1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-N-(2,3-dihydro-1H-inden-1-yl)piperidine-4-carboxamide **(CCG-205472).** The following was added sequentially to DCM (2 mL): 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carboxylic acid **71** (50 mg, 0.13 mmol), DIPEA (0.07 mL, 0.38 mmol), EDC·HCl (30 mg, 0.15 mmol), HOBT (24 mg, 0.15 mmol), and 1-aminoindane (20 μL, 0.15 mmol). The mixture was

stirred for 18 h at rt, at which time the solution diluted with a 2:1 solution of EtOAc:Et$_2$O and washed with 1M HCl (2X), 10% aq. Na$_2$CO$_3$ (1X) and brine (1X). The organic phase was dried with MgSO$_4$ and concentrated *in vacuo*. The resulting residue was dissolved in EtOAc and precipitated out Hex, and then collected over a filter and washed with Et$_2$O to give the title compound as a white solid. Yield: 33 mg, 0.06 mmol, 51%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.66 (d, *J* = 7.9 Hz, 1H), 7.34 (d, *J* = 8.3 Hz, 1H), 7.29 − 7.20 (m, 7H), 7.17 (t, *J* = 7.4 Hz, 1H), 7.04 (d, *J* = 8.0 Hz, 2H), 6.66 (s, 1H), 5.66 (d, *J* = 8.4 Hz, 1H), 5.47 (s, 2H), 4.49 (bs, 1H), 4.33 (bs, 1H), 3.04 − 2.95 (m, 1H), 2.89 (dt, *J* = 15.9, 8.0 Hz, 3H), 2.62 (dtd, *J* = 12.2, 7.8, 3.8 Hz, 1H), 2.33 (tt, *J* = 11.1, 3.9 Hz, 1H), 1.94 − 1.54 (m, 6H). ES+ TOF MS: 512.2 (M + H), 534.2 (M + Na). HPLC t_R = 5.93 min, > 95%.

(1-(4-Chlorobenzyl)-1H-indol-2-yl)(4-(2-phenylpyrrolidine-1-carbonyl)piperidin-1-yl)methanone. **(CCG-205473)** The following was added sequentially to DCM (2 mL): 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carboxylic acid **71** (50 mg, 0.126 mmol), DIPEA (0.07 mL, 0.378 mmol), EDC·HCl (30 mg, 0.151 mmol), HOBT (24 mg, 0.151 mmol), and 2-phenylpyrrolidine (0.023 mL, 0.151 mmol). The mixture was stirred for 18 h at rt, at which time the solution diluted with a 1:1 solution of EtOAc:Et$_2$O and washed with 1M HCl (1X), 10% aq. sodium carbonate (1X) and brine (1X). The organic phase was dried with magnesium sulfate and concentrated *in vacuo*. The resulting residue was dissolved in EtOAc and precipitated out hexanes, and then collected over a filter and washed with diethyl ether to give the title compound as a white solid. Yield: 20 mg, 0.038 mmol, 30%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.66 − 7.57 (m, 1H), 7.33 − 7.14 (m, 10H), 7.00 (d, *J* = 8.1 Hz, 2H), 6.58 (s, 1H), 5.41 (s, 2H), 4.34

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(s, 4H), 3.73 (dd, J = 19.2, 11.3 Hz, 2H), 3.07 − 2.88 (m, 1H), 2.73 (s, 1H), 2.51 − 2.26 (m, 3H), 2.04 − 1.44 (m, 5H). ES+ TOF MS: 526.2 (M + H), 548.2 (M + Na). HPLC t_R = 6.62 min, > 95%.

(1-(4-Chlorobenzyl)-1H-indol-2-yl)(4-(3-phenylpyrrolidine-1-carbonyl)piperidin-1-yl)methanone. **(CCG-205474)** The following was added sequentially to DCM (2 mL): 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carboxylic acid (50 mg, 0.126 mmol), DIPEA (0.07 mL, 0.378 mmol), EDC·HCl (30 mg, 0.151 mmol), HOBT (24 mg, 0.151 mmol), and 3-phenylpyrrolidine (0.023 mL, 0.151 mmol). The mixture was stirred for 36 h at rt, at which time the solution diluted with a 1:1 solution of EtOAc:Et$_2$O and washed with 1M HCl (1X), 10% aq. sodium carbonate (1X) and brine (1X). The organic phase was dried with magnesium sulfate and concentrated *in vacuo*. The resulting residue was dissolved in EtOAc and precipitated out hexanes, and then collected over a filter and washed with diethyl ether to give the title compound as a slightly yellow solid. Yield: 21 mg, 0.040 mmol, 32%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.66 (dd, J = 7.8, 2.8 Hz, 1H), 7.34 (dq, J = 14.5, 7.4 Hz, 3H), 7.29 − 7.20 (m, 6H), 7.16 (t, J = 7.8 Hz, 1H), 7.06 (dd, J = 7.9, 5.4 Hz, 2H), 6.68 (d, J = 6.3 Hz, 1H), 5.48 (d, J = 5.0 Hz, 2H), 4.57 (s, 1H), 4.33 (s, 1H), 4.03 (dd, J = 11.9, 7.5 Hz, 1H), 3.98 − 3.91 (m, 1H), 3.85 (ddd, J = 11.6, 8.3, 2.4 Hz, 1H), 3.73 (td, J = 9.1, 8.3, 2.9 Hz, 1H), 3.60 (td, J = 9.6, 6.8 Hz, 1H), 3.49 (ddd, J = 21.3, 9.6, 4.0 Hz, 2H), 3.38 (p, J = 7.9 Hz, 1H), 2.92 (s, 2H), 2.62 (dp, J = 21.4, 7.5 Hz, 1H), 2.46 − 2.36 (m, 1H), 2.31 (dq, J = 13.1, 4.1, 2.6 Hz, 1H), 2.16 − 2.00 (m, 2H). ES+ TOF MS: 526.3 (M + H), 548.2 (M + Na). HPLC t_R = 6.37 min, > 95%.

(4-(Cyclohexanecarbonyl)piperidin-1-yl)(1-(4-fluorobenzyl)-1H-indol-2-yl)methanone

(CCG-205475). 4-(Cyclohexyl(hydroxy)methyl)piperidin-1-yl)(1-(4-fluorobenzyl)-1*H*-indol-2-yl)methanone (85 mg, 0.189 mmol) was dissolved in DCM (4 mL) and cooled to -78 °C. Anhydrous DMSO (34 μL, 0.474 mmol) and $(COCl)_2$ (34 μL, 0.379 mmol) were added, and the solution was stirred for 30 min, at which time TEA (130 μL, 0.947 mmol) was added dropwise and the reaction was allowed to stir for an additional 30 min, then the bath was removed and it was allowed to stir until it reached RT. The reaction was diluted with 1:1 EtOAc:Et_2O and washed with H_2O (2X) and brine (1X), dried ($MgSO_4$), and concentrated *in vacuo*. The residue was purified via silica flash chromatography (50 g silica, 10% EtOAc:Hex). Yield: 49 mg, 0.106 mmol, 58%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.64 (d, *J* = 7.8 Hz, 1H), 7.37 (d, *J* = 8.4 Hz, 1H), 7.29 − 7.24 (m, 1H), 7.15 (t, *J* = 7.5 Hz, 1H), 7.08 (dd, *J* = 8.2, 5.5 Hz, 2H), 6.99 − 6.89 (m, 2H), 6.62 (s, 1H), 5.47 (s, 2H), 4.54 (bs, 1H), 4.15 (bs, 1H), 2.94 − 2.81 (m, 2H), 2.68 (tt, *J* = 11.2, 3.6 Hz, 1H), 2.46 (tt, *J* = 11.1, 2.9 Hz, 1H), 1.82 − 1.64 (m, 7H), 1.38 − 1.17 (m, 7H). TOF ES+ MS: (M + Na) 469.2. HPLC t_R = 8.93 min, > 90%.

(1-(4-Chlorobenzyl)-1H-indol-2-yl)(4-(piperidin-1-ylmethyl)piperidin-1-yl)methanone **(CCG-205476).** *Tert*-Butyl 4-(piperidin-1-ylmethyl)piperidine-1-carboxylate (238 mg, 0.843 mmol) was dissolved in DCM (10 mL), and TFA (2 mL) was added. The reaction was allowed to stir for 12 h, after which time the solvent was stripped off *in vacuo*, then thoroughly removed under high vacuum. The crude residue was then taken directly into the next step.

The following was added to DCM (6 mL): 1-(4-chlorobenzyl)-1*H*-indole-2-carboxylic acid (110 mg, 0.380 mmol), DIPEA (0.50 mL, 2.95 mmol), EDC·HCl (100 mg, 0.506 mmol), HOBT (80 mg, 0.506 mmol), and 1-(piperidin-4-ylmethyl)piperidine (150 mg, 0.843 mmol). The solution was stirred at RT for 15 h, at which time the solution was diluted with a 1:1 solution of EtOAc:Et$_2$O and washed with H$_2$O (1X), 10% aq. Na$_2$CO$_3$ (1X), and brine (1X). The organic phase was then dried over MgSO$_4$ and concentrated *in vacuo*. The resulting residue was purified via silica flash chromatography (40 g silica, 10% EtOAc:Hex) and triturated with EtOAc and Hex to afford the title compound as a white powder. Yield: 109 mg, 0.242 mmol, 63%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.63 (d, *J* = 7.9 Hz, 1H), 7.35 (d, *J* = 8.3 Hz, 1H), 7.28 − 7.11 (m, 4H), 7.03 (d, *J* = 8.4 Hz, 2H), 6.59 (s, 1H), 5.46 (s, 2H), 4.59 (bs, 2H), 4.09 (bs, 2H), 2.73 (bs, 2H), 2.34 (bs, 2H), 2.08 (d, *J* = 6.8 Hz, 2H), 1.67 (s, 6H), 1.44 − 1.32 (m, 2H), 0.95 (bs, 1H), 0.56 (bs, 1H). ES+ TOF MS: 450.2 (M + H). HPLC t_R = 6.18 min, > 95%.

(4-Benzylpiperazin-1-yl)(1-(4-chlorobenzyl)-1H-indol-2-yl)methanone **(CCG-206327).** The following was added to DCM (5 mL): 1-(4-chlorobenzyl)-1*H*-indole-2-carboxylic acid (70 mg, 0.245 mmol), DIPEA (0.130 mL, 0.735 mmol), EDC·HCl (57 mg, 0.294 mmol), HOBT (45 mg, 0.294 mmol), and 1-benzylpiperazine (0.085 mL, 0.490 mmol). The solution was stirred at RT for 16 h, at which time the solution was diluted with a 1:1 solution of EtOAc:Et$_2$O and washed with H$_2$O (1X), 10% aq. Na$_2$CO$_3$ (1 x), and brine (1 x). The organic phase was then dried over MgSO$_4$ and concentrated *in vacuo*. The resulting residue was purified via silica flash chromatography (30 g silica, 10% EtOAc:Hex) and triturated with EtOAc and Hex to afford the title compound as a

white powder. Yield: 52 mg, 0.117 mmol, 48%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.62 (d, J = 7.9 Hz, 1H), 7.36 (d, J = 8.3 Hz, 1H), 7.32 − 7.19 (m, 8H), 7.14 (t, J = 7.5 Hz, 1H), 7.02 (d, J = 8.2 Hz, 2H), 6.60 (s, 1H), 5.48 (s, 2H), 3.63 (bs, 4H), 3.42 (s, 2H), 2.35 (bs, 2H), 2.04 (bs, J = 11.7 Hz, 2H). TOF ES+ MS: (M + H) 444.2, (M + Na) 466.2. HPLC t_R = 6.12 min, > 95%.

(1-(4-Chlorobenzyl)-1H-indol-2-yl)(4-(indoline-1-carbonyl)piperidin-1-yl)methanone. **(CCG-206328)** The following was added sequentially to DCM (5 mL): 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carboxylic acid (100 mg, 0.252 mmol), DIPEA (1.4 mL, 0.756 mmol), EDC·HCl (60 mg, 0.302 mmol), HOBT (48 mg, 0.302 mmol), and indoline (0.040 mL, 0.302 mmol). The mixture was stirred for 24 h at rt, at which time the solution diluted with EtOAc and washed with water (2X) and brine (1X). The organic phase was dried with magnesium sulfate and concentrated *in vacuo*. The resulting residue was dissolved in EtOAc and precipitated out hexanes, and then collected over a filter and washed with diethyl ether to give the title compound as a white solid. Yield: ^{1}H NMR (400 MHz, Chloroform-d) δ 7.66 (d, J = 7.8 Hz, 1H), 7.33 (t, J = 8.8 Hz, 1H), 7.30 − 7.13 (m, 8H), 7.04 (d, J = 8.2 Hz, 2H), 6.65 (s, 1H), 5.48 (s, 2H), 4.36 (s, 2H), 3.39 (t, J = 5.8 Hz, 2H), 3.08 (q, J = 9.6, 9.1 Hz, 2H), 2.99 − 2.82 (m, 2H), 2.71 − 2.64 (m, 2H), 2.26 (tt, J = 11.3, 3.9 Hz, 1H), 1.76 (s, 2H), 1.50 (s, 2H). TOF ES+ MS: (M + H) 498.2. HPLC t_R = 8.58 min, >95% purity.

1-(1-(4-Chlorobenzyl)-1H-pyrrole-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-206381).** The following was added

sequentially to DCM: 1-(1-(4-chlorobenzyl)-1H-pyrrole-2-carbonyl)piperidine-4-carboxylic acid (600 mg, 1.73 mmol), TEA (0.725 mL, 5.19 mmol), EDC·HCl (365 mg, 1.903 mmol), and HOBt (291 mqg, 1.903 mmol). This was allowed to stir at rt for 30 min, at which time pyridylethylamine (0.227 mL, 1.903 mmol) was added. Stirring continued for 18 h. At this time, the DCM and TEA were stripped off, and the residue was taken up in EtOAc and washed with 10% aqueous sodium carbonate (3X). The organic phase was collected, dried over magnesium sulfate, and concentrated *in vacuo*. The resulting solid/oil mixture was recrystallized from EtOAc to afford the title compound as small, white crystals. Yield: 586 mg, 1.29 mmol, 75%. ^{1}HNMR (400 MHz, CDCl3) δ 8.50 (d, J = 5.7 Hz, 2H), 7.21 (d, J = 8.3 Hz, 2H), 7.09 (d, J = 5.6 Hz, 2H), 7.02 (d, J = 8.2 Hz, 2H), 6.78 (s, 1H), 6.30 (dd, J = 3.6, 1.2 Hz, 1H), 6.13−6.09 (m, 1H), 5.48 (t, J = 5.4 Hz, 1H), 5.26 (s, 2H), 4.32 (d, J = 12.7 Hz, 2H), 3.53 (q, J = 6.8 Hz, 2H), 2.82 (t, J = 7.0 Hz, 4H), 2.19 (tt, J = 11.2, 3.6 Hz, 1H), 1.68 (d, J = 14.4 Hz, 2H), 1.35 (q, J = 11.0, 9.8 Hz, 2H). Anal. Calcd for C25H27ClN4O2: C, 66.58%; H, 6.04%; N, 12.42%. Found: C, 66.68%; H, 6.32%; N, 12.38%. TOF ES+ MS: (M + H) 451.2, (M + Na) 473.2. HPLC t_R = 5.03 min, >95% purity.

1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-N-((2,3-dihydro-1H-inden-2-yl)methyl)piperidine-4-carboxamide **(CCG-206382).** The following was added sequentially to DCM (8 mL): 1-(1-(4-chlorobenzyl)-1*H*-indole-2-carbonyl)piperidine-4-carboxylic acid (180 mg, 0.454 mmol), DIPEA (0.317 mL, 1.814 mmol), EDC·HCl (96 mg, 0.499 mmol), HOBT (76 mg, 0.499 mmol), and crude (2,3-dihydro-1*H*-inden-2-yl)methanamine (105 mg, 0.713 mmol). The mixture was stirred for 18 h at RT, at which

time the solution diluted with a 1:1 solution of EtOAc:Et$_2$O and washed with 1 M HCl (1X), 10% aq. Na$_2$CO$_3$ (1X). The organic phase was dried with MgSO$_4$ and concentrated *in vacuo*. Purification was accomplished via silica flash chromatography (60 g silica, 50% EtOAc:Hex), followed by EtOAc/Hex tritutation to give the title compound as a white powder. Yield: 137 mg, 0.260 mmol, 57%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.66 (d, *J* = 7.8 Hz, 1H), 7.33 (t, *J* = 8.8 Hz, 1H), 7.30 − 7.13 (m, 8H), 7.04 (d, *J* = 8.2 Hz, 2H), 6.65 (s, 1H), 5.48 (s, 2H), 4.36 (s, 2H), 3.39 (t, *J* = 5.8 Hz, 2H), 3.08 (q, *J* = 9.6, 9.1 Hz, 2H), 2.99 − 2.82 (m, 2H), 2.71 − 2.64 (m, 2H), 2.26 (tt, *J* = 11.3, 3.9 Hz, 1H), 1.76 (s, 2H), 1.50 (s, 2H). TOF ES+ MS: (M + H) 526.2, (M + Na) 548.2. HPLC t_R = 8.25 min, > 95%.

N-Benzyl-1-(1-(4-chlorobenzyl)-7-methyl-1H-indole-2-carbonyl)piperidine-4-carboxamide (**CCG-206447**). The following was added to sequentially to anhydrous DMF (2 mL): 1-(4-chlorobenzyl)-7-methyl-1*H*-indole-2-carboxylic acid (50 mg, 0.167 mmol), DIPEA (0.10 mL, 0.584 mmol), EDC·HCl (38 mg, 0.200 mmol), and HOBT (31 mg, 0.200 mmol). This was allowed to stir for 30 min at RT, after which time the crude *N*-benzylpiperidine-4-carboxamide (36 mg, 0.20 mmol) was added. The reaction was stirred at RT for 18 h, at which time the reaction was diluted with 1:1 EtOAc:Et$_2$O and washed with 10% aq. Na$_2$CO$_3$ (2x), dried over MgSO$_4$, and concentrated. The residue was purified via silica flash chromatography (20 g silica, 50% EtOAc:Hex) to afford the title compound as a white powder. Yield: 39 mg, 0.078 mmol, 47%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.51 (d, *J* = 7.7 Hz, 1H), 7.37 − 7.33 (m, 2H), 7.31 − 7.26 (m, 3H), 7.19 (dd, *J* = 10.0, 3.6 Hz, 2H), 7.08 − 6.99 (m, 2H), 6.82 (d, *J* = 8.2 Hz, 2H), 6.64 (s, 1H),

5.73 (s, 2H), 4.58 (bs, 1H), 4.45 (t, *J* = 5.9 Hz, 2H), 4.11 (bs, 1H), 2.83 (bs, 2H), 2.58 (s, 3H), 2.34 – 2.27 (m, 1H), 1.80 (bs, 2H), 1.37 (bs, 2H). TOF ES+ MS: (M + H) 500.2, (M + Na) 522.2. HPLC *t*R = 8.01 min, > 95%.

(1-(4-Fluorobenzyl)-1H-indol-2-yl)(4-(hydroxymethyl)piperidin-1-yl)methanone

(CCG-206462). The following was added sequentially to DMF (10 mL): 1-(4-fluorobenzyl)-1*H*-indole-2-carboxylic acid (1.175 g, 4.36 mmol), DIPEA (2.29 mL, 13.09 mmol), EDC·HCl (1.00 g, 5.24 mmol), and HOBT (802 mg, 5.24 mmol). This solution was allowed to stir at RT for 1.5 h, at which time 4-piperidinemethanol (754 mg, 6.55 mmol) was added. Stirring continued for 22 h at RT, at which time the solution was partitioned between H₂O and 1:1 solution of EtOAc:Et₂O. The organic extract was then washed with saturated aq. Na₂CO₃, dried with MgSO₄, and concentrated *in vacuo*. Purification was accomplished via silica flash chromatography (100 g silica, 80% EtOAc/Hexanes.) Resulting residue was crystallized via Et₂O:Hex trituration to give the title compound as a white solid. Yield: 1.51 g, 4.11 mmol, 94%. ¹H NMR (400 MHz, Chloroform-d) δ 7.63 (d, *J* = 7.9 Hz, 1H), 7.37 (d, *J* = 8.3 Hz, 1H), 7.29 – 7.22 (m, 1H), 7.14 (t, *J* = 7.5 Hz, 1H), 7.10 – 7.04 (m, 2H), 6.91 (t, *J* = 8.2 Hz, 2H), 6.60 (s, 1H), 5.46 (s, 2H), 4.64 (bs, 1H), 4.14 (bs, 1H), 3.42 (t, *J* = 5.1 Hz, 2H), 2.76 (s, 2H), 1.44 (t, *J* = 5.0 Hz, 1H), 1.05 (bs, 1H), 0.71 (bs, 1H). HPLC *t*R = 6.62 min, >95% purity.

2-Benzyl-5-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)octahydro-1H-pyrrolo[3,4-c]pyridin-1-one. **(CCG-206485)** 5-(1-(4-chlorobenzyl)-1*H*-indole-2-carbonyl)octahydro-1*H*-pyrrolo[3,4-c]pyridin-1-one (64 mg, 0.245 mmol) was dissolved in anhydrous DMF

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(5 mL). A 60% oil suspension of NaH (29 mg, 0.735 mmol) was added at RT under N_2 and stirred for 1 h. BnBr (0.035 mL, 0.294 mmol) was added and stirring continued for 18 h under the same conditions. After this time, the solution was diluted with EtOAc and washed with 10% aq. Na_2CO_3 (2X), dried with $MgSO_4$, and concentrated *in vacuo*. Purification was done via silica flash chromatography (25 g silica, 80% EtOAc:Hex.) The residue was dissolved in DCM, and rapid solvent removal afforded the title compound as a white powder. Yield: 56 mg, 0.112 mmol, 72%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.71 (d, *J* = 7.9 Hz, 1H), 7.37 − 7.26 (m, 5H), 7.26 − 7.20 (m, 5H), 7.15 (t, *J* = 7.4 Hz, 1H), 7.10 (d, *J* = 8.2 Hz, 2H), 5.49 (d, *J* = 3.7 Hz, 2H), 4.55 (d, *J* = 14.6 Hz, 2H), 4.39 (d, *J* = 14.5 Hz, 1H), 4.12 (bs, 1H), 3.43 − 3.30 (m, 2H), 2.98 − 2.89 (m, 1H), 2.84 (d, *J* = 1.6 Hz, 1H), 2.63 − 2.44 (m, 2H), 1.75 (bs, 1H), 1.41 (bs, *J* = 13.5 Hz, 1H). TOF ES+ MS: (M + H) 498.2, (M + Na) 520.2. HPLC t_R = 8.23 min, >95% purity.

(1-(4-Fluorobenzyl)-1H-indol-2-yl)(4-(((4-methylphenethyl)amino)methyl)-piperidin-1-yl)methanone **(CCG-206486).** 1-(1-(4-fluorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carbaldehyde (46 mg, 0.126 mmol) was dissolved in THF (2 mL) and 2-(p-tolyl)ethanamine (0.030 mL, 0.189 mmol) was added. The solution was stirred at rt for 10 h, at which time the THF was removed *in vacuo* and the residue dissolved in EtOH (5 mL), and sodium cyanoborohydride (24 mg, 0.349 mmol) and a catalytic drop of glacial acetic acid were added. Stirring was permitted for 14 h, at which time the solvent was removed *in vacuo*, the residue was taken up in EtOAc. The organic phase was washed with 10% aq. sodium carbonate, dried over magnesium sulfate, and concentrated. Purification was accomplished via silica gel flash chromatography (30 g

silica gel, 5:95 7M methanolic ammonia:ethyl acetate.) The title compound was obtained

as a yellow oil. Yield: 26 mg, 0.054 mmol, 43%. ^{1}H NMR (500 MHz, Chloroform-d) δ

7.64 (d, J = 8.4 Hz, 1H), 7.38 (d, J = 8.3 Hz, 1H), 7.27 (t, J = 7.7 Hz, 2H), 7.18 − 7.13

(m, 1H), 7.12 − 7.06 (m, 5H), 6.91 (t, J = 8.5 Hz, 2H), 6.60 (s, 1H), 5.47 (s, 2H), 4.62 (bs,

1H), 4.09 (bs, 1H), 2.84 (t, J = 7.0 Hz, 2H), 2.76 (t, J = 7.0 Hz, 2H), 2.70 (bs, 2H), 2.44

(d, J = 5.9 Hz, 2H), 2.32 (s, 3H), 1.79 − 1.62 (m, 5H). TOF ES+ MS: (M + H) 484.1, (M

+ Na) 506.1. HPLC t_R = 6.43 min, >95% purity.

(R)-(1-(4-Fluorobenzyl)-1H-indol-2-yl)(4-(((1-phenylethyl)amino)methyl)-

piperidin-1-yl)methanone **(CCG-206499).** 1-(1-(4-fluorobenzyl)-1H-indole-2-

carbonyl)piperidine-4-carbaldehyde (91 mg, 0.250 mmol) was dissolved in THF (4 mL)

and (R)-α-methylbenzylamine (0.048 mL, 0.375 mmol) was added. The solution was

stirred at rt for 10 h, at which time the THF was removed *in vacuo* and the residue

dissolved in EtOH (5 mL), and sodium cyanoborohydride (47 mg, 0.749 mmol) and a

catalytic drop of glacial acetic acid were added. Stirring was permitted for 15 h, at which

time the solvent was removed *in vacuo*, the residue was taken up in EtOAc. The organic

phase was washed with 10% aq. sodium carbonate, dried over magnesium sulfate, and

concentrated. Purification was accomplished via silica gel flash chromatography (30 g

silica gel, 5:95 7M methanolic ammonia:ethyl acetate.) The title compound was obtained

as a yellow oil. Yield: 52 mg, 0.111 mmol, 44%. ^{1}H NMR (500 MHz, Chloroform-d) δ

7.64 (d, J = 7.9 Hz, 1H), 7.45 − 7.25 (m, 7H), 7.16 (t, J = 7.5 Hz, 1H), 7.10 − 7.01 (m,

2H), 6.91 − 6.80 (m, 2H), 6.60 (s, 1H), 5.46 (s, 2H), 4.60 (s, 1H), 4.05 (s, 1H), 3.80 (q, J

= 6.3 Hz, 1H), 2.80 (s, 2H), 2.38 (dd, *J* = 11.6, 6.0 Hz, 1H), 2.23 (dd, *J* = 16.5, 4.8 Hz, 1H), 1.70 (d, *J* = 54.9 Hz, 4H), 1.42 (d, *J* = 6.6 Hz, 3H), 0.99 (s, 1H), 0.57 (s, 1H). TOF ES+ MS: (M + H) 470.3, (M + Na) 492.2. HPLC t_R = 5.96 min, >95% purity.

(4-((Benzylamino)methyl)piperidin-1-yl)(1-(4-fluorobenzyl)-1H-indol-2-yl)methanone. **(CCG-206500).** 1-(1-(4-fluorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carbaldehyde (52 mg, 0.143 mmol) was dissolved in THF (2 mL) and benzylamine (0.025 mL, 0.285 mmol) was added. The solution was stirred at RT for 10 h, at which time the THF was removed *in vacuo* and the residue dissolved in EtOH (5 mL), and NaCNBH₃ (18 mg, 0.285 mmol) and a catalytic drop of glacial AcOH were added. Stirring was permitted for 14 h, at which time the solvent was removed *in vacuo*, the residue was taken up in EtOAc. The organic phase was washed with 10% aq. Na₂CO₃, dried MgSO₄, and concentrated. Purification was accomplished via silica flash chromatography (30 g silica gel, 5:95 7M methanolic ammonia:EtOAc.) The title compound was obtained as a yellow oil. Yield: 23 mg, 0.050 mmol, 40%. ¹H NMR (500 MHz, Chloroform-d) δ 7.65 (d, *J* = 7.9 Hz, 1H), 7.39 − 7.27 (m, 7H), 7.16 (t, *J* = 7.5 Hz, 1H), 7.09 (dd, *J* = 8.2, 5.5 Hz, 2H), 6.91 (t, *J* = 8.6 Hz, 2H), 6.61 (s, 1H), 5.48 (s, 2H), 4.65 (bs, 1H), 4.10 (bs, 1H), 3.78 (s, 2H), 2.77 (bd, *J* = 50.6 Hz, 2H), 2.46 (d, *J* = 6.4 Hz, 2H), 1.79 − 1.61 (m, 3H), 1.09 (bs, 1H), 0.66 (bs, 1H). TOF ES+ MS: (M + H) 456.2, (M + Na) 478.2. HPLC t_R = 6.04 min, >90% purity.

(S)-(1-(4-Fluorobenzyl)-1H-indol-2-yl)(4-(((1-phenylethyl)amino)methyl)-piperidin-1-yl)methanone **(CCG-206501).** 1-(1-(4-fluorobenzyl)-1*H*-indole-2-

carbonyl)piperidine-4-carbaldehyde (35 mg, 0.096 mmol) was dissolved in THF (2 mL) and (S)-α-methylbenzylamine (0.013 mL, 0.115 mmol) was added. The solution was stirred at RT for 10 h, at which time the THF was removed *in vacuo* and the residue dissolved in EtOH (5 mL), and sodium cyanoborohydride (13 mg, 0.192 mmol) and a catalytic drop of glacial AcOH were added. Stirring was permitted for 14 h, at which time the solvent was removed *in vacuo*, the residue was taken up in EtOAc. The organic phase was washed with 10% aq. Na_2CO_3, dried over $MgSO_4$, and concentrated. Purification was accomplished via silica flash chromatography (30 g silica gel, 5:95 7M methanolic ammonia:EtOAc.) The title compound was obtained as a yellow oil. Yield: 21 mg, 0.045 mmol, 47%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.64 (d, *J* = 8.0 Hz, 1H), 7.39 − 7.22 (m, 7H), 7.16 (t, *J* = 7.5 Hz, 1H), 7.08 (dd, *J* = 8.1, 5.5 Hz, 2H), 6.88 (t, *J* = 7.9 Hz, 2H), 6.59 (s, 1H), 5.47 (s, 2H), 4.61 (s, 1H), 4.05 (s, 1H), 3.74 − 3.69 (m, 1H), 2.75 (d, *J* = 49.0 Hz, 2H), 2.41 − 2.33 (m, 1H), 2.22 (dd, *J* = 11.6, 7.1 Hz, 1H), 1.59 (d, *J* = 10.4 Hz, 4H), 1.35 (d, *J* = 6.6 Hz, 3H), 1.01 (s, 1H), 0.63 (s, 1H). TOF ES+ MS: (M + H) 470.3, (M + Na) 492.2. HPLC t_R = 5.98 min, > 95% purity.

(1-(4-Fluorobenzyl)-1H-indol-2-yl)(4-(morpholinomethyl)piperidin-1-yl)methanone. **(CCG-206502)** 1-(1-(4-fluorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carbaldehyde (52 mg, 0.143 mmol) was dissolved in EtOH (5 mL) and morpholine (0.025 mL, 0.285 mmol), sodium cyanoborohydride (18 mg, 0.285 mmol), and a catalytic drop of glacial acetic acid were added. Stirring was permitted for 13 h, at which time the solvent was removed *in vacuo*, the residue was taken up in EtOAc. The organic phase was washed with 10% aq. sodium carbonate, dried over magnesium sulfate, and

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concentrated. Purification was accomplished via silica gel flash chromatography (30 g silica gel, 100% EtOAc.) The title compound was obtained as an oil. Yield: 15 mg, 0.034 mmol, 24%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.65 (d, *J* = 8.0 Hz, 1H), 7.40 (d, *J* = 8.3 Hz, 1H), 7.30 – 7.27 (m, 1H), 7.17 (t, *J* = 8.3 Hz, 1H), 7.12 – 7.07 (m, 2H), 6.97 – 6.89 (m, 2H), 6.63 – 6.59 (m, 1H), 5.49 (s, 2H), 4.65 (bs, 1H), 4.11 (bs, 1H), 3.69 (s, 4H), 2.76 (bd, *J* = 43.7 Hz, 2H), 2.39 (s, 4H), 2.11 (s, 2H), 1.83 – 1.57 (m, 3H), 0.99 (bs, 1H), 0.60 (bs, 1H). TOF ES+ MS: (M + H) 436.2, (M + Na) 458.2. HPLC t_R = 5.38 min, >95% purity.

(4-((Benzyl(methyl)amino)methyl)piperidin-1-yl)(1-(4-fluorobenzyl)-1H-indol-2-yl)methanone **(CCG-206503).** 1-(1-(4-fluorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carbaldehyde (86 mg, 0.236 mmol) was dissolved in EtOH (5 mL) and N-methylbenzylamine (0.027 mL, 0.283 mmol), NaCNBH$_3$ (30 mg, 0.472 mmol), and a catalytic drop of glacial AcOH were added. Stirring was permitted for 14 h, at which time the solvent was removed *in vacuo*, the residue was taken up in EtOAc. The organic phase was washed with 10% aq. Na$_2$CO$_3$, dried over MgSO$_4$, and concentrated. Purification was accomplished via silica flash chromatography (30 g silica gel, 100% EtOAc.) The title compound was obtained as an oil. Yield: 23 mg, 0.049 mmol, 20%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.64 (t, *J* = 7.4 Hz, 1H), 7.39 (t, *J* = 7.7 Hz, 1H), 7.35 – 7.22 (m, 6H), 7.16 (q, *J* = 7.3 Hz, 1H), 7.07 (td, *J* = 8.0, 5.4 Hz, 2H), 6.87 – 6.79 (m, 2H), 6.59 (d, *J* = 7.0 Hz, 1H), 5.47 (s, 2H), 4.61 (bs, 1H), 4.06 (bs, 1H), 3.46 (s, 2H), 2.70 (bs, 2H), 2.18 (d, *J* = 7.2 Hz, 3H), 2.10 – 1.60 (m, 5H), 0.95 (bs, 1H), 0.50 (bs, 1H). TOF ES+ MS: (M + H) 470.2, (M + Na) 492.2. HPLC t_R = 6.04 min, > 95% purity.

(1-(4-Fluorobenzyl)-1H-indol-2-yl)(4-(((2-(pyridin-4-yl)ethyl)amino)methyl)-piperidin-1-yl)methanone **(CCG-206504).** 1-(1-(4-fluorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carbaldehyde (100 mg, 0.273 mmol) was dissolved in EtOH (5 mL) and 4-(2-aminoethyl)pyridine (50 μL, 0.410 mmol), NaCNBH$_3$ (51 mg, 0.819 mmol), and a catalytic drop of glacial AcOH were added. Stirring was permitted for 10 h, at which time the solvent was removed *in vacuo* and the residue was taken up in EtOAc. The organic phase was washed with 10% aq. Na$_2$CO$_3$, dried over MgSO$_4$, and concentrated. Purification was accomplished via silica flash chromatography (30 g silica gel, 100% EtOAc.) The title compound was obtained as an oil. Yield: 51 mg, 0.109 mmol, 45%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.51 (d, *J* = 4.7 Hz, 2H), 7.64 (d, *J* = 7.9 Hz, 1H), 7.39 (d, *J* = 8.3 Hz, 1H), 7.29 − 7.26 (m, 1H), 7.17 − 7.11 (m, 3H), 7.10 − 7.06 (m, 2H), 6.97 − 6.89 (m, 3H), 6.60 (s, 1H), 5.47 (s, 2H), 4.65 (bs, 1H), 4.08 (bs, 1H), 2.87 (t, *J* = 7.1 Hz, 2H), 2.78 (t, *J* = 7.0 Hz, 2H), 2.70 (bs, 2H), 2.43 (s, 2H), 1.72 − 1.54 (m, 5H). TOF ES+ MS: (M + H) 471.3, (M + Na) 493.2. HPLC *t*$_R$ = 5.51 min, >95% purity.

1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-N-(1-(2,3-dihydro-1H-inden-2-yl)ethyl)piperidine-4-carboxamide **(CCG-206549).** The following was added sequentially to DCM (5 mL): 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carboxylic acid (109 mg, 0.275 mmol), DIPEA (0.144 mL, 0.825 mmol), EDC·HCl (63 mg, 0.330 mmol), HOBT (51 mg, 0.330 mmol), and crude 2-(1-aminoethyl)indane (~44

mg, 0.275 mmol). The mixture was stirred for 16 h at rt, at which time the solution diluted with a 1:1 solution of ethyl acetate:diethyl ether and washed with 1M HCl (1X), 10% aq. sodium carbonate (1X) and brine (1X). The organic phase was dried with magnesium sulfate and concentrated *in vacuo*. The resulting residue was dissolved in EtOAc and precipitated out hexanes, and then collected over a filter and washed with diethyl ether to give the title compound as a white solid. Yield: 59 mg, 0.109 mmol, 40%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.66 (d, *J* = 7.9 Hz, 1H), 7.33 (d, *J* = 8.3 Hz, 1H), 7.29 – 7.13 (m, 8H), 7.04 (d, *J* = 8.2 Hz, 2H), 6.65 (s, 1H), 5.48 (s, 2H), 5.30 (d, *J* = 8.9 Hz, 1H), 4.45 (s, 1H), 4.25 – 4.14 (m, 1H), 3.02 (td, *J* = 16.7, 8.3 Hz, 2H), 2.91 – 2.65 (m, 4H), 2.51 (h, *J* = 8.1 Hz, 1H), 2.22 (tq, *J* = 10.8, 4.1 Hz, 1H), 1.63 (d, *J* = 95.6 Hz, 5H), 1.21 (d, *J* = 6.7 Hz, 3H). TOF ES+ MS: (M + H) 540.2. HPLC t_R = 8.55 min, >95% purity.

1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-N-((2,3-dihydro-1H-inden-1-yl)methyl)piperidine-4-carboxamide. **(CCG-206550)** The following was added sequentially to DCM (6 mL): 1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)piperidine-4-carboxylic acid (134 mg, 0.340 mmol), DIPEA (0.178 mL, 1.019 mmol), EDC·HCl (78 mg, 0.408 mmol), HOBT (62.4 mg, 0.408 mmol), and 2-indanmethanamine hydrochloride (62 mg, 0.340 mmol). The mixture was stirred for 18 h at rt, at which time the solution diluted with a 1:1 solution of ethyl acetate:diethyl ether and washed with 1M HCl (1X), 10% aq. sodium carbonate (1X) and brine (1X). The organic phase was dried with magnesium sulfate and concentrated *in vacuo*. The resulting residue was dissolved in EtOAc and precipitated out hexanes, and then collected over a filter and washed with

diethyl ether to give the title compound as a white solid. Yield: 72 mg, 0.137 mmol, 41%.

^{1}H NMR (400 MHz, Chloroform-d) δ 7.63 (d, J = 7.9 Hz, 1H), 7.32 (d, J = 8.2 Hz, 1H), 7.27 − 7.22 (m, 3H), 7.21 − 7.13 (m, 5H), 7.01 (d, J = 8.2 Hz, 2H), 6.62 (s, 1H), 5.45 (s, 2H), 5.38 (t, J = 5.3 Hz, 1H), 4.36 (s, 2H), 3.62 (dt, J = 12.2, 5.8 Hz, 1H), 3.50 − 3.34 (m, 2H), 2.88 (tt, J = 15.8, 8.2 Hz, 4H), 2.23 (ddd, J = 13.3, 9.5, 5.8 Hz, 2H), 1.78 (ddd, J = 13.3, 8.2, 6.4 Hz, 3H), 1.49 (s, 2H). TOF ES+ MS: (M + H) 526.2. HPLC t_R = 8.35 min, >95% purity.

N-Benzyl-1-(1-(4-chlorobenzyl)-1H-imidazole-2-carbonyl)piperidine-4-carboxamide **(CCG-206586).** Ethyl 1-(4-chlorobenzyl)-1*H*-imidazole-2-carboxylate (7.28 g, 27.5 mmol), was dissolved in EtOH (10 mL) and 10% aq. NaOH (20 mL) and stirred at rt for 15 h. The solvent was then stripped off, water was added, and the solution acidified with HCl. The resulting precipitate was collected over a filter and washed with 1M HCl and dried to afford the carboxylic acid as a white powder which was taken directly into the next step. Yield: 6.506 g, 27.5 mmol, 100%.

The following was added sequentially to DMF: 1-(4-chlorobenzyl)-1H-imidazole-2-carboxylic acid (71 mg, 0.298 mmol), DIPEA (0.16 mL, 0.894 mmol), EDC·HCl (69 mg, 0.358 mmol), and HOBT (55 mg, 0.358 mmol). This was allowed to stir at rt for 30 min, at which time N-benzylpiperidine-4-carboxamide (~ 65 mg, 0.298 mmol) was added and syirring continued at rt for 14 h. Addition of water caused precipitation, which was collected over a filter and washed with water and small amount of diethyl ether and EtOAc to give the title compound as a white solid. Yield: 37 mg, 0.085 mmol, 28%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.34 − 7.22 (m, 7H), 7.12 (d, J = 8.3 Hz, 2H), 7.04 (s,

1H), 6.93 (s, 1H), 5.72 (t, *J* = 5.1 Hz, 1H), 5.36 (d, *J* = 5.1 Hz, 2H), 4.63 (t, *J* = 15.1 Hz, 2H), 4.43 (d, *J* = 5.6 Hz, 2H), 3.14 (t, *J* = 12.6 Hz, 1H), 2.82 (t, *J* = 12.8 Hz, 1H), 2.38 (tt, *J* = 11.3, 3.9 Hz, 1H), 1.95 (d, *J* = 13.4 Hz, 1H), 1.83 (d, *J* = 13.3 Hz, 1H), 1.72 − 1.55 (m, 2H). TOF ES+ MS: (M + H) 437.2, (M + Na) 459.2. HPLC t_R = 5.59 min, >95% purity.

1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-N-(3-phenylpropyl)piperidine-4-carboxamide **(CCG-206587).** The following were added sequentially to DCM (10 mL): 1-(1-(4-chlorobenzyl)-1*H*-indole-2-carbonyl)piperidine-4-carboxylic acid (100 mg, 0.252 mmol), DIPEA (132 µL, 0.756 mmol), EDC·HCl (58 mg, 0.302 mmol), HOBt (46 mg, 0.302 mmol), and then 3-phenylpropylamine (43 µL, 0.302 mmol). The reaction was allowed to stir at RT for 13 h, at which time it was diluted with EtOAc, washed with 1 M HCl (3X), H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), dried over $MgSO_4$, and concentrated *in vacuo*. The residue was the purified by silica flash chromatography (20 g silica, 70% EtOAc:Hex) to provide the desired material as a colorless oil. Yield: 101 mg, 0.197 mmol, 78%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.65 (d, *J* = 7.9 Hz, 1H), 7.37 − 7.23 (m, 5H), 7.24 − 7.12 (m, 6H), 6.64 (s, 1H), 5.45 (s, 2H), 5.39 (t, *J* = 5.4 Hz, 1H), 4.50 (bs, 1H), 4.21 (bs, 1H), 3.29 (q, *J* = 6.5 Hz, 2H), 2.84 (bs, 2H), 2.65 (t, *J* = 7.5 Hz, 2H), 2.20 (tt, *J* = 11.0, 3.5 Hz, 1H), 1.85 (p, *J* = 7.5 Hz, 2H), 1.72 (bs, 2H), 1.44 (bs, 2H). TOF ES+ MS: (M + H) 515.1, (M + Na) 537.0. HPLC t_R = 8.16 min, > 90%.

1-(1-(4-Chlorobenzyl)-1H-pyrrole-2-carbonyl)-N-((2,3-dihydro-1H-inden-2-yl)methyl)azetidine-3-carboxamide **(CCG-208793).** Methyl 1-(1-(4-chlorobenzyl)-1*H*-pyrrole-2-carbonyl)azetidine-3-carboxylate **29** (300 mg, 0.90 mmol) was dissolved in 200 proof EtOH (10 mL) and 10% aq. NaOH (10 mL) was added. The reaction was allowed to stir at RT for 24 h, at which time the solvent was stripped off *in vacuo* and the residue was taken up in H₂O and acidified to pH 1 by addition of conc. HCl at 0°C. The resulting precipitate was collected over a filter and dried by aspirator then high vacuum to afford the desired carboxylic acid as a white powder. Yield: 250 mg, 0.78 mmol, 87%.

1-(1-(4-chlorobenzyl)-1*H*-pyrrole-2-carbonyl)azetidine-3-carboxylic acid (50 mg, 0.16 mmol) was added to DCM (5 mL), followed by indanyl amine **75** (29 mg, 0.17 mmol), TEA (66 μL, 0.47 mmol), EDC·HCl (33 mg, 0.17 mmol), and HOBT (26 mg, 0.17 mmol). The reaction was allowed to stir at RT for 21 h, at which time the reaction was concentrated and the residue taken up in EtOAc. This solution was then washed with 1M HCl (2X), H₂O (3X) and 10% aq. Na₂CO₃ (3X), and brine (1X), dried (MgSO₄), and concentrated. The residue was purified by silica flash chromatography (4 g silica, 10% to 60% EtOAc:Hex) to provide the final compound as a white powder. Yield: 39 mg, 0.09 mmol, 56%. ¹H NMR (500 MHz, Chloroform-d) δ 7.28 − 7.24 (m, 3H), 7.21 − 7.18 (m, 2H), 7.16 − 7.13 (m, 2H), 7.09 − 7.04 (m, 2H), 6.81 (dd, *J* = 2.6, 1.6 Hz, 1H), 6.52 (dd, *J* = 4.0, 1.5 Hz, 1H), 6.19 − 6.16 (m, 1H), 5.56 (d, *J* = 9.0 Hz, 3H), 4.30 (bs, 4H), 3.42 (t, *J* = 6.0 Hz, 2H), 3.22 (p, *J* = 7.4 Hz, 1H), 3.09 (q, *J* = 10.0, 9.3 Hz, 2H), 2.75 − 2.65 (m, 3H). TOF ES+ MS: (M + H) 448.2, (M + Na) 470.2. HPLC *t*$_R$ = 7.56 min, > 95%.

N¹-(4-Chlorobenzyl)-N4-(2-(pyridin-4-yl)ethyl)piperidine-1,4-dicarboxamide

(208825). Ethyl 1-((4-chlorobenzyl)carbamoyl)piperidine-4-carboxylate **31a** (810 mg, 2.50 mmol) was dissolved in 200 proof EtOH (20 mL) at RT, followed by the addition of 10% aq. NaOH, which elicited precipitation. The mixture was then stirred at 50 °C for 2 h over which time it became a solution. The reaction was then allowed to cool to RT and the solvent was then removed *in vacuo*. The residue was taken up in H_2O and acidified to pH 1 with conc. HCl at 0 °C and the resulting precipitate was collected over a filter, washed with cold 1 M aq. HCl, and dried over the filter then under high vacuum to give the desired compound as a white powder. No further purification as necessary. Yield: 675 mg, 2.27 mmol, 91%.

1-((4-chlorobenzyl)carbamoyl)piperidine-4-carboxylic acid (60 mg, 0.20 mmol) and TEA (93 µL, 0.61 mmol) were dissolved in DCM (5 mL), followed by EDC·HCl (47 mg, 0.22 mmol), HOBT (37 mg, 0.22 mmol), and amine **27** (30 µL, 0.20 mmol). The reaction was allowed to stir at RT for 15 h. After this time, the reaction was diluted with EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried with $MgSO_4$ and concentrated. The residue was then crystallized from EtOAc and washed with Et_2O to provide an off-white, slightly tanned solid. Yield: 42 mg, 0.11 mmol, 52%. ¹H NMR (500 MHz, Chloroform-d) δ 8.52 (d, *J* = 4.4 Hz, 2H), 7.32 − 7.20 (m, 4H), 7.11 (d, *J* = 4.4 Hz, 2H), 5.56 (s, 1H), 4.79 (d, *J* = 5.9 Hz, 1H), 4.37 (d, *J* = 5.2 Hz, 2H), 3.96 (d, *J* = 13.2 Hz, 2H), 3.55 (q, *J* = 6.4, 5.8 Hz, 2H), 2.85 − 2.78 (m, 4H), 2.19 (t, *J* = 11.5 Hz, 1H), 1.77 (d, *J* = 13.0 Hz, 2H), 1.63 (dd, *J* = 18.8, 5.5 Hz, 2H). TOF ES+ MS: (M + H) 401.2, (M + Na) 423.2. HPLC t_R = 4.61 min, > 95% purity.

N¹-(4-Chlorobenzyl)-N⁴-((2,3-dihydro-1H-inden-2-yl)methyl)piperidine-1,4-

dicarboxamide **(CCG-208826).** Ethyl 1-((4-chlorobenzyl)carbamoyl)piperidine-4-carboxylate **31a** (810 mg, 2.50 mmol) was dissolved in 200 proof EtOH (20 mL) at RT, followed by the addition of 10% aq. NaOH, which elicited precipitation. The mixture was then stirred at 50 °C for 2 h over which time it became a solution. The reaction was then allowed to cool to RT and the solvent was then removed *in vacuo*. The residue was taken up in H_2O and acidified to pH 1 with conc. HCl at 0 °C and the resulting precipitate was collected over a filter, washed with cold 1 M aq. HCl, and dried over the filter then under high vacuum to give the desired compound as a white powder. No further purification as necessary. Yield: 675 mg, 2.27 mmol, 91%.

1-((4-chlorobenzyl)carbamoyl)piperidine-4-carboxylic acid (60 mg, 0.20 mmol) and TEA (93 µL, 0.61 mmol) were dissolved in DCM (5 mL), followed by EDC·HCl (47 mg, 0.22 mmol), HOBT (37 mg, 0.22 mmol), and amine **27** (30 µL, 0.20 mmol). The reaction was allowed to stir at RT for 16 h. After this time, the reaction was diluted with EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried with $MgSO_4$ and concentrated. The residue was then crystallized from EtOAc and washed with Et_2O to provide a tan solid. Yield: 47 mg, 0.11 mmol, 54%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.32 − 7.28 (m, 2H), 7.25 (d, *J* = 8.4 Hz, 2H), 7.21 − 7.18 (m, 2H), 7.16 − 7.13 (m, 2H), 5.57 (s, 1H), 4.39 (s, 2H), 3.98 (dt, *J* = 13.4, 3.7 Hz, 2H), 3.38 (t, *J* = 6.0 Hz, 2H), 3.08 (q, *J* = 10.3, 9.4 Hz, 2H), 2.85 (ddd, *J* = 13.3, 11.8, 2.8 Hz, 2H), 2.70 − 2.64 (m, 3H), 2.23 (ddt, *J* = 11.4, 7.7, 3.8 Hz, 1H), 1.83 − 1.58 (m, 4H). TOF ES+ MS: (M + H) 426.2. HPLC t_R = 6.69 min, > 95%.

N^1-(4-Chlorophenethyl)-N^4-(2-(pyridin-4-yl)ethyl)piperidine-1,4-dicarboxamide

(CCG-208827). Ethyl 1-((4-chlorophenethyl)carbamoyl)-piperidine-4-carboxylate **31b** (807 mg, 2.382 mmol) was dissolved in EtOH (20 mL), and 10% aq NaOH (15 mL) was added. The resulting mixture was warmed to 50 °C and stirred for 2 h. The now homogeneous solution was then concentrated in vacuo, a small amount of water was added back, and the solution chilled in an ice bath. The solution was then acidified with concentrated HCl, and the resulting precipitate was collected over a filter, washed with 1 M HCl and ice-cold water, filter-dried, then further dried under high vacuum to afford 1-((4-chlorophenethyl)carbamoyl)piperidine-4-carboxylic acid as a white powder. Yield: 631 mg (85%). ^{1}H NMR (400 MHz, DMSO-d6) δ 12.21 (s, 1H), 7.30 (d, J = 8.4 Hz, 2H), 7.18 (d, J = 8.4 Hz, 2H), 6.54 (t, J = 5.4 Hz, 1H), 3.80 (d, J = 13.3 Hz, 2H), 3.18 (q, J = 7.0 Hz, 2H), 2.76−2.62 (m, 4H), 2.36 (ddt, J = 11.0, 7.6, 3.9 Hz, 1H), 1.75−1.64 (m, 2H), 1.40−1.25 (m, 2H).

The carboxylic acid (60 mg, 0.193 mmol), TEA (81 μL, 0.579 mmol), EDC·HCl (41 mg, 0.212 mmol), HOBT (33 mg, 0.212 mmol), and 4-(2-aminoethyl)pyridine **27** (25 μL, 0.212 mmol) were all added sequentially to DCM (5 mL) at RT. This solution was allowed to stir for 16 h at RT, after which time the reaction was diluted with ethyl acetate and washed with water (3×), 10% aq sodium carbonate (3×), and brine (1×), then dried (magnesium sulfate) and concentrated in vacuo. The resulting residue was then crystallized from ethyl acetate and washed with diethyl ether to provide the title compound as a tan solid. Yield: 46 mg (57%). ^{1}HNMR (400 MHz, CDCl3) δ 8.61−8.26 (m, 2H), 7.30−7.19 (m, 2H), 7.17−7.01 (m, 4H), 5.91−5.66 (m, 1H), 4.59 (dd, J = 13.4, 5.6 Hz, 1H), 3.86 (d, J = 13.2 Hz, 2H), 3.53 (q, J = 6.7 Hz, 2H), 3.42 (q, J = 6.8 Hz, 2H),

2.82 (t, J = 6.8 Hz, 2H), 2.79−2.61 (m, 4H), 2.17 (t, J = 9.6 Hz, 1H), 1.73 (d, J = 12.9 Hz, 2H), 1.57 (qd, J = 12.9, 12.5, 3.8 Hz, 2H). TOF ES+ MS: (M + H) 415.2, (M + Na) 437.2. HPLC t_R = 4.70 min, > 95%.

N^1-*(4-Chlorophenethyl)-N4-((2,3-dihydro-1H-inden-2-yl)methyl)piperidine-1,4-*

dicarboxamide **(CCG-208828).** Ethyl 1-((4-chlorophenethyl)carbamoyl)-piperidine-4-carboxylate **31b** (807 mg, 2.382 mmol) was dissolved in EtOH (20 mL), and 10% aq NaOH (15 mL) was added. The resulting mixture was warmed to 50 °C and stirred for 2 h. The now homogeneous solution was then concentrated in vacuo, a small amount of water was added back, and the solution chilled in an ice bath. The solution was then acidified with concentrated HCl, and the resulting precipitate was collected over a filter, washed with 1 M HCl and ice-cold water, filter-dried, then further dried under high vacuum to afford 1-((4-chlorophenethyl)carbamoyl)piperidine-4-carboxylic acid as a white powder. Yield: 631 mg (85%). ^{1}H NMR (400 MHz, DMSO-d6) δ 12.21 (s, 1H), 7.30 (d, J = 8.4 Hz, 2H), 7.18 (d, J = 8.4 Hz, 2H), 6.54 (t, J = 5.4 Hz, 1H), 3.80 (d, J = 13.3 Hz, 2H), 3.18 (q, J = 7.0 Hz, 2H), 2.76−2.62 (m, 4H), 2.36 (ddt, J = 11.0, 7.6, 3.9 Hz, 1H), 1.75−1.64 (m, 2H), 1.40−1.25 (m, 2H).

The carboxylic acid (60 mg, 0.19 mmol), TEA (81 μL, 0.58 mmol), EDC·HCl (41 mg, 0.21 mmol), HOBT (33 mg, 0.21 mmol), and indanyl amine **75** (39 mg, 0.21 mmol) were all added sequentially to DCM (5 mL) at RT. This solution was allowed to stir for 15 h at RT, after which time the reaction was diluted with EtOAc and washed with 05M HCl (3X), H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried (magnesium sulfate) and concentrated in vacuo. The resulting residue was then crystallized from

EtOAc to give tan crystals. Yield: 55 mg, 0.12 mmol, 64%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.28 − 7.27 (m, 2H), 7.21 − 7.18 (m, 2H), 7.16 − 7.12 (m, 4H), 5.54 (s, 1H), 4.45 (s, 1H), 3.88 (d, J = 9.9 Hz, 1H), 3.46 (q, J = 6.7 Hz, 2H), 3.38 (t, J = 6.0 Hz, 2H), 3.08 (q, J = 7.3, 6.5 Hz, 2H), 2.83 − 2.75 (m, 3H), 2.72 − 2.65 (m, 3H), 2.21 (tt, J = 11.5, 3.9 Hz, 1H), 1.79 (dd, J = 13.0, 3.2 Hz, 2H), 1.62 (td, J = 12.7, 12.2, 4.4 Hz, 4H). TOF ES+ MS: (M + H) 440.2, (M + Na) 462.2. HPLC t_R = 7.32 min, > 95%.

N-Benzyl-1-(1-(4-chlorobenzyl)-1H-pyrrole-2-carbonyl)azetidine-3-carboxamide

(CCG-208829). Methyl 1-(1-(4-chlorobenzyl)-1*H*-pyrrole-2-carbonyl)azetidine-3-carboxylate **29** (300 mg, 0.90 mmol) was dissolved in 200 proof EtOH (10 mL) and 10% aq. NaOH (10 mL) was added. The reaction was allowed to stir at RT for 24 h, at which time the solvent was stripped off *in vacuo* and the residue was taken up in H_2O and acidified to pH 1 by addition of conc. HCl at 0°C. The resulting precipitate was collected over a filter and dried by aspirator then high vacuum to afford the desired carboxylic acid as a white powder. Yield: 250 mg, 0.78 mmol, 87%.

1-(1-(4-chlorobenzyl)-1*H*-pyrrole-2-carbonyl)azetidine-3-carboxylic acid (50 mg, 0.16 mmol) was added to DCM (5 mL), followed by benzylamine (19 μL, 0.17 mmol), TEA (66 μL, 0.47 mmol), EDC·HCl (33 mg, 0.17 mmol), and HOBT (26 mg, 0.17 mmol). The reaction was allowed to stir at RT for 15 h, at which time the reaction was concentrated and the residue taken up in EtOAc. This solution was then washed with 0.5M HCl (2X), H_2O (3X) and 10% aq. Na_2CO_3 (3X), and brine (1X), dried (MgSO₄), and concentrated. The residue was purified by silica flash chromatography (4 g silica, 30% to 60% EtOAc:Hex) to provide the final compound as a white powder. Yield: 29

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mg, 0.07 mmol, 45%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.38 – 7.23 (m, 7H), 7.06 (d, J = 8.2 Hz, 2H), 6.80 (s, 1H), 6.53 (d, J = 3.5 Hz, 1H), 6.19 – 6.16 (m, 1H), 5.82 (s, 1H), 5.54 (s, 2H), 4.57 (s, 1H), 4.48 (d, J = 5.7 Hz, 2H), 4.32 (s, 2H), 3.28 (p, J = 7.4 Hz, 1H). TOF ES+ MS: (M + H) 408.1, (M + Na) 430.1. HPLC t_R = 7.06 min, > 95%.

1-(3-(4-Chlorophenyl)propanoyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide (**CCG-208846**). Ethyl 1-(3-(4-chlorophenyl)propanoyl)piperidine-4-carboxylate **33b** (850 mg, 2.62 mmol) was dissolved in EtOH (10 mL), followed by the addition of 10% aq. NaOH (10 mL). This was stirred 11 h at RT, after which time a majority of the solvent was stripped off *in vacuo*. The remaining concentrated aq. solution was chilled to 0 °C and acidified to pH<1 with conc. HCl. The aq. solution was decanted off of the resulting gummy residue. H$_2$O was then added, sonicated, and decanted. The residue was then dried under high vacuum, and the thick viscous residue was sonicated in Et$_2$O to afford the carboxylic acid as a white solid that was collected over a filter. Yield: 730 mg, 2.47 mmol, 94%.

1-(3-(4-chlorophenyl)propanoyl)piperidine-4-carboxylic acid (50 mg, 0.17 mmol) was added to DCM (2 mL) at RT followed by TEA (71 μL, 0.51 mmol), EDC ·HCl (36 mg, 0.19 mmol), HOBT (28 mg, 0.19 mmol), and amine **27** (22 μL, 0.19 mmol). The reaction was allowed to stir at RT for 14 h, after which time the DCM was removed *in vacuo* and the residue taken up in EtOAc, which was subsequently washed with 0.5 M aq. HCl (1X), H$_2$O (2X), 10% aq. Na$_2$CO$_3$ (2X), and brine (1X), then dried (MgSO$_4$) and decanted. The material was crystallized from the dropwise addition of EtOAc to boiling hexanes, then cooling of the solution. Yield: 42 mg, 0.11 mmol, 62%. ^{1}H NMR (500

MHz, Chloroform-d) δ 8.52 – 8.48 (m, 2H), 7.29 – 7.20 (m, 2H), 7.17 – 7.07 (m, 4H), 5.66 (s, 1H), 4.57 (d, J = 12.9 Hz, 1H), 3.81 (d, J = 13.2 Hz, 1H), 3.54 (q, J = 5.9 Hz, 2H), 2.96 – 2.89 (m, 3H), 2.82 (t, J = 6.1 Hz, 2H), 2.63 – 2.54 (m, 3H), 2.22 (dt, J = 11.2, 5.8 Hz, 1H), 1.79 – 1.69 (m, 2H), 1.60 – 1.46 (m, 2H). TOF ES+ MS: 400.2 (M + H), 422.2 (M + Na). HPLC t_R = 4.89 min, > 95% purity.

1-(3-(4-Chlorophenyl)propanoyl)-N-((2,3-dihydro-1H-inden-2-yl)methyl)piperidine-4-carboxamide **(CCG-208847).** Ethyl 1-(3-(4-chlorophenyl)propanoyl)piperidine-4-carboxylate **33b** (850 mg, 2.62 mmol) was dissolved in EtOH (10 mL), followed by the addition of 10% aq. NaOH (10 mL). This was stirred 11 h at RT, after which time a majority of the solvent was stripped off *in vacuo*. The remaining concentrated aq. solution was chilled to 0 °C and acidified to pH<1 with conc. HCl. The aq. solution was decanted off of the resulting gummy residue. H_2O was then added, sonicated, and decanted. The residue was then dried under high vacuum, and the thick viscous residue was sonicated in Et_2O to afford the carboxylic acid as a white solid that was collected over a filter. Yield: 730 mg, 2.47 mmol, 94%.

The following were added sequentially to DCM (2 mL) at RT: 1-(2-(4-chlorophenyl)acetyl)piperidine-4-carboxylic acid (50 mg, 0.17 mmol), TEA (71 µL, 0.51 mmol), EDC·HCl (36 mg, 0.19 mmol), HOBT (29 mg, 0.19 mmol), and indanyl amine **75** (34 mg, 0.19 mmol). This solution was stirred a RT for 15 h, at which time the DCM was removed *in vacuo* and the residue was diluted with EtOAc and washed with 1M HCl (3X), H_2O (2X), 10% aq. Na_2CO_3 (2X), and brine (1X), then dried ($MgSO_4$) and concentrated. The crude residue was purified by crystallization from boiling EtOAc to

give crystals. Yield: 44 mg, 0.10 mmol, 61%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.26 (d, J = 8.7 Hz, 2H), 7.21 − 7.18 (m, 2H), 7.17 − 7.12 (m, 4H), 5.54 (t, J = 5.9 Hz, 1H), 4.58 (dd, J = 13.6, 3.6 Hz, 1H), 3.83 (dt, J = 14.4, 3.9 Hz, 1H), 3.38 (t, J = 5.7 Hz, 2H), 3.08 (q, J = 7.5, 6.6 Hz, 2H), 3.00 − 2.89 (m, 3H), 2.74 − 2.65 (m, 3H), 2.60 (dd, J = 8.6, 6.3 Hz, 2H), 2.26 (tt, J = 11.4, 3.9 Hz, 1H), 1.83 − 1.74 (m, 2H), 1.60 − 1.47 (m, 2H). TOF ES+ MS: (M + H) 425.2, (M + Na) 447.2. HPLC t_R = 7.14 min, > 95%.

1-(2-(4-Chlorophenyl)acetyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-208848)**. Ethyl 1-(2-(4-chlorophenyl)acetyl)piperidine-4-carboxylate **3a** (811 mg, 2.62 mmol) was added to EtOH (15 mL) and 10% aq. NaOH (5 mL). This mixture was heated to 50 °C and stirred for 5 h, then allowed to cool to RT. Solvent was removed *in vacuo* until precipitation began to occur, at which point the material was cooled to 0 °C and acidified to pH<1 by the dropwise addition of conc. HCl. The solution was decanted off the resulting gum, and the residue was air dried, then dried under high vacuum to afford a highly gummy material that could be solidified by sonication in Et$_2$O to provide the carboxylic acid as a white powder without further purification. Yield: 723 mg, 2.57 mmol, 98%.

The following were added sequentially to DCM (5 mL) at RT: 1-(2-(4-chlorophenyl)acetyl)piperidine-4-carboxylic acid (60 mg, 0.22 mmol), TEA (93 μL, 0.67 mmol), EDC·HCl (47 mg, 0.25 mmol), HOBT (37 mg, 0.25 mmol), and amine **27** (29 μL, 0.25 mmol). This solution was stirred a RT for 18 h, at which time the reaction was diluted with EtOAc and washed with H$_2$O (3X), 10% aq. Na$_2$CO$_3$ (3X), and brine (1X), then dried (MgSO$_4$) and concentrated. The crude residue was purified by crystallization

from the dropwise addition of EtOAc to boiling hexanes, then cooling of the solution, to give crystals. Yield: 40 mg, 0.105 mmol, 54%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.52 (d, J = 4.7 Hz, 2H), 7.29 (d, J = 7.8 Hz, 2H), 7.17 (d, J = 8.1 Hz, 2H), 7.10 (d, J = 4.8 Hz, 2H), 5.50 (bs, 1H), 4.57 (d, J = 13.3 Hz, 1H), 3.86 (d, J = 13.5 Hz, 1H), 3.68 (s, 2H), 3.54 (q, J = 6.6 Hz, 2H), 3.03 − 2.96 (m, 1H), 2.82 (t, J = 6.9 Hz, 2H), 2.70 − 2.62 (m, 1H), 2.21 (tt, J = 11.1, 3.6 Hz, 1H), 1.77 (d, J = 12.3 Hz, 2H), 1.57 (qd, J = 12.2, 4.1 Hz, 1H), 1.46 (qd, J = 12.3, 3.9 Hz, 1H). TOF ES+ MS: 386.2 (M + H), 408.2 (M + Na). HPLC t_R = 6.87 min, > 95% purity.

1-(2-(4-Chlorophenyl)acetyl)-N-((2,3-dihydro-1H-inden-2-yl)methyl)piperidine-4-carboxamide **(CCG-208849).** Ethyl 1-(2-(4-chlorophenyl)acetyl)piperidine-4-carboxylate (811 mg, 2.62 mmol) was added to EtOH (15 mL) and 10% aq. NaOH (5 mL). This mixture was heated to 50 °C and stirred for 5 h, then allowed to cool to RT. Solvent was removed *in vacuo* until precipitation began to occur, at which point the material was cooled to 0 °C and acidified to pH<1 by the dropwise addition of conc. HCl. The solution was decanted off the resulting gum, and the residue was air dried, then dried under high vacuum to afford a highly gummy material that could be solidified by sonication in Et$_2$O to provide the carboxylic acid as a white powder without further purification. Yield: 723 mg, 2.57 mmol, 98%.

The following were added sequentially to DCM (2 mL) at RT: 1-(2-(4-chlorophenyl)acetyl)piperidine-4-carboxylic acid (50 mg, 0.18 mmol), TEA (74 µL, 0.53 mmol), EDC·HCl (37 mg, 0.20 mmol), HOBT (30 mg, 0.20 mmol), and indanyl amine **75** (36 mg, 0.20 mmol). This solution was stirred a RT for 10 h, at which time the DCM was

removed *in vacuo* and the residue was diluted with EtOAc and washed with 0.5M HCl (2X), H_2O (2X), 10% aq. Na_2CO_3 (2X), and brine (1X), then dried ($MgSO_4$) and concentrated. The crude residue was purified by crystallization from boiling EtOAc to give crystals. Yield: 43 mg, 0.11 mmol, 59%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.32 − 7.24 (m, 2H), 7.23 − 7.02 (m, 6H), 5.52 (t, $J = 6.0$ Hz, 1H), 4.63 − 4.53 (m, 1H), 3.95 − 3.82 (m, 1H), 3.69 (s, 2H), 3.37 (td, $J = 6.0$, 1.8 Hz, 2H), 3.13 − 2.97 (m, 3H), 2.70 − 2.57 (m, 4H), 2.24 (tt, $J = 11.2$, 3.9 Hz, 1H), 1.83 − 1.70 (m, 2H), 1.61 − 1.45 (m, 2H). TOF ES+ MS: (M + H) 411.2. HPLC $t_R = 6.87$ min, > 95%.

1-(1-(4-Chlorobenzyl)-1H-imidazole-2-carbonyl)-N-((2,3-dihydro-1H-inden-2-yl)methyl)piperidine-4-carboxamide **(CCG-208913).** Ethyl 1-(4-chlorobenzyl)-1*H*-imidazole-2-carboxylate **23b** (7.28 g, 27.5 mmol), was dissolved in EtOH (10 mL) and 10% aq. NaOH (20 mL) and stirred at rt for 15 h. The solvent was then stripped off, water was added, and the solution acidified with HCl. The resulting precipitate was collected over a filter and washed with 1M HCl and dried to afford the carboxylic acid as a white powder which was taken directly into the next step. Yield: 6.506 g, 27.5 mmol, 100%.

1-(4-chlorobenzyl)-1*H*-imidazole-2-carboxylic acid (50 mg, 0.21 mmol) was added to DCM (5 mL), followed by *N*-((2,3-dihydro-1*H*-inden-2-yl)methyl)piperidine-4-carboxamide (69 mg, 0.23 mmol), TEA (88 μL, 0.63 mmol), EDC·HCl (45 mg, 0.23 mmol), and HOBT (36 mg, 0.23 mmol). The reaction was allowed to stir at RT for 20 h, at which time the reaction was concentrated and the residue taken up in EtOAc. This solution was then washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), dried

(MgSO$_4$), and concentrated. The residue was purified by silica flash chromatography (10 g silica, 60 to 100% EtOAc:Hex) to provide the final compound as a white powder. Yield: 48 mg, 0.10 mmol, 47%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.31 (d, *J* = 8.3 Hz, 2H), 7.21 – 7.12 (m, 6H), 7.07 (s, 1H), 6.96 (s, 1H), 5.67 – 5.61 (m, 1H), 5.38 (d, *J* = 5.6 Hz, 2H), 4.62 (t, *J* = 14.0 Hz, 2H), 3.37 (t, *J* = 5.8 Hz, 2H), 3.15 (t, *J* = 12.1 Hz, 1H), 3.07 (q, *J* = 9.6, 8.8 Hz, 2H), 2.87 – 2.79 (m, 1H), 2.71 – 2.65 (m, 3H), 2.34 (tt, *J* = 11.4, 4.0 Hz, 1H), 1.89 (d, *J* = 13.4 Hz, 1H), 1.82 – 1.75 (m, 1H), 1.61 (dtd, *J* = 36.5, 12.2, 3.5 Hz, 2H). TOF ES+ MS: (M + H) 477.2, (M + Na) 499.2. HPLC *t*$_R$ = 6.07 min, > 95%.

1-(1-(4-Chlorobenzyl)-1H-pyrrole-2-carbonyl)-N-((2,3-dihydro-1H-inden-2-yl)methyl)piperidine-4-carboxamide **(CCG-208914).** 1-(1-(4-chlorobenzyl)-1*H*-pyrrole-2-carbonyl)piperidine-4-carboxylic acid **25a** (50 mg, 0.14 mmol) was added to DCM (5 mL), followed by indanyl amine **75** (29 mg, 0.16 mmol), TEA (60 μL, 0.43 mmol), EDC·HCl (30 mg, 0.16 mmol), and HOBT (24 mg, 0.16 mmol). The reaction was allowed to stir at RT for 20 h, at which time the reaction was concentrated and the residue taken up in EtOAc. This solution was then washed with 1M HCl (3X), H$_2$O (3X) and 10% aq. Na$_2$CO$_3$ (3X), and brine (1X), dried (MgSO$_4$), and concentrated. The residue was purified by silica flash chromatography (10 g silica, 60% EtOAc:Hex) to provide the final compound as a white powder. Yield: 42 mg, 0.09 mmol, 61%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.29 – 7.25 (m, 2H), 7.23 – 7.17 (m, 2H), 7.17 – 7.13 (m, 2H), 7.05 (d, *J* = 8.4 Hz, 2H), 6.81 – 6.80 (m, 1H), 6.34 (dd, *J* = 3.7, 1.6 Hz, 1H), 6.14 (d, *J* = 6.4 Hz, 1H), 5.52 (t, *J* = 5.0 Hz, 1H), 5.29 (s, 2H), 4.36 (d, *J* = 11.7 Hz, 2H), 3.38 (t, *J* = 6.0 Hz, 2H), 3.08 (q, *J* = 9.4, 8.9 Hz, 2H), 2.84 (t, *J* = 11.9 Hz, 2H), 2.73 – 2.65 (m, 3H), 2.25 (tq,

$J = 11.2$, 3.8 Hz, 1H), 1.77 − 1.68 (m, 2H), 1.43 (d, $J = 11.7$ Hz, 2H). TOF ES+ MS: (M + H) 476.2, (M + Na) 498.2. HPLC t_R = 7.60 min, > 95%.

1-(1-(4-Chlorobenzyl)-1H-pyrrole-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)azetidine-3-carboxamide **(CCG-208915)**. Methyl 1-(1-(4-chlorobenzyl)-1H-pyrrole-2-carbonyl)azetidine-3-carboxylate **29** (300 mg, 0.90 mmol) was dissolved in 200 proof EtOH (10 mL) and 10% aq. NaOH (10 mL) was added. The reaction was allowed to stir at RT for 24 h, at which time the solvent was stripped off *in vacuo* and the residue was taken up in H_2O and acidified to pH 1 by addition of conc. HCl at 0°C. The resulting precipitate was collected over a filter and dried by aspirator then high vacuum to afford the desired carboxylic acid as a white powder. Yield: 250 mg, 0.78 mmol, 87%.

1-(1-(4-chlorobenzyl)-1H-pyrrole-2-carbonyl)azetidine-3-carboxylic acid (50 mg, 0.16 mmol) was added to DCM (5 mL), followed by 4-(2-aminoethyl)pyridine (20 μL, 0.17 mmol), TEA (66 μL, 0.47 mmol), EDC·HCl (33 mg, 0.17 mmol), and HOBT (26 mg, 0.17 mmol). The reaction was allowed to stir at RT for 17 h, at which time the reaction was concentrated and the residue taken up in EtOAc. This solution was then washed with H_2O (3X) and 10% aq. Na_2CO_3 (3X), and brine (1X), dried ($MgSO_4$), and concentrated. The residue was purified by silica flash chromatography (4 g silica, 1% to 30% 7 M methanolic ammonia/EtOAc) to provide the final compound as a white powder. Yield: 32 mg, 0.076 mmol, 48%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.48 (s, 2H), 7.25 (d, $J = 8.3$ Hz, 2H), 7.10 (d, $J = 4.1$ Hz, 2H), 7.04 (d, $J = 8.2$ Hz, 2H), 6.80 (s, 1H), 6.50 (d, $J = 3.9$ Hz, 1H), 6.17 (t, $J = 3.2$ Hz, 1H), 5.97 (t, $J = 5.2$ Hz, 1H), 5.52 (s, 2H), 4.25

(bs, 4H), 3.58 (q, $J = 6.6$ Hz, 2H), 3.20 (p, $J = 7.4$ Hz, 1H), 2.84 (t, $J = 6.9$ Hz, 2H). TOF ES+ MS: (M + H) 423.1, (M + Na) 445.1. HPLC rt = 5.09 min, >95% purity.

1-(1-(4-Chlorobenzyl)-1H-imidazole-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-208916).** Ethyl 1-(4-chlorobenzyl)-1*H*-imidazole-2-carboxylate **23b** (7.28 g, 27.5 mmol), was dissolved in EtOH (10 mL) and 10% aq. NaOH (20 mL) and stirred at rt for 15 h. The solvent was then stripped off, water was added, and the solution acidified with HCl. The resulting precipitate was collected over a filter and washed with 1M HCl and dried to afford the carboxylic acid as a white powder which was taken directly into the next step. Yield: 6.506 g, 27.5 mmol, 100%.

The following were added sequentially to DCM (5 mL): 1-(4-chlorobenzyl)-1*H*-imidazole-2-carboxylic acid (73 mg, 0.309 mmol), *N*-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide dihydrochloride (100 mg, 0.371 mmol), DIPEA (162 μL, 0.927 mmol), EDC·HCl (65 mg, 0.340 mmol), and HOBt (52 mg, 0.340 mmol). The solution was allowed to stir at RT for 24 h, after which time the DCM was stripped off in vacuo and the resulting residue was taken up on 10% aq. Na_2CO_3. Material was then extracted with EtOAc. The extract was dried (MgSO$_4$) and concentrated *in vacuo*. The residue was then triturated with EtOAc:Hex and collected over a filter. The impure yellow powder was then recrystallized from EtOAc to provide the title compound as off-white crystals. Yield: 89 mg (64%). ^{1}H NMR (400 MHz, CDCl$_3$) δ 8.50 (d, $J = 5.2$ Hz, 2H), 7.28 (d, $J = 8.3$ Hz, 2H), 7.15–7.07 (m, 4H), 7.05 (s, 1H), 6.95 (s, 1H), 5.59 (t, $J = 5.7$ Hz, 1H), 5.35 (d,$J = 8.9$ Hz, 2H), 4.58 (d, $J = 13.4$ Hz, 2H), 3.53 (q, $J = 6.7$ Hz, 2H), 3.10 (t, $J = 13.7$

Hz, 1H), 2.87–2.71 (m, 3H), 2.29 (tt, *J* = 11.0, 3.4 Hz, 1H), 2.00–1.41 (m, 4H). TOF ES+ MS: (M + H) 452.2, (M + Na) 474.2. HPLC rt = 4.24 min, >90% purity.

1-(1-(4-Chlorobenzyl)-4-fluoro-1H-pyrrole-2-carbonyl)-N-((2,3-dihydro-1H-inden-2-yl)methyl)piperidine-4-carboxamide **(CCG-209020).** The following reagents were dissolved in DCM (3 mL) at RT: 1-(4-chlorobenzyl)-4-fluoro-1*H*-pyrrole-2-carboxylic acid **136** (50 mg, 0.197 mmol), *N*-((2,3-dihydro-1*H*-inden-2-yl)methyl)piperidine-4-carboxamide dihydrochloride (70 mg, 0.237 mmol), TEA (110 µL, 0.788 mmol), EDC·HCl (45 mg, 0.237 mmol), and HOBT (36 mg, 0.237 mmol). The reaction was allowed to stir for 16 h, after which time it was diluted with EtOAc and washed with HCl (3X), H$_2$O (3X), 10% aq. Na$_2$CO$_3$ (3X), and brine (1X), then dried (MgSO$_4$) and concentrated. The residue was purified by silica flash chromatography (10 g silica, 5% to 50% EtOAc:Hex) to give the desired product as a white solid. Yield: ^{1}H NMR (400 MHz, Chloroform-d) δ 7.26 (d, *J* = 8.0 Hz, 2H), 7.21 − 7.09 (m, 4H), 7.05 (d, *J* = 8.2 Hz, 2H), 6.56 − 6.50 (m, 1H), 6.05 (s, 1H), 5.54 (t, *J* = 5.1 Hz, 1H), 5.15 (s, 2H), 4.28 (d, *J* = 13.2 Hz, 2H), 3.36 (t, *J* = 5.7 Hz, 2H), 3.05 (q, *J* = 9.2 Hz, 2H), 2.82 (t, *J* = 11.7 Hz, 2H), 2.71 − 2.61 (m, 3H), 2.22 (tt, *J* = 11.1, 3.7 Hz, 1H), 1.71 (d, *J* = 12.0 Hz, 2H), 1.42 (d, *J* = 11.4 Hz, 2H). TOF ES+ MS: (M + H) 494.2. HPLC rt = 7.75 min, >95% purity.

1-(1-(4-Chlorobenzyl)-4-fluoro-1H-pyrrole-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-209021).** The following were added sequentially to DCM (3 mL): 1-(4-chlorobenzyl)-4-fluoro-1*H*-pyrrole-2-carboxylic

acid **139** (50 mg, 0.20 mmol), TEA (0.11 mL, 0.79 mmol), EDC·HCl (45 mg, 0.24 mmol), HOBT (36 mg, 0.24 mmol), and then amine dihydrochloride **36** (64 mg, 0.24 mmol). The reaction was allowed to stir at RT for 14 h, at which time it was diluted with EtOAc, washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), dried with $MgSO_4$, and concentrated *in vacuo*. The residue was the purified by silica flash chromatography (10 g silica, 1% 7 M methanolic ammonia/EtOAc) to provide the desired material as a white powder. Yield: 48 mg (52%). ^{1}H NMR (400 MHz, CDCl3) δ 8.54 (d, J = 4.3 Hz, 2H), 7.25–7.20 (m, 2H), 7.14 (d, J = 4.4 Hz, 2H), 7.06 (d, J = 6.9 Hz, 2H), 6.58–6.54 (m, 1H), 6.05 (s, 1H), 5.46–5.38 (m, 1H), 5.17 (s, 2H), 4.29 (d, J = 6.7 Hz, 2H), 3.55 (q, J = 7.3 Hz, 2H), 2.83 (dt, J = 17.9, 9.0 Hz, 4H), 2.27–2.15 (m, 1H), 1.77–1.63 (m, 2H), 1.38 (d, J = 13.4 Hz, 2H). TOF ES+ MS: (M + H) 469.2, (M + Na) 491.2. HPLC rt = 5.18 min, >95% purity.

1-(1-(4-Chlorobenzyl)-1H-pyrrolo[2,3-c]pyridine-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-211751).** Azaindole ethyl ester **144** (300 mg, 0.953 mmol) was dissolved into ethanol (8 mL) at RT, followed by the addition of 10% aqueous NaOH (8 mL), which initially caused precipitation, but homogeneity was achieved over time. The reaction was allowed to stir at RT for 12 h, at which time the solvent was removed in vacuo. The residue was taken up in a small amount of water (5 mL), the pH was adjusted to ~4, and material was extracted with ethyl acetate (8×). The extracts were combined and the solvent removed again in vacuo to provide the title compound as a fine white powder. Yield: 226 mg (83%). ^{1}H NMR (400 MHz, DMSO-

d6) δ 9.64 (s, 1H), 8.40 (d, J = 6.3 Hz, 1H), 8.26 (d, J = 6.3 Hz, 1H), 7.61 (s, 1H), 7.35 (d, J = 8.5 Hz, 2H), 7.10 (d, J = 8.5 Hz, 2H), 6.06 (s, 2H).

The azaindole carboxylic acid (50 mg, 0.174 mmol) was dissolved into DCM (2 mL) at RT, followed by amine dihydrochloride **36** (59 mg, 0.192 mmol) and TEA (122 μL, 0.872 mmol). Once all material was in solution, EDC·HCl (37 mg, 0.192 mmol) and HOBT (29 mg, 0.192 mmol) were added and the reaction was allowed to stir for 18 h at RT. At this time, the reaction was diluted with a solution of ethyl acetate/diethyl ether (1:1) and washed with water (3×), 10% aq sodium carbonate (3×), and brine (1×), then dried (magnesium sulfate) and concentrated in vacuo. The resulting residue was then purified by flash chromatography (50 g silica, 5% 7.0 M methanolic ammonia/95% ethyl acetate) to give the title compound as white solid. Yield: 53 mg (61%). ^{1}H NMR (400 MHz, CDCl3) δ 8.78 (s, 1H), 8.49 (d, J = 5.4 Hz, 2H), 8.27 (d, J = 5.5 Hz, 1H), 7.51 (d, J = 5.5 Hz, 1H), 7.21 (d, J = 8.2 Hz, 2H), 7.09 (d, J = 5.3 Hz, 2H), 7.05 (d, J = 8.2 Hz, 2H), 6.56 (s, 1H), 5.67 (t, J = 5.6 Hz, 1H), 5.48 (s, 2H), 4.57 (s, 1H), 3.91 (s, 1H), 3.53 (q, J = 6.6 Hz, 2H), 2.96−2.69 (m, 4H), 2.21 (tt, J =11.1, 7.5, 3.7 Hz, 1H), 1.88−1.69 (m, 1H), 1.67−1.46 (m, 2H), 1.33−1.22 (m, 1H). TOF ES+ MS: (M + H) 502.2. HPLC t_R = 4.01 min, > 95%.

(R)-1-(1-(4-Chlorobenzoyl)pyrrolidine-2-carbonyl)-N-((2,3-dihydro-1H-inden-2-yl)methyl)piperidine-4-carboxamide **(CCG-211753).** (R)-Ethyl 1-(1-(4-chlorobenzoyl)pyrrolidine-2-carbonyl)piperidine-4-carboxylate **(R)-40** (300 mg, 0.76 mmol) was dissolved in a 9:1 DCM:MeOH solution (20 mL), and a solution of methanolic NaOH (92 mg NaOH/2mL MeOH) was added. This was stirred at RT for 2 h,

after which time the solvent was stripped off *in vacuo* to give a white solid that was dissolved in a small amount of H_2O and acidified by the addition of conc. HCl, and the material was extracted with EtOAc (3X). The combined organic extracts were dried (MgSO$_4$) and concentrated. The clear oily residue was then dissolved in minimum EtOAc and precipitated with Hex to afford a white powder. No further purification necessary. Yield: 260 mg, 0.71 mmol, 93%.

(R)-1-(4-chlorobenzoyl)pyrrolidine-2-carboxylic acid (50 mg, 0.14 mmol) was added to DCM (5 mL), followed by the sequential addition of TEA (76 µL, 0.55 mmol), HATU (63 mg, 0.16 mmol), and indanyl amine **75** (24 mg, 0.16 mmol). The reaction was stirred for 22 h at RT, after which time the reaction was diluted in sat. aq. NH$_4$Cl and extracted with EtOAc. The organic extract was then washed with 1M HCl (3X), H$_2$O (3X), 10% aq. Na$_2$CO$_3$ (3X), and sat. aq. Na$_2$CO$_3$ (1X), then dried with MgSO$_4$ and concentrated. The residue was purified via silica flash chromatography (4 g silica, 5% to 20% 7 M methanolic ammonia:EtOAc) to give the desired compound as a white powder. Yield: 37 mg, 0.08 mmol, 57%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.52 (d, *J* = 8.1 Hz, 2H), 7.35 (d, *J* = 8.0 Hz, 2H), 7.18 − 7.11 (m, 4H), 6.04 (s, 0.5H), 5.75 − 5.67 (m, 0.5H), 5.03 (m, 1H), 4.60 (d, *J* = 13.3 Hz, 0.5H), 4.47 (d, *J* = 13.6 Hz, 0.5H), 4.07 (dd, *J* = 30.7, 13.8 Hz, 1H), 3.80 (m, 0.5H), 3.68 (m, 1.5H), 3.51 (m, 1H), 3.32 (m, 2H), 3.04 (m, 2H), 2.85 − 2.74 (m, 1H), 2.72 − 2.56 (m, 3H), 2.43 (m, 0.5H), 2.36 − 2.14 (m, 1.5H), 2.14 − 2.00 (m, 2H), 2.02 − 1.76 (m, 5H), 1.65 (dd, *J* = 29.2, 12.8 Hz, 2H). TOF ES+ MS: (M + H) 494.2. HPLC t_R = 6.37 min, > 95% purity.

(S)-1-(1-(4-Chlorobenzoyl)pyrrolidine-2-carbonyl)-N-(2-(pyridin-4-

yl)ethyl)piperidine-4-carboxamide **(CCG-211751)**. (S)-Ethyl 1-(1-(4-

chlorobenzoyl)pyrrolidine-2-carbonyl)piperidine-4-carboxylate **(S)-40** (300 mg, 0.76

mmol) was dissolved in a 9:1 DCM:MeOH solution (20 mL), and a solution of

methanolic NaOH (92 mg NaOH/2mL MeOH) was added. This was stirred at RT for 2 h,

after which time the solvent was stripped off *in vacuo* to give a white solid that was

dissolved in a small amount of H_2O and acidified by the addition of conc. HCl, and the

material was extracted with EtOAc (3X). The combined organic extracts were dried

($MgSO_4$) and concentrated. The clear oily residue was then dissolved in minimum EtOAc

ad precipitated with Hex to afford a white powder. No further purification necessary.

Yield: 265 mg, 0.73 mmol, 95%.

(S)-1-(4-chlorobenzoyl)pyrrolidine-2-carboxylic acid (50 mg, 0.14 mmol) was

added to DCM (5 mL), followed by the sequential addition of TEA (76 μL, 0.55 mmol),

HATU (63 mg, 0.16 mmol), and amine **27** (20 μL, 0.16 mmol). The reaction was stirred

for 17 h at RT, after which time the reaction was diluted in sat. aq. NH_4Cl and extracted

with EtOAc. The organic extract was then washed with 10% aq. NH_4Cl (2X), 10% aq.

Na_2CO_3 (3X), and sat. aq. Na_2CO_3 (1X), then dried with $MgSO_4$ and concentrated. The

residue was purified via silica flash chromatography (4 g silica, 5% to 20% 7 M

methanolic ammonia:EtOAc) to give the desired compound as a white powder. Yield: 42

mg, 0.09 mmol, 65%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.52 (m, 2H), 7.53 (d, *J* =

8.0 Hz, 2H), 7.37 (t, *J* = 7.0 Hz, 2H), 7.16 − 7.09 (m, 2H), 5.91 (d, *J* = 2.1 Hz, 0.5H),

5.58 − 5.49 (m, 0.5H), 5.07 − 4.97 (m, 1H), 4.63 (d, *J* = 14.5 Hz, 0.5H), 4.48 (d, *J* = 13.9

Hz, 1H), 4.09 (dd, *J* = 41.8, 14.7 Hz, 2H), 3.68 (t, *J* = 9.9 Hz, 1H), 3.54 (m, 3H), 3.32 −

3.21 (m, 0.5H), 3.12 (t, J = 12.8 Hz, 0.5H), 2.83 (dt, J = 14.6, 6.8 Hz, 3H), 2.64 (t, J = 12.6 Hz, 0.5H), 2.47 –1.15 (m, 7H). TOF ES+ MS: (M + H) 469.3, (M + Na) 491.2. HPLC t_R = 4.36 min, > 95% purity.

(S)-1-(1-(4-Chlorobenzoyl)pyrrolidine-2-carbonyl)-N-((2,3-dihydro-1H-inden-2-yl)methyl)piperidine-4-carboxamide **(CCG-211755).** (S)-Ethyl 1-(1-(4-chlorobenzoyl)pyrrolidine-2-carbonyl)piperidine-4-carboxylate **(S)-40** (300 mg, 0.76 mmol) was dissolved in a 9:1 DCM:MeOH solution (20 mL), and a solution of methanolic NaOH (92 mg NaOH/2mL MeOH) was added. This was stirred at RT for 2 h, after which time the solvent was stripped off *in vacuo* to give a white solid that was dissolved in a small amount of H_2O and acidified by the addition of conc. HCl, and the material was extracted with EtOAc (3X). The combined organic extracts were dried ($MgSO_4$) and concentrated. The clear oily residue was then dissolved in minimum EtOAc ad precipitated with Hex to afford a white powder. No further purification necessary. Yield: 265 mg, 0.73 mmol, 95%.

(S)-1-(4-chlorobenzoyl)pyrrolidine-2-carboxylic acid (50 mg, 0.14 mmol) was added to DCM (5 mL), followed by the sequential addition of TEA (76 µL, 0.55 mmol), HATU (63 mg, 0.16 mmol), and indanyl amine **75** (24 mg, 0.16 mmol). The reaction was stirred for 19 h at RT, after which time the reaction was diluted in sat. aq. NH_4Cl and extracted with EtOAc. The organic extract was then washed with 1M HCl (3X), H_2O (3X), 10% aq. Na_2CO_3 (3X), and sat. aq. Na_2CO_3 (1X), then dried with $MgSO_4$ and concentrated. The residue was purified via silica flash chromatography (4 g silica, 5% to 20% 7 M methanolic ammonia:EtOAc) to give the desired compound as a white powder.

Yield: 44 mg, 0.09 mmol, 68%. ^{1}H NMR (400 MHz, Chloroform-d) δ 7.61 − 7.54 (m, 1H), 7.54 − 7.49 (m, 1H), 7.44 − 7.37 (m, 1H), 7.37 − 7.28 (m, 2H), 7.25 − 7.06 (m, 4H), 6.14 − 6.02 (m, 0.5H), 5.81 − 5.70 (m, 0.5H), 5.19 − 4.96 (m, 1H), 4.60 (d, J = 13.6 Hz, 0.5H), 4.47 (d, J = 13.7 Hz, 0.5H), 4.06 (dd, J = 30.8, 14.3 Hz, 1H), 3.73 − 3.61 (m, 1H), 3.59 − 3.43 (m, 1H), 3.42 − 3.21 (m, 2H), 3.19 − 2.94 (m, 3H), 2.76 − 2.53 (m, 3H), 2.35 − 2.14 (m, 2H), 2.11 − 1.77 (m, 5H), 1.77 − 1.51 (m, 2H). TOF ES+ MS: (M + H) 494.2. HPLC t_R = 6.37 min, > 95% purity.

(R)-1-(1-(4-Chlorobenzoyl)pyrrolidine-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-211756)**. (R)-Ethyl 1-(1-(4-chlorobenzoyl)pyrrolidine-2-carbonyl)piperidine-4-carboxylate **(R)-40** (300 mg, 0.76 mmol) was dissolved in a 9:1 DCM:MeOH solution (20 mL), and a solution of methanolic NaOH (92 mg NaOH/2mL MeOH) was added. This was stirred at RT for 2 h, after which time the solvent was stripped off *in vacuo* to give a white solid that was dissolved in a small amount of H_2O and acidified by the addition of conc. HCl, and the material was extracted with EtOAc (3X). The combined organic extracts were dried (MgSO$_4$) and concentrated. The clear oily residue was then dissolved in minimum EtOAc and precipitated with Hex to afford a white powder. No further purification necessary. Yield: 260 mg, 0.71 mmol, 93%.

(R)-1-(4-chlorobenzoyl)pyrrolidine-2-carboxylic acid (50 mg, 0.14 mmol) was added to DCM (5 mL), followed by the sequential addition of TEA (76 μL, 0.55 mmol), HATU (63 mg, 0.16 mmol), and amine **27** (20 μL, 0.16 mmol). The reaction was stirred for 18 h at RT, after which time the reaction was diluted in sat. aq. NH$_4$Cl and extracted

with EtOAc. The organic extract was then washed with 10% aq. NH$_4$Cl (2X), 10% aq. Na$_2$CO$_3$ (3X), and sat. aq. Na$_2$CO$_3$ (1X), then dried with MgSO$_4$ and concentrated. The residue was purified via silica flash chromatography (4 g silica, 5% to 20% 7 M methanolic ammonia:EtOAc) to give the desired compound as a white powder. Yield: 41 mg, 0.09 mmol, 62%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.59 − 8.47 (m, 2H), 7.53 (d, *J* = 8.1 Hz, 2H), 7.38 (t, *J* = 7.6 Hz, 2H), 7.12 (dd, *J* = 9.5, 4.8 Hz, 2H), 5.90 (s, 0.5H), 5.50 (s, 0.5H), 5.10 − 4.99 (m, 1H), 4.63 (d, *J* = 13.3 Hz, 0.5H), 4.48 (d, *J* = 13.2 Hz, 1H), 4.09 (dd, *J* = 40.4, 14.2 Hz, 2H), 3.70 (t, *J* = 8.6 Hz, 1H), 3.54 (m, 3H), 3.29 (t, *J* = 12.8 Hz, 0.5H), 3.12 (t, *J* = 12.7 Hz, 0.5H), 2.83 (dt, *J* = 14.7, 6.6 Hz, 3H), 2.64 (t, *J* = 12.6 Hz, 0.5H), 2.40-1.20 (m, 7H). TOF ES+ MS: (M + H) 469.3, (M + Na) 491.3. HPLC *t*$_R$ = 4.26 min, > 95% purity.

(S)-1-(1-(4-Chlorobenzyl)pyrrolidine-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-211757)**. (S)-1-(4-chlorobenzyl)pyrrolidine-2-carboxylic acid **(S)-38** (63 mg, 0.27 mmol) was added to DCM (2 mL), followed by the sequential addition of TEA (150 µL, 1.06 mmol), HATU (111 mg, 0.29 mmol), and amine dihydrochloride **36** (75 mg, 0.28 mmol). The reaction was stirred for 24 h at RT, after which time the reaction was diluted in sat. aq. NH$_4$Cl and extracted with EtOAc. The organic extract was then washed with 10% aq. NH$_4$Cl (2X), 10% aq. Na$_2$CO$_3$ (3X), and sat. aq. Na$_2$CO$_3$ (1X), then dried with MgSO$_4$ and concentrated. The residue was purified via silica flash chromatography (4 g silica, 5% to 20% 7 M methanolic ammonia/EtOAc) to give a slightly yellow oil that was triturated from Et$_2$O/Hex to provide a white powder. Yield: 64 mg, 0.14 mmol, 53%. ^{1}H NMR (500 MHz,

Chloroform-d) δ 8.48 (d, *J* = 4.6 Hz, 2H), 7.31 − 7.22 (m, 4H), 7.11 (bs, 2H), 5.82 (bs, 1H), 4.52 (bs, 1H), 4.13 (d, *J* = 11.9 Hz, 1H), 3.89 (dd, *J* = 18.3, 13.1 Hz, 1H), 3.58 − 3.49 (m, 2H), 3.46 − 3.34 (m, 2H), 3.10 − 3.00 (m, 1H), 2.94 (d, *J* = 9.3 Hz, 1H), 2.87 − 2.79 (m, 2H), 2.63 − 2.52 (m, 1H), 2.33 − 2.26 (m, 2H), 2.18 − 2.08 (m, 1H), 1.89 − 1.75 (m, 4H), 1.60 − 1.50 (m, 2H). TOF ES+ MS: (M + H) 455.2, (M + Na) 477.2. HPLC t_R = 3.39 min, > 90% purity.

(R)-1-(1-(4-Chlorobenzyl)pyrrolidine-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-211758)**. (R)-1-(4-chlorobenzyl)pyrrolidine-2-carboxylic acid **(R)-38** (64 mg, 0.27 mmol) was added to DCM (2 mL), followed by the sequential addition of TEA (150 µL, 1.06 mmol), HATU (111 mg, 0.29 mmol), and amine dihydrochloride **36** (75 mg, 0.28 mmol). The reaction was stirred for 20 h at RT, after which time the reaction was diluted in sat. aq. NH$_4$Cl and extracted with EtOAc. The organic extract was then washed with 10% aq. NH$_4$Cl (2X), 10% aq. Na$_2$CO$_3$ (3X), and sat. aq. Na$_2$CO$_3$ (1X), then dried with MgSO$_4$ and concentrated. The residue was purified via silica flash chromatography (4 g silica, 5% to 20% 7 M methanolic ammonia:EtOAc) to give a slightly yellow oil that was triturated from Et$_2$O/Hex to provide a white powder. Yield: 75 mg, 0.17 mmol, 62%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.52 (d, *J* = 5.1 Hz, 2H), 7.26 (s, 4H), 7.12 (s, 2H), 5.57 (s, 1H), 4.55 (d, *J* = 11.7 Hz, 1H), 4.15 (bs, 1H), 3.90 (dd, *J* = 18.8, 13.1 Hz, 1H), 3.61 − 3.50 (m, 2H), 3.39 (d, *J* = 12.0 Hz, 2H), 3.06 (d, *J* = 7.0 Hz, 1H), 2.98 − 2.91 (m, 1H), 2.84 (q, *J* = 6.2 Hz, 2H), 2.59 (q, *J* = 13.9, 13.3 Hz, 1H), 2.33 − 2.21 (m, 2H), 2.14 (s, 1H), 1.80 − 1.77

(m, 2H), 1.59 – 1.48 (m, 2H). TOF ES+ MS: (M + H) 455.2, (M + Na) 477.2. HPLC t_R = 3.44 min, > 90% purity.

(E)-(1-(4-Chlorobenzyl)-1H-indol-2-yl)(4-(4-(pyridin-4-yl)but-1-en-1-yl)piperidin-1-yl)methanone **(CCG-211822).** (E)-tert-butyl 4-(4-(pyridin-4-yl)but-1-en-1-yl)piperidine-1-carboxylate **99b** (100 mg, 0.316 mmol) was dissolved in anhydrous 1,4-dioxane (1 mL) and 4 M HCl in 1,4-dioxane (0.55 mL, 2.21 mmol) was added. The reaction was allowed to stir for 1 h, at which time the solvent was stripped off and replaced with Et$_2$O. The oily residue was solidified with sonication, washed (decantation) with Et$_2$O (3X) and then dried under high vacuum to give the dihydrochloride as a white powder. Yield: 91 mg, 0.316 mmol, 100%.

(E)-4-(4-(pyridin-4-yl)but-1-en-1-yl)piperidine dihydrochloride (75 mg, 0.259 mmol) was dissolved in DCM (10 mL) and TEA (217 µL, 1.56 mmol), followed by the indole carboxylic acid **4a** (89 mg, 0.311 mmol), EDC·HCl (60 mg, 0.311 mmol), and HOBT (48 mg, 0.311 mmol), and the reaction was allowed to stir at RT for 18 h. After this time, the reaction was diluted with EtOAc and washed with H$_2$O (3X), 10% Na$_2$CO$_3$ (3X), and brine (1X), then dried (Na$_2$SO$_4$) and concentrated *in vacuo*. The resulting residue was purified via silica flash chromatography (10 g silica, 1% to 20% methanolic ammonia:EtOAc) to provide a clear, colorless oil. Yield: 72 mg, 0.148 mmol, 57%. ^{1}H NMR (400 MHz, Chloroform-d) δ 8.54 – 8.39 (m, 2H), 7.63 (d, *J* = 7.8 Hz, 1H), 7.35 (d, *J* = 8.2 Hz, 1H), 7.25 (t, *J* = 7.6 Hz, 1H), 7.21 – 7.10 (m, 3H), 7.08 (s, 2H), 7.01 (d, *J* = 8.0 Hz, 2H), 6.59 (s, 1H), 5.46 (s, 2H), 5.37 – 5.31 (m, 1H), 5.26 (d, *J* = 11 Hz, 1H), 4.57 (bs, 1H), 4.04 (bs, 1H), 2.85 (bs, 2H), 2.65 (t, *J* = 7.3 Hz, 2H), 2.39 – 2.23 (m, 2H), 2.07

(s, 1H), 1.64 (bs, 1H), 1.41 (bs, 1H), 1.08 (bs, 1H), 0.64 (bs, 1H). TOF ES+ MS: (M + H) 484.2. HPLC t_R = 6.24 min, > 95% purity.

1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)-1,2,3,6-tetrahydropyridine-4-carboxamide **(CCG-211823).** Ethyl 1-(1-(4-chlorobenzyl)-1*H*-indole-2-carbonyl)-1,2,3,6-tetrahydropyridine-4-carboxylate (53 mg, 0.125 mmol) was dissolved in EtOH (5 mL), followed by the addition of 10% aq. NaOH (2 mL). This reaction was allowed to stir for 4 h, at which time the solvent was removed *in vacuo*. The residue was dissolved in H_2O and acidified with conc. HCl at 0 °C. The resulting precipitate was collected over a filter and dried under high vacuum to give a white powder. Yield: 46 mg, 0.116 mmol, 93%.

1-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)-1,2,3,6-tetrahydropyridine-4-carboxylic acid (33 mg, 0.084 mmol) was dissolved in DCM (5 mL) and TEA (35 μL, 0.251 mmol), followed by EDC·HCl (18 mg, 0.092 mmol), and HOBT (14 mg, 0.092 mmol), and the reaction was allowed to stir at RT for 21 h. After this time, the reaction was diluted with EtOAc:Et$_2$O and washed with H_2O (2X), 10% Na_2CO_3 (3X), and brine (1X), then dried (Na_2SO_4) and concentrated *in vacuo*. The resulting residue was purified via silica flash chromatography (4 g silica, 80% to 100% EtOAc:Hex) to provide a fine white powder. Yield: 38 mg, 0.076 mmol, 91%. ^{1}H NMR (400 MHz, Chloroform-d) δ 8.51 (d, *J* = 4.1 Hz, 2H), 7.66 (d, *J* = 7.9 Hz, 1H), 7.30 − 7.05 (m, 8H), 6.97 (d, *J* = 8.1 Hz, 2H), 6.78 (s, 1H), 5.77 (s, 1H), 5.51 (s, 2H), 4.83 (bs, 1H), 3.89 (bs, 1H), 3.61 − 3.43 (m, 3H), 2.99 (s, 1H), 2.84 (t, *J* = 6.9 Hz, 2H), 2.16 (bs,

1H), 1.94 (bs, 1H). TOF ES+ MS: (M + H) 499.2, (M + Na) 521.2. HPLC t_R = 5.46 min, > 95%.

1-(2-((4-Chlorophenyl)amino)benzoyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-211824)**. Ethyl 1-(2-((4-chlorophenyl)amino)benzoyl)piperidine-4-carboxylate (72 mg, 0.19 mmol) was dissolved in EtOH (5 mL), followed by the addition of 10% aq. NaOH (2 mL). This reaction was allowed to stir at RT for 24 h, after which time the solvent was stripped off *in vacuo* and the residue was taken up in H_2O and acidified to pH<1 with conc. HCl at 0 °C. The resulting white precipitate was collected over a filter, dried via aspirator then high vacuum to afford the carboxylic acid as a white powder requiring no further purification. Yield: 47 mg, 0.13 mmol, 70%.

1-(2-((4-chlorophenyl)amino)benzoyl)piperidine-4-carboxylic acid (36 mg, 0.10 mmol), 4-(2-aminoethyl)pyridine (13 µL, 0.11 mmol), TEA (42 µL, 0.30 mmol), EDC·HCl (21 mg, 0.11 mmol), and HOBT (17 mg, 0.11 mmol) were all added to DCM (5 mL) in the listed order and stirred at RT for 20 h, at which time the reaction was diluted with 1:1 EtOAc:Et$_2$O and washed with 0.5 M aq. HCl (1X), H_2O (1X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic phase was then dried (MgSO$_4$) and concentrated, and the residue was purified via silica flash chromatography (4 g silica, 80% to 100% EtOAc:Hex gradient) to give the title compound as a clear, colorless oil. Yield: 36 mg, 0.078 mmol, 78%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.51 (d, *J* = 4.9 Hz, 2H), 7.32 (d, *J* = 8.3 Hz, 1H), 7.26 – 7.19 (m, 3H), 7.17 (d, *J* = 7.6 Hz, 1H), 7.13 – 7.09 (m, 2H), 7.00 (d, *J* = 8.6 Hz, 2H), 6.89 (t, *J* = 7.4 Hz, 1H), 5.56 (s, 1H), 4.23 (bs, 2H), 3.53 (q, *J* = 6.6 Hz, 2H), 2.93 (s, 2H), 2.82 (t, *J* = 6.9 Hz, 2H), 2.28 (tt, *J* = 11.2, 3.7

Hz, 1H), 1.80 – 1.66 (m, 4H). TOF ES+ MS: (M + H) 463.2, (M + Na) 485.2. HPLC t_R = 5.23 min, > 95%.

1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-4-methyl-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-211825).** 1-(1-(4-chlorobenzyl)-1*H*-indole-2-carbonyl)-4-methylpiperidine-4-carboxylic acid (100 mg, 0.243 mmol), 4-(2-aminoethyl)pyridine (32 μL, 0.268 mmol), TEA (100 μL, 0.730 mmol), EDC·HCl (51 mg, 0.268 mmol), and HOBT (41 mg, 0.268 mmol) were dissolved in DCM (5 mL) and stirred at RT for 16 h. After this time, the reaction was diluted with a 2:1 solution of EtOAc:Et$_2$O and washed with H$_2$O (3X), 10% aq. Na$_2$CO$_3$ (3X), and brine (1X), then dried (MgSO$_4$) and concentrated. The residue was purified by silica flash chromatography (4 g silica, 1% to 5% methanolic ammonia:EtOAc) to give a clear colorless oil. Yield: 113 mg, 0.219 mmol, 90%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.46 (d, *J* = 4.7 Hz, 2H), 7.63 (d, *J* = 8.0 Hz, 1H), 7.35 (d, *J* = 8.3 Hz, 1H), 7.26 (t, *J* = 7.7 Hz, 1H), 7.19 – 7.12 (m, 3H), 7.08 (d, *J* = 4.8 Hz, 2H), 7.00 (d, *J* = 8.0 Hz, 2H), 6.58 (s, 1H), 5.95 (t, *J* = 5.6 Hz, 1H), 5.44 (s, 2H), 4.02 (s, 1H), 3.68 (s, 1H), 3.52 (q, *J* = 6.5 Hz, 2H), 3.22 (s, 2H), 2.81 (t, *J* = 6.9 Hz, 2H), 2.22 (s, 1H), 1.86 (s, 1H), 1.67 (s, 1H), 1.28 (s, 1H), 1.05 (s, 3H), 0.77 (s, 1H). TOF ES+ MS: (M + H) 515.0. HPLC t_R = 5.49 min, > 95%.

(1-(4-Chlorobenzyl)-1H-indol-2-yl)(4-(3-(pyridin-4-yl)propyl)piperazin-1-yl)methanone **(CCG-211826).** t-Butyl 4-(3-(pyridin-4-yl)propyl)piperazine-1-carboxylate (160 mg, 0.524 mmol) was dissolved in 1,4-dioxane (2 mL) at RT, and a 4 M HCl in 1,4-

dioxane solution (1.31 mL, 5.24 mmol) was added. The reaction was allowed to stir for 15 min, over which time precipitation occurred. The solvent was removed *in vacuo* and dried under high vacuum to give the trishydrochloride salt as a tan powder. Yield: 168 mg, 0.524 mmol, 100%.

1-(4-Chlorobenzyl)-1*H*-indole-2-carboxylic acid (91 mg, 0.318 mmol), 4-(3-(pyridin-4-yl)propyl)piperazine trishydrochloride (100 mg, 0.318 mmol), TEA (266 µL, 1.907 mmol), EDC·HCl (67 mg, 0.350 mmol), and HOBT (54 mg, 0.350 mmol) were dissolved in DCM (5 mL) and stirred at RT for 14 h. After this time, the reaction was diluted with a 2:1 solution of EtOAc:Et$_2$O and washed with H$_2$O (3X), 10% aq. Na$_2$CO$_3$ (3X), and brine (1X), then dried (MgSO$_4$) and concentrated. The residue was purified by silica flash chromatography (4 g silica, 1% to 5% MeOH:EtOAc) to give a clear colorless oil. Yield: 94 mg, 0.199 mmol, 63%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.49 (d, $J =$ 5.2 Hz, 2H), 7.65 (d, $J =$ 7.9 Hz, 1H), 7.38 (d, $J =$ 8.3 Hz, 1H), 7.31 − 7.26 (m, 1H), 7.19 (dd, $J =$ 13.2, 7.9 Hz, 3H), 7.11 (d, $J =$ 5.2 Hz, 2H), 7.03 (d, $J =$ 8.2 Hz, 2H), 6.62 (s, 1H), 5.49 (s, 2H), 3.64 (bs, 4H), 2.64 (t, $J =$ 7.6 Hz, 2H), 2.36 − 2.26 (m, 4H), 2.00 (bs, 2H), 1.79 (p, $J =$ 7.5 Hz, 2H). TOF ES+ MS: (M + H) 473.2, (M + Na) 495.2. HPLC $t_R =$ 4.78 min, > 95%.

(1-(4-Chlorobenzyl)-1H-indol-2-yl)(4-phenethylpiperazin-1-yl)methanone **(CCG-211827).** 1-(4-Chlorobenzyl)-1*H*-indole-2-carboxylic acid (100 mg, 0.350 mmol), *N*-(2-phenylethyl)piperazine (73 µL, 0.385 mmol), TEA (146 µL, 1.050 mmol), EDC·HCl (74 mg, 0.385 mmol), and HOBT (59 mg, 0.385 mmol) were dissolved in DCM (6 mL) and stirred at RT for 14 h. After this time, the reaction was diluted with a 2:1 solution of

EtOAc:Et$_2$O and washed with H$_2$O (3X), 10% aq. Na$_2$CO$_3$ (3X), and brine (1X), then dried (MgSO$_4$) and concentrated. The residue was purified by silica flash chromatography (4 g silica, 60% EtOAc:Hex) to give a clear colorless oil. Yield: 109 mg, 0.238 mmol, 68%. ^{1}H NMR (500 MHz, Chloroform-d) δ 7.67 (d, J = 7.9 Hz, 1H), 7.39 (d, J = 8.4 Hz, 1H), 7.33 − 7.26 (m, 3H), 7.20 (dt, J = 19.0, 7.8 Hz, 6H), 7.05 (d, J = 8.3 Hz, 2H), 6.65 (s, 1H), 5.50 (s, 2H), 3.68 (s, 4H), 2.77 (dd, J = 9.6, 6.5 Hz, 2H), 2.57 (dd, J = 9.7, 6.5 Hz, 2H), 2.28 (d, J = 147.4 Hz, 5H). TOF ES+ MS: (M + H) 458.2. HPLC t_R = 5.76 min, > 95%.

(Z)-(1-(4-Chlorobenzyl)-1H-indol-2-yl)(4-(4-(pyridin-4-yl)but-1-en-1-yl)piperidin-1-yl)methanone (**CCG-211829**). (*Z*)-tert-butyl 4-(4-(pyridin-4-yl)but-1-en-1-yl)piperidine-1-carboxylate **99a** (100 mg, 0.316 mmol) was dissolved in anhydrous 1,4-dioxane (1 mL) and 4 M HCl in 1,4-dioxane (0.55 mL, 2.21 mmol) was added. The reaction was allowed to stir for 1 h, at which time the solvent was stripped off and replaced with Et$_2$O. The oily residue was solidified with sonication, washed (decantation) with Et$_2$O (3X) and then dried under high vacuum to give the dihydrochloride as a white powder. Yield: 90 mg, 0.316 mmol, 100%.

(*Z*)-4-(4-(pyridin-4-yl)but-1-en-1-yl)piperidine dihydrochloride (75 mg, 0.259 mmol) was dissolved in DCM (10 mL) and TEA (217 µL, 1.56 mmol), followed by the indole carboxylic acid **4a** (89 mg, 0.311 mmol), EDC·HCl (60 mg, 0.311 mmol), and HOBT (48 mg, 0.311 mmol), and the reaction was allowed to stir at RT for 16 h. After this time, the reaction was diluted with EtOAc and washed with H$_2$O (3X), 10% Na$_2$CO$_3$ (3X), and brine (1X), then dried (Na$_2$SO$_4$) and concentrated *in vacuo*. The resulting

residue was purified via silica flash chromatography (10 g silica, 1% to 20% methanolic ammonia:EtOAc) to provide a clear, colorless oil. Yield: 54 mg, 0.111 mmol, 43%. ^{1}H NMR (400 MHz, Chloroform-d) δ 8.47 (d, J = 5.5 Hz, 2H), 7.62 (d, J = 7.9 Hz, 1H), 7.35 (d, J = 8.3 Hz, 1H), 7.28 − 7.22 (m, 1H), 7.17 (dd, J = 15.8, 7.9 Hz, 3H), 7.09 (d, J = 5.5 Hz, 2H), 7.01 (d, J = 8.3 Hz, 2H), 6.58 (s, 1H), 5.46 (s, 2H), 5.32 (dt, J = 8.1, 7.4 Hz, 1H), 5.11 (t, J = 8.1 Hz, 1H), 4.54 (bs, 1H), 4.05 (bs, 1H), 2.78 (bs, 2H), 2.64 (t, J = 7.2 Hz, 2H), 2.36 (q, J = 7.4 Hz, 2H), 2.31 − 2.19 (m, 1H), 1.43 (bs, 1H), 1.12 (bs, 2H), 0.64 (bs, 1H). TOF ES+ MS: (M + H) 484.2. HPLC t_R = 6.29 min, > 95%.

N-(1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)piperidin-4-yl)-3-(pyridin-4-yl)propanamide **(CCG-212052).** *t*-Butyl 4-(3-(pyridin-4-yl)propanamido)piperidine-1-carboxylate (250 mg, 0.750 mmol) was dissolved in 1,4-dioxane (1 mL) and 4 M HCl (1.0 mL, 4.12 mmol) in 1,4-dioxane was added. The reaction was allowed to stir for 1 h, then the solvent was removed and replaced with Et$_2$O and the oily material was solidified with sonication. Washing with Et$_2$O and thorough drying under high vacuum afforded the desired dihydrochloride salt. Yield: 230 mg, 0.750 mmol, 100%.

4-(3-(pyridin-4-yl)propanamido)piperidine dihydrochloride (100 mg, 0.327 mmol), 1-(4-chlorobenzyl)-1*H*-indole-2-carboxylic acid **4a** (112 mg, 0.392 mmol), TEA (273 μL, 1.959 mmol), EDC·HCl (75 mg, 0.392 mmol), and HOBT (60 mg, 0.392 mmol) were all dissolved in DCM (10 mL) and stirred at RT for 22 h, after which time the DCM was removed *in vacuo* and the residue was taken up in EtOAc and washed with H$_2$O (2X), 10% Na$_2$CO$_3$ (3X), and brine (1X), then dried (MgSO$_4$) and concentrated. The resulting residue was purified via silica flash chromatography (10 g silica, 1% to 20%

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methanolic ammonia:EtOAc) to provide a white solid. Yield: 100 mg, 0.20 mmol, 61%.
^{1}H NMR (500 MHz, Chloroform-d) δ 8.50 (d, J = 4.3 Hz, 2H), 7.65 (d, J = 7.9 Hz, 1H), 7.41 (d, J = 8.4 Hz, 1H), 7.30 (t, J = 7.7 Hz, 1H), 7.22 – 7.17 (m, 3H), 7.14 – 7.11 (m, 2H), 7.07 – 7.03 (m, 2H), 6.59 (d, J = 1.8 Hz, 1H), 5.48 (s, 2H), 5.30 (d, J = 8.0 Hz, 1H), 4.50 (s, 1H), 4.09 (s, 1H), 3.89 (d, J = 7.1 Hz, 1H), 2.95 (t, J = 7.3 Hz, 2H), 2.82 (s, 2H), 2.45 (t, J = 7.1 Hz, 2H), 1.83 (bs, 1H), 1.62 (bs, 1H), 1.02 (bs, 1H), 0.36 (bs, 1H). TOF ES+ MS: (M + H) 501.2, (M + Na) 523.2.

4-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)-piperazine-1-carboxamide **(CCG-212390).** *t*-Butyl 4-((2-(pyridin-4-yl)ethyl)carbamoyl)piperazine-1-carboxylate (233 mg, 0.70 mmol) was dissolved in anhydrous 1,4-dioxane (1 mL), and 4 M HCl in 1,4-dioxane (1.0 mL, 4.18 mmol) was added. The reaction was allowed to stir for 2 h, after which time the solvent was removed and the residue was solidified via sonication in Et$_2$O. The solid was washed with Et$_2$O then thoroughly dried under high vacuum to give the dihydrochloride salt as a tan powder. Yield: 214 mg, 0.70 mmol, 100%.

4-((2-(pyridin-4-yl)ethyl)carbamoyl)piperazine dihydrochloride (214 mg, 0.70 mmol), 1-(4-chlorobenzyl)-1*H*-indole-2-carboxylic acid (200 mg, 0.70 mmol), TEA (0.388 mL, 2.79 mmol), EDC·HCl (147 mg, 0.766 mmol), and HOBT (117 mg, 0.766 mmol) were dissolved in DCM (10 mL) and stirred for 16 h at RT. After this time, the reaction was diluted with EtOAc and washed with H$_2$O (3X), 10% Na$_2$CO$_3$ (3X), and brine (1X), then dried (MgSO$_4$) and concentrated. The resulting residue was purified via silica flash chromatography (25 g silica, 1% to 20% methanolic ammonia:EtOAc) to

provide a white solid. Yield: 224 mg, 0.446 mmol, 64%. ^{1}H NMR (400 MHz, Chloroform-d) δ 8.47 (d, J = 5.4 Hz, 2H), 7.64 (d, J = 7.9 Hz, 1H), 7.36 (d, J = 8.4 Hz, 1H), 7.31 − 7.26 (m, 1H), 7.17 (dd, J = 12.9, 7.8 Hz, 3H), 7.11 (d, J = 5.4 Hz, 2H), 6.99 (d, J = 8.2 Hz, 2H), 6.63 (s, 1H), 5.47 (s, 2H), 4.72 (t, J = 5.4 Hz, 1H), 3.60 (s, 4H), 3.49 (q, J = 6.6 Hz, 2H), 3.16 (bs, 4H), 2.82 (t, J = 6.9 Hz, 2H). TOF ES+ MS: (M + H) 502.2, (M + Na) 524.2. HPLC t_R = 5.40 min, > 95%.

1-(2-(4-Chlorobenzoyl)benzoyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide (**CCG-212391**). 2-(4-chlorobenzoyl)benzoic acid (366 mg, 1.404 mmol) was added to DCM (20 mL), followed by amine dihydrochloride **36** (473 mg, 1.544 mmol), TEA (978 µL, 7.02 mmol), EDC·HCl (296 mg, 1.544 mmol), and HOBT (237 mg, 1.544 mmol.) The reaction was allowed to stir at RT for 20 h, at which time the solvent was removed *in vacuo*. The residue was taken up in EtOAc and washed with H_2O (1X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated. The residue was then purified by silica flash chromatography (25 g silica, 1% to 20% 7 M methanolic ammonia:EtOAc.) ^{1}H NMR (400 MHz, Chloroform-d) δ 8.47 (d, J = 5.5 Hz, 2H), 7.72 (d, J = 8.4 Hz, 2H), 7.61 − 7.53 (m, 1H), 7.52 − 7.24 (m, 5H), 7.10 (d, J = 5.4 Hz, 2H), 5.98 (t, J = 5.5 Hz, 1H), 4.47 (d, J = 12.9 Hz, 1H), 3.62 (d, J = 13.5 Hz, 1H), 3.51 (q, J = 6.6 Hz, 2H), 3.02 (t, J = 11.6 Hz, 1H), 2.81 (t, J = 6.9 Hz, 2H), 2.71 (t, J = 11.9 Hz, 1H), 2.28 (ddd, J = 14.9, 11.1, 3.9 Hz, 1H), 1.91 − 1.52 (m, 4H). TOF ES+ MS: (M + H) 476.2, (M + Na) 498.2. HPLC t_R = 4.83 min, > 95%.

1-(2-(4-chlorophenoxy)benzoyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-

carboxamide **(CCG-212392)**. Methyl 2-(4-chlorophenoxy)benzoate **41** was dissolved in

EtOH (5 mL), followed by the addition of 10% aq. NaOH (2 mL). The reaction was

allowed to stir for 14 h, after which time the solvent was stripped off *in vacuo* and the

residue re-dissolved in H_2O. This aqueous solution was then cooled to 0 °C and acidified

to pH<1 with conc. HCl, and the resulting precipitate was collected over a filter, then

dried via aspirator and high vacuum to provide the desired carboxylic acid as a white

powder. No further purification necessary. Yield: 92 mg, 0.37 mmol, 97%.

All of the previously prepared carboxylic acid (92 mg, 0.37 mmol) was added to

DCM (5 mL), followed by amine dihydrochloride **36** (136 mg, 0.44 mmol), TEA (310

µL, 2.22 mmol), EDC·HCl (85 mg, 0.44 mmol), and HOBT (68 mg, 0.44 mmol). The

solution was allowed to stir at RT for 18 h, after which time the reaction was

concentrated *in vacuo* and the resulting residue was taken up in EtOAc and washed with

H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated.

The resulting residue was purified by silica flash chromatography (10 g silica, 1% to 20%

methanolic ammonia:EtOAc) to give the desired compound as a thick oil. ^{1}H NMR (500

MHz, Chloroform-d) δ 8.43 (d, J = 4.7 Hz, 2H), 7.28 (dt, J = 29.2, 7.9 Hz, 4H), 7.14 (t, J

= 7.4 Hz, 1H), 7.06 (d, J = 4.8 Hz, 2H), 6.97 − 6.76 (m, 3H), 6.15 (d, J = 47.8 Hz, 1H),

4.57 (d, J = 12.6 Hz, 1H), 3.63 (bs, 1H), 3.48 (q, J = 6.5 Hz, 2H), 3.04 − 2.88 (m, 1H),

2.77 (t, J = 6.9 Hz, 2H), 2.75 − 2.62 (m, 1H), 2.24 (t, J = 11.0 Hz, 1H), 1.81 − 1.73 (m,

1H), 1.72 − 1.61 (m, 2H), 1.61 − 1.51 (m, 1H). TOF ES+ MS: (M + H) 464.1, (M + Na)

486.1. HPLC t_R = 4.79 min, > 90%.

1-(2-((4-Chlorophenyl)(methyl)amino)benzoyl)-N-(2-(pyridin-4-yl)ethyl)-

piperidine-4-carboxamide **(CCG-212393)**. Methyl 2-((4-chlorophenyl)(methyl)amino)benzoate **45** (120 mg, 0.44 mmol) was dissolved in EtOH (8 mL), followed by the addition of 10% aq. NaOH (3 mL). The reaction was allowed to stir at RT for 6 h, after which time the solvent was removed and the residue was taken up in H_2O, cooled to 0 °C, and acidified to pH<1. The resulting white precipitate was collected over a filter, dried under high vacuum, and used without further purification. Yield: 108 mg, 0.41 mmol, 95%. ^{1}H NMR (400 MHz, Chloroform-d) δ 9.44 (s, 1H), 7.96 (dd, *J* = 8.0, 1.5 Hz, 1H), 7.35 – 7.24 (m, 3H), 7.17 (t, *J* = 9.0 Hz, 3H), 6.79 – 6.72 (m, 1H), 3.89 (s, 3H).

2-((4-chlorophenyl)(methyl)amino)benzoic acid (85 mg, 0.33 mmol), amine dihydrochloride **36** (119 mg, 0.39 mmol), TEA (270 µL, 1.95 mmol), EDC·HCl (75 mg, 0.39 mmol), and HOBT (60 mg, 0.39 mmol) were added to DCM (5 mL) sequentially, and the solution was allowed to stir at RT for 20 h. After this time, the solvent was removed *in vacuo* and the residue was dissolved in EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated. The residue was then purified via silica flash chromatography (10 g silica, 1% to 20% methanolic ammonia:EtOAc) to provide the desired compound as a thick colorless oil. Yield: 90 mg, 0.19 mmol, 58%. ^{1}H NMR (400 MHz, Chloroform-d, mixture of rotamers) δ 8.48 (m, 2H), 7.39 (t, *J* = 8.1 Hz, 1H), 7.34 – 7.19 (m, 3H), 7.09 (d, *J* = 7.9 Hz, 4H), 6.63 (t, *J* = 9.6 Hz, 2H), 5.75 (t, *J* = 5.4 Hz, 0.5H), 5.41 (t, *J* = 5.4 Hz, 0.5H), 4.64 – 4.44 (m, 1H), 3.61 – 3.39 (m, 3H), 3.22 (s, 1.5H), 3.17 (s, 1.5H), 2.93 – 2.84 (m, 0.5H), 2.83 – 2.74 (m, 2H), 2.74 – 2.63 (m, 1H), 2.53 – 2.44 (m, 0.5H), 2.22 – 2.09 (m, 1H), 1.82 (bd, *J* = 13.1

Hz, 0.5H), 1.78 – 1.51 (m, 3.5H), 1.41 (bd, *J* = 13.2 Hz, 0.5H), 1.30 (qd, *J* = 12.9, 4.2 Hz, 0.5H). TOF ES+ MS: (M + H) 477.2, 499.1. HPLC t_R = 5.13 min, > 90% purity.

(3aS,7aR)-5-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-2-(2-(pyridin-4-yl)ethyl)octahydro-1H-pyrrolo[3,4-c]pyridin-1-one **(CCG-212394).** 5-(1-(4-chlorobenzyl)-1*H*-indole-2-carbonyl)octahydro-1*H*-pyrrolo[3,4-c]pyridin-1-one **91** (35 mg, 0.09 mmol) was dissolved in anhydrous DMF (5 mL). A 60% oil suspension of NaH (17 mg, 0.43 mmol) was added at RT under N_2 and stirred for 1 h. 4-(2-Bromoethyl)pyridine (19 mg, 0.10 mmol) was added and stirring continued for 22 h under the same conditions. After this time, the solution was diluted with EtOAc and washed with H_2O (1X), 10% aq. Na_2CO_3 (2X), and brine (1X), dried with $MgSO_4$, and concentrated *in vacuo*. Purification was done via silica flash chromatography (10 g silica, 1 to 20% methanolic ammonia:EtOAc.) The residue was dissolved in DCM, and rapid solvent removal afforded the title compound as a white powder. Yield: 36 mg, 0.07 mmol, 82%. ^{1}H NMR (400 MHz, Chloroform-d) δ 8.51 (d, *J* = 4.8 Hz, 2H), 7.68 (d, *J* = 7.9 Hz, 1H), 7.29 – 7.23 (m, 2H), 7.23 – 7.17 (m, 3H), 7.17 – 7.11 (m, 3H), 7.05 (d, *J* = 8.0 Hz, 2H), 5.45 (d, *J* = 7.0 Hz, 2H), 4.43 (d, *J* = 13.7 Hz, 1H), 4.10 (d, *J* = 3.8 Hz, 1H), 3.66 – 3.49 (m, 2H), 3.41 – 3.34 (m, 1H), 3.32 – 3.25 (m, 1H), 2.92 – 2.77 (m, 4H), 2.42 (bs, 2H), 2.10 (bs, 1H), 1.75 (bs, 1H), 1.31 (bs, 1H). TOF ES+ MS: (M + H) 513.2, (M + Na) 535.2. HPLC t_R = 5.53 min, > 95% purity.

1-(2-((4-Chlorobenzyl)amino)benzoyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-222660)**. Methyl 2-((4-chlorobenzyl)amino)benzoate **47a** (80 mg,

0.29 mmol) was dissolved in MeOH (8 mL) and 10% aq. NaOH (4 mL) was added and the reaction was stirred at RT for 24 h. At this time, the solvent was removed *in vacuo* and the residue was dissolved in H_2O and acidified with conc. HCl to pH<1 at 0 °C. The resulting precipitate was collected over a filter and dried via aspirator then high vacuum to afford the desired carboxylic acid as a white powder. No further purification necessary. Yield: 75 mg, 0.29 mmol, 99%.

2-((4-chlorobenzyl)amino)benzoic acid (75 mg, 0.29 mmol) was dissolved in DCM (12 mL) along with TEA (325 μL, 2.32 mmol), EDC·HCl (67 mg, 0.35 mmol), HOBT (53 mg, 0.35 mmol), and *N*-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide dihydrochloride (133 mg, 0.44 mmol). The solution was stirred at RT for 20 h, after which time the DCM was stripped off *in vacuo* and the resulting residue was taken up in EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated. The residue was then purified via silica flash chromatography (10 g silica, 1% to 30% methanolic ammonia:EtOAc) to give the desired compound. Yield: 63 mg, 0.132 mmol, 46%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.60 − 8.51 (m, 2H), 7.30 − 7.25 (m, 4H), 7.22 − 7.15 (m, 1H), 7.14 − 7.10 (m, 2H), 7.08 (dd, *J* = 7.6, 1.6 Hz, 1H), 6.67 (t, *J* = 7.4 Hz, 1H), 6.58 (d, *J* = 8.4 Hz, 1H), 5.58 (t, *J* = 5.2 Hz, 1H), 5.53 (t, *J* = 5.7 Hz, 1H), 4.33 (m, 4H), 3.57 (q, *J* = 6.7 Hz, 2H), 2.95 (t, *J* = 12.8 Hz, 2H), 2.85 (t, *J* = 6.9 Hz, 2H), 2.30 (tt, *J* = 11.1, 3.8 Hz, 1H), 1.82 (m, 2H), 1.74 (dt, *J* = 15.9, 7.9 Hz, 2H). TOF ES+ MS: (M + H) 477.2, (M + Na) 499.2. HPLC t_R = 5.33 min, > 90%.

1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)azepane-4-carboxamide **(CCG-222661).** The ethyl ester **132** (160 mg, 0.37 mmol) was dissolved in

EtOH (6 mL) and 10% aq. NaOH (4 mL) was added and the reaction was stirred at RT for 24 h. At this time, the solvent was removed *in vacuo* and the white residue was dissolved in H_2O and acidified with conc. HCl to pH<1 at 0 °C. The resulting precipitate was collected over a filter and dried via aspirator then high vacuum to afford the desired carboxylic acid as a white powder. No further purification necessary. Yield: 144 mg, 0.35 mmol, 96%.

The carboxylic acid (60 mg, 0.15 mmol), amine **27** (21 μL, 0.18 mmol), EDC·HCl (34 mg, 0.18 mmol), HOBT (27 mg, 0.18 mmol), and TEA (61 μL, 0.44 mmol) were dissolved in DCM (10 mL) and stirred at RT for 29 h, at which time the DCM was removed *in vacuo* and the residue was taken up in EtOAc and washed with H_2O (1X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic extract was dried ($MgSO_4$) and concentrated, and the residue purified via silica flash chromatography (10 g silica, 1% to 20% methanolic ammonia:EtOAc) to give a colorless oil. Yield: 40 mg, 0.76 mmol, 52%. HPLC t_R = 5.53 min, > 95%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.51 (bs, 2H), 7.65 (d, *J* = 7.6 Hz, 1H), 7.39 − 7.32 (m, 1H), 7.28 (s, 1H), 7.16 (d, *J* = 7.4 Hz, 3H), 7.12 (bs, 2H), 7.02 (d, *J* = 8.0 Hz, 2H), 6.65 (d, *J* = 6.3 Hz, 1H), 5.48 (d, *J* = 9.4 Hz, 2H), 3.77 − 3.64 (m, 2H), 3.60 − 3.36 (m, 4H), 3.31 − 3.15 (m, 1H), 2.86 − 2.77 (m, 2H), 2.08 − 1.97 (m, 2H), 1.91 − 1.69 (m, 3H), 1.56 − 1.48 (m, 1H), 1.43 − 1.34 (m, 1H). TOF ES+ MS: (M + H) 515.2, (M + Na) 537.2. HPLC t_R = 5.52 min, > 95%.

1-(2-(4-Chlorobenzyl)benzoyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-222821)**. 2-(4-chlorobenzyl)benzoic acid (40 mg, 0.16 mmol), *N*-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide dihydrochloride (55 mg, 0.18 mmol), TEA (225 μL,

1.62 mmol), EDC·HCl (37 mg, 0.20 mmol), and HOBT (30 mg, 0.20 mmol) were dissolved in DCM (10 mL) and stirred at RT for 24 h. The DCM was then removed *in vacuo* and the residue was taken up in EtOAc, then washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated. Purification via silica flash chromatography (4 g silica, 1% to 20% methanolic ammonia:EtOAc) provided the desired compound as white microcrystals upon concentration. ^{1}H NMR (500 MHz, Chloroform-d, 3:2 mixture of rotamers) δ 8.51 (bs, 2H), 7.33 (t, J = 7.3 Hz, 1H), 7.27 − 7.18 (m, 4H), 7.17 − 7.05 (m, 5H), 5.61 − 5.42 (m, 1H), 4.73 − 4.59 (m, 1H), 4.18 − 4.03 (m, 1H), 3.93 − 3.80 (m, 1H), 3.53 (q, J = 6.5 Hz, 2H), 3.37 − 3.15 (m, 1H), 2.86 − 2.81 (m, 2H), 2.50 (td, J = 12.7, 3.1 Hz, 1H), 2.23 − 1.99 (m, 2H), 1.90 − 1.75 (m, 2H), 1.73 − 1.49 (m, 2H). TOF ES+ MS: (M + H) 462.2, (M + Na) 484.2. HPLC t_R = 4.90 min, > 90%.

1-(1-(4-Chlorobenzyl)-2-oxo-1,2-dihydroquinolin-4-yl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-222822)**. Ethyl 1-(1-(4-chlorobenzyl)-2-oxo-1,2-dihydroquinolin-4-yl)piperidine-4-carboxylate (75 mg, 0.18 mmol) was dissolved into a mixture of EtOH (4 mL), THF (4 mL), and 10% aq. NaOH (2 mL) and stirred at RT for 18 h. After this time, the reaction was concentrated and the residue dissolved in H_2O, chilled to 0 °C, and acidified to pH<1 with conc. HCl. The resulting precipitate was collected over a filter and washed with cold 1M HCl, air dried via aspirator, then dried under high vacuum to give 1-(1-(4-chlorobenzyl)-2-oxo-1,2-dihydroquinolin-4-yl)piperidine-4-carboxylic acid as a white solid. No further purification necessary. Yield: 67 mg, 0.17 mmol, 95%.

1-(1-(4-chlorobenzyl)-2-oxo-1,2-dihydroquinolin-4-yl)piperidine-4-carboxylic acid (48 mg, 0.12 mmol), 4-(2-aminoethyl)pyridine (17 µL, 0.15 mmol), TEA (51 µL, 0.36 mmol), EDC·HCl (28 mg, 0.15 mmol), and HOBT (22 mg, 0.15 mmol) were dissolved in DCM (5 mL) and stirred at RT for 20 h. DCM was then removed *in vacuo* and the residue taken up in EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated. The residue was purified by silica flash chromatography (10 g silica, 1% to 20% methanolic ammonia:EtOAc). Yield: 45 mg, 0.05 mmol, 45%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.54 (s, 2H), 7.78 (d, J = 8.1 Hz, 1H), 7.40 (t, J = 7.8 Hz, 1H), 7.26 (d, J = 8.0 Hz, 2H), 7.21 − 7.00 (m, 6H), 6.22 (s, 1H), 5.81 (t, J = 5.4 Hz, 1H), 5.46 (s, 2H), 3.58 (dt, J = 16.9, 8.7 Hz, 4H), 2.87 (t, J = 6.9 Hz, 2H), 2.74 (t, J = 11.6 Hz, 2H), 2.28 (tt, J = 11.4, 3.7 Hz, 1H), 2.05 (qd, J = 12.5, 12.0, 3.5 Hz, 2H), 1.94 (d, J = 11.2 Hz, 2H). TOF ES+ MS: (M + H) 501.2, (M + Na) 523.2. HPLC t_R = 5.25 min, >95% purity.

1-(1-(4-Chlorophenyl)-2-oxo-1,2-dihydroquinolin-4-yl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide (**CCG-222823**). Ethyl 1-(1-(4-chlorophenyl)-2-oxo-1,2-dihydroquinolin-4-yl)piperidine-4-carboxylate **58** (150 mg, 0.37 mmol) was dissolved in THF (10 mL) followed by the addition of 10% aq. NaOH (5 mL). This biphasic reaction was stirred at RT for 20 h. The solvent was then stripped off *in vacuo* and the resulting residue was dissolved in H_2O, chilled to 0°C, then acidified to pH < 1. The resulting precipitate was collected over a filter, air dried, then high vacuum dried to provide   1-(1-(4-chlorophenyl)-2-oxo-1,2-dihydroquinolin-4-yl)piperidine-4-carboxylic

acid as a white powder. No further purification necessary. Yield: 130 mg, 0.33 mmol, 91%.

1-(1-(4-Chlorophenyl)-2-oxo-1,2-dihydroquinolin-4-yl)piperidine-4-carboxylic acid (66 mg, 0.17 mmol), 4-(2-aminoethyl)pyridine (25 μL, 0.21 mmol), TEA (72 μL, 0.52 mmol), ED C (40 mg, 0.21 mmol), and HOBT (32 mg, 0.21 mmol) were dissolved in DCM (5 mL) and stirred at RT for 19 h. The reaction was then diluted with EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated. The residue was purified via silica flash chromatography (10 g silica, 1 to 17% methanolic ammonia/EtOAc) to give the title compound as an off-white powder. Yield: 47 mg, 0.10 mmol, 56%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.55 (d, J = 5.1 Hz, 2H), 7.79 (d, J = 7.9 Hz, 1H), 7.55 (d, J = 8.2 Hz, 2H), 7.32 (t, J = 7.5 Hz, 1H), 7.25 − 7.18 (m, 3H), 7.15 (d, J = 4.9 Hz, 2H), 6.66 (d, J = 8.4 Hz, 1H), 6.20 (s, 1H), 5.78 (t, J = 6.0 Hz, 1H), 5.30 (s, 1H), 3.58 (q, J = 9.6, 6.8 Hz, 4H), 2.85 (t, J = 6.8 Hz, 2H), 2.77 (t, J = 11.9 Hz, 2H), 2.34 − 2.21 (m, 2H), 2.07 (q, J = 13.3 Hz, 2H), 1.95 (d, J = 13.5 Hz, 2H). TOF ES+ MS: (M + H) 487.2, (M + Na) 509.2. HPLC t_R = 5.09 min, >95% purity.

1-(9H-Carbazole-1-carbonyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide (**CCG-222824**). 9H-carbazole-1-carboxylic acid was prepared according to *Journal of Organic Chemistry*, Vol. 53, No. 4, 1988, 794-799.

9*H*-Carbazole-1-carboxylic acid (120 mg, 0.57 mmol), *N*-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide dihydrochloride (190 mg, 0.63 mmol), TEA (475 μL, 3.41 mmol), EDC·HCl (120 mg, 0.63 mmol) and HOBT (96 mg, 0.63 mmol) were dissolved in DCM (10 mL) and stirred at RT for 14 h. At this time, the reaction was

diluted with EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated. The residue was purified via silica flash chromatography (10 g silica, 10% to 30% methanolic ammonia/EtOAc) to give a tan powder. Yield: 170 mg, 0.40 mmol, 70%. ^{1}H NMR (500 MHz, Chloroform-d) δ 9.13 (s, 1H), 8.53 (d, J = 5.6 Hz, 2H), 8.14 (d, J = 7.7 Hz, 1H), 8.08 (d, J = 7.8 Hz, 1H), 7.50 – 7.42 (m, 2H), 7.41 (d, J = 7.5 Hz, 1H), 7.26 – 7.23 (m, 1H), 7.21 (t, J = 7.6 Hz, 1H), 7.11 (d, J = 5.3 Hz, 2H), 5.59 (t, J = 5.5 Hz, 1H), 4.46 (s, 2H), 3.56 (q, J = 6.6 Hz, 2H), 3.04 (t, J = 12.3 Hz, 2H), 2.84 (t, J = 6.9 Hz, 2H), 2.32 (ddd, J = 15.0, 10.9, 4.1 Hz, 1H), 1.91 – 1.72 (m, 4H). TOF ES+ MS: (M + H) 427.2, (M + Na) 449.2. HPLC t_R = 4.80 min, >95% purity.

1-(6-Chloro-9H-carbazole-1-carbonyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide (**CCG-222825**). Methyl 6-chloro-9*H*-carbazole-1-carboxylate (54 mg, 0.21 mmol) was dissolved in EtOH (4 mL) and THF (5 mL), then 10% aq. NaOH (2 mL) was added. The reaction was allowed to stir at RT for 22 h, after which time the solvent was stripped off *in vacuo*. The residue was taken up in H_2O and acidified to pH<1 with conc. HCl, and the resulting white precipitate was collected over a filter, air dried, then high vacuum dried to give 6-chloro-9H-carbazole-1-carboxylic acid as a white powder with no further purification. Yield: 51 mg, 0.21 mmol, 100%.

6-Chloro-9*H*-carbazole-1-carboxylic acid (51 mg, 0.21 mmol) was added to DCM (5 mL), followed by *N*-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide dihydrochloride (76 mg, 0.25 mmol), TEA (145 µL, 1.04 mmol), EDC·HCl (48 mg, 0.25 mmol) and HOBT (38 mg, 0.25 mmol). This solution was stirred at RT for 16 h, at which time the

solvent was removed *in vacuo* and the residue taken up in EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brined (1X). The extract was dried ($MgSO_4$) and concentrated, and the residue purified via silica flash chromatography (10 g silica, 1% to 20% methanolic ammonia: EtOAc). Yield: 48 mg, 0.10 mmol, 50%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.53 (d, *J* = 4.7 Hz, 2H), 8.10 − 8.06 (m, 1H), 8.04 − 7.98 (m, 1H), 7.48 − 7.36 (m, 3H), 7.22 (ddd, *J* = 7.6, 5.9, 1.0 Hz, 1H), 7.13 (d, *J* = 5.1 Hz, 2H), 5.61 − 5.52 (m, 1H), 4.45 (bs, 2H), 3.57 (q, *J* = 6.6 Hz, 2H), 3.06 (s, 2H), 2.85 (t, *J* = 6.9 Hz, 2H), 2.34 (tt, *J* = 10.9, 4.6 Hz, 1H), 1.91 − 1.69 (m, 4H). TOF ES+ MS: (M + H) 461.2, (M + Na) 483.2. HPLC t_R = 5.23 min, > 90% purity.

Exo-(1R,5S,6r)-3-(1-(4-chlorobenzyl)-1H-indole-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)-3-azabicyclo[3.1.0]hexane-6-carboxamide (**CCG-222980**). The ethyl ester **113** (206 mg, 0.49 mmol) was dissolved in anhydrous THF (10 mL), followed by the addition of KOTMS (66 mg, 0.51 mmol). The reaction stirred at RT for 48 h, solvent was removed *in vacuo*, the residue was dried under high vacuum overnight, leaving the potassium salt residue that was taken directly into the next step. Yield: 213 mg, 0.49 mmol, 100%.

The potassium salt (211 mg, 0.49 mmol), amine **27** (64 µL, 0.54 mmol), HATU (204 mg, 0.54 mmol), and DIPEA (170 µL, 0.97 mmol) were dissolved in DCM (10 mL) and stirred at RT for 24 h, at which time the DCM was removed *in vacuo* and the residue was diluted in EtOAc and washed with H_2O (2X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic extract was dried ($MgSO_4$) and concentrated, and the residue purified via silica flash chromatography (25 g silica, 1% to 20% methanolic ammonia:EtOAc) to give

an off-white powder. Yield: 134 mg, 0.27 mmol, 55%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.44 (d, J = 5.0 Hz, 2H), 7.61 (d, J = 7.9 Hz, 1H), 7.33 – 7.24 (m, 2H), 7.17 – 7.10 (m, 3H), 7.06 (d, J = 5.0 Hz, 2H), 6.88 (d, J = 8.1 Hz, 2H), 6.66 (s, 1H), 6.07 (t, J = 5.9 Hz, 1H), 5.55 – 5.34 (m, 2H), 4.02 (d, J = 12.3 Hz, 1H), 3.72 – 3.58 (m, 2H), 3.53 – 3.32 (m, 4H), 2.75 (t, J = 7.0 Hz, 2H), 1.99 (d, J = 9.4 Hz, 2H). TOF ES+ MS: (M + H) 499.2, (M + Na) 521.2. HPLC t_R = 5.36 min, > 95%.

1-(2-(Phenylamino)benzoyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide **(CCG-222981)**. Methyl 2-(phenylamino)benzoate (74 mg, 0.33 mmol) was dissolved in 10% aq. NaOH (1 mL), THF (5 mL), and EtOH (2 mL), and stirred at RT for 5 h. At this time, the solvent was stripped off *in vacuo* and the residue was dissolved in H_2O and acidified with conc. HCl to pH < 1. Material was collected over a filter and air-dried then dried under high vacuum to give 2-(phenylamino)benzoic acid as a white powder. No further purification necessary. Yield: 62 mg, 0.29 mmol, 89%.

2-(Phenylamino)benzoic acid (56 mg, 0.26 mmol), *N*-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide dihydrochloride (97 mg, 0.32 mmol), EDC·HCl (60 mg, 0.32 mmol), HOBT (48 mg, 0.32 mmol), and TEA (220 μL, 1.58 mmol) were dissolved in DCM (10 mL) and stirred at RT for 17 h. After this time, the DCM was removed *in vacuo*. The residue was taken up in EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X), and dried ($MgSO_4$) and concentrated. The residue was purified via silica flash chromatography (10 g silica, 1% to 20% methanolic NH_3/EtOAc) to give the desired compound as a colorless oil. Yield: 53 mg, 0.12 mmol, 47%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.47 (d, J = 5.0 Hz, 2H), 7.37 (d, J = 8.3 Hz, 1H), 7.28 –

7.21 (m, 3H), 7.15 (d, J = 7.6 Hz, 1H), 7.10 – 7.02 (m, 4H), 6.93 (t, J = 7.8 Hz, 1H), 6.86 (t, J = 7.4 Hz, 1H), 5.78 (s, 1H), 4.26 (s, 2H), 3.50 (q, J = 6.5 Hz, 2H), 2.92 (bs, 2H), 2.80 (t, J = 6.9 Hz, 2H), 2.28 (ddd, J = 14.7, 11.0, 3.4 Hz, 1H), 1.77 (bs, 2H), 1.70 – 1.57 (m, 2H). TOF ES+ MS: (M + H) 429.2, (M + Na) 451.2. HPLC t_R = 5.05 min, > 95%.

1-(2-(Benzylamino)benzoyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide

(**CCG-222982**). Methyl 2-(benzylamino)benzoate (50 mg, 0.21 mmol) was dissolved in EtOH (2 mL) and THF (5 mL), then 10% aq. NaOH (2 mL) was added and the reaction was stirred at RT for 15 h. At this time, the solvent was removed *in vacuo* and the residue was dissolved in H_2O and acidified with conc. HCl to pH<1. The resulting precipitate was collected over a filter and dried via aspirator then high vacuum to afford the desired 2-(benzylamino)benzoic acid as a white powder. No further purification necessary. Yield: 41 mg, 0.18 mmol, 88%.

Ethyl 2-(benzylamino)benzoate (30 mg, 0.13 mmol) was dissolved in DCM (5 mL), followed by *N*-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide dihydrochloride (45 mg, 0.15 mmol), TEA (0.11 mL, 0.79 mmol), EDC·HCl (30 mg, 0.16 mmol), and HOBT (24 mg, 0.16 mmol). The reaction was then stirred at RT for 16 h, at which time the reaction was diluted with EtOAc and washed with H_2O (2X), 10% aq. Na_2CO_3 (2X), and brine (1X), then dried (MgSO$_4$) and concentrated *in vacuo*. The resulting residue was then purified by silica flash chromatography (10 g silica, 1% to 20% methanolic ammonia:EtOAc) to provide the desired product as a colorless oil. Yield: 26 mg, 0.06 mmol, 48%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.48 (d, J = 5.7 Hz, 2H), 7.32 (d, J = 6.7 Hz, 4H), 7.26 – 7.21 (m, 1H), 7.17 (t, J = 7.8 Hz, 1H), 7.10 (d, J = 5.7 Hz, 2H), 7.07

− 7.02 (m, 1H), 6.63 (dd, *J* = 7.8, 3.8 Hz, 2H), 5.92 (t, *J* = 5.6 Hz, 1H), 5.44 (t, *J* = 5.4

Hz, 1H), 4.33 (d, *J* = 5.3 Hz, 2H), 4.26 (bs, 2H), 3.52 (q, *J* = 6.7 Hz, 2H), 2.90 (t, *J* = 11.7

Hz, 2H), 2.81 (t, *J* = 6.9 Hz, 2H), 2.28 (tt, *J* = 11.2, 3.5 Hz, 1H), 1.78 (d, *J* = 12.1 Hz,

2H), 1.69 (qd, *J* = 12.9, 12.4, 4.0 Hz, 2H). TOF ES+ MS: (M + H) 443.2, (M + Na)

465.2. HPLC t_R = 4.92 min, > 95%.

1-(2-((4-Chlorophenethyl)amino)benzoyl)-N-(2-(pyridin-4-yl)ethyl)piperidine-4-

carboxamide (**CCG-222983**). 2-((4-chlorophenethyl)amino)benzoic acid (40 mg, 0.15

mmol), *N*-(2-(pyridin-4-yl)ethyl)piperidine-4-carboxamide dihydrochloride (53 mg, 0.17

mmol), EDC·HCl (33 mg, 0.17 mmol), HOBT (26 mg, 0.17 mmol), and TEA (0.12 mL,

0.87 mmol) were dissolved in DCM (10 mL) and stirred at RT for 24 h, at which time the

DCM was removed *in vacuo* and the residue was taken up in EtOAc and washed with

H_2O (2X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic extract was dried ($MgSO_4$)

and concentrated, and the residue purified via silica flash chromatography (10 g silica,

1% to 20% methanolic ammonia:EtOAc) to give a colorless oil. Yield: 41 mg, 0.083

mmol, 57%. HPLC t_R = 5.55 min, > 95%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.52 (d,

J = 5.2 Hz, 2H), 7.30 − 7.21 (m, 3H), 7.16 (d, *J* = 8.2 Hz, 2H), 7.12 (d, *J* = 5.2 Hz, 2H),

7.05 (d, *J* = 7.5 Hz, 1H), 6.69 (d, *J* = 8.3 Hz, 1H), 6.65 (d, *J* = 7.4 Hz, 1H), 5.57 (t, *J* =

5.6 Hz, 1H), 4.97 (bs, 1H), 4.21 (bs, 2H), 3.56 (q, *J* = 6.5 Hz, 2H), 3.34 (t, *J* = 7.2 Hz,

2H), 2.89 (t, *J* = 7.2 Hz, 2H), 2.83 (q, *J* = 12.6, 9.7 Hz, 4H), 2.24 (tt, *J* = 11.2, 3.7 Hz,

1H), 1.77 − 1.62 (m, 7H). TOF ES+ MS: (M + H) 491.2, (M + Na) 513.2. HPLC t_R =

5.44 min, >95% purity.

(1R,3s,5S)-8-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)-8-azabicyclo[3.2.1]octane-3-carboxamide **(CCG-224000).** The Boc-protected right-hand half **115** (100 mg, 0.35 mmol) was dissolved into 4M HCl (5 mL) and stirred for 15 min at RT. After this time, Et_2O (15 mL) was added to precipitate the material and the reaction was sonicated for 15 min. The resulting residue did not solidify, so the liquid phase was decanted off and the residue was dried under aspirator vacuum, then high vacuum, for 48 h. The resulting solid aminde dihydrochloride **116** was then taken directly into the subsequent reaction. Yield: 115 mg, 0.35 mmol, 99%.

The indole carboxylic acid **4a** (52 mg, 0.18 mmol), amine dihydrochloride **116** (60 mg, 0.18 mmol), EDC·HCl (45 mg, 0.24 mmol), HOBT (36 mg, 0.24 mmol), and TEA (252 μL, 0.24 mmol) were dissolved in DCM (3 mL) and stirred at RT for 20 h, at which time the DCM was removed *in vacuo* and the residue was diluted in EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic extract was dried ($MgSO_4$) and concentrated, and the residue purified via silica flash chromatography (10 g silica, 1% to 20% methanolic ammonia:EtOAc) to give an off-white powder. Yield: 59 mg, 0.11 mmol, 61%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.44 (d, J = 4.8 Hz, 2H), 7.63 (d, J = 7.9 Hz, 1H), 7.34 (d, J = 8.4 Hz, 1H), 7.26 (t, J = 7.7 Hz, 1H), 7.19 − 7.12 (m, 3H), 7.07 (d, J = 5.0 Hz, 2H), 6.98 (d, J = 8.1 Hz, 2H), 6.74 (s, 1H), 6.11 (t, J = 5.7 Hz, 1H), 5.51 (d, J = 9.8 Hz, 2H), 4.82 (bs, 1H), 4.39 (bs, 1H), 3.48 (q, J = 6.6 Hz, 2H), 2.78 (t, J = 7.0 Hz, 2H), 2.60 (tt, J = 11.5, 5.2 Hz, 1H), 2.30 (bs, 2H), 1.76 (dd, J = 143.8, 60.1 Hz, 7H). TOF ES+ MS: (M + H) 527.2, (M + Na) 549.2. HPLC t_R = 5.55 min, >95%.

2-(1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)piperidin-4-yl)-N-(pyridin-4-

ylmethyl)acetamide **(CCG-224001).** The ethyl ester **96** (350 mg, 0.80 mmol) was

dissolved in EtOH (10 mL), THF (10 mL), and 10% aq. KOH (3.22 mL) was added and

the reaction was stirred at RT for 13 h. At this time, the solvent was removed *in vacuo*

and the thick white residue was diluted in H_2O, sonicated, and acidified with conc. HCl to

pH<1 at 0 °C. The resulting precipitate was collected over a filter and dried via aspirator

then high vacuum to afford the desired carboxylic acid as a white powder. No further

purification necessary. Yield: 311 mg, 0.76 mmol, 95%.

The carboxylic acid (62 mg, 0.15 mmol), amine **27** (18 μL, 0.18 mmol),

EDC·HCl (35 mg, 0.18 mmol), HOBT (28 mg, 0.18 mmol), and TEA (84 μL, 0.60

mmol) were dissolved in DCM (5 mL) and stirred at RT for 18 h, at which time the DCM

was removed *in vacuo* and the residue was taken up in EtOAc and washed with H_2O

(3X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic extract was dried ($MgSO_4$) and

concentrated, and the residue purified via silica flash chromatography (10 g silica, 1% to

20% methanolic ammonia:EtOAc) to give a colorless oil. Yield: 42 mg, 0.08 mmol, 54%.

^{1}H NMR (500 MHz, Chloroform-d) δ 8.51 (d, J = 5.6 Hz, 2H), 7.64 (d, J = 7.9 Hz, 1H),

7.39 (d, J = 8.3 Hz, 1H), 7.29 (t, J = 7.7 Hz, 1H), 7.20 − 7.14 (m, 3H), 7.13 (d, J = 5.3

Hz, 2H), 7.02 (d, J = 8.2 Hz, 2H), 6.59 (s, 1H), 6.34 (t, J = 5.7 Hz, 1H), 5.45 (s, 2H), 4.59

(bs, 1H), 4.39 (s, 2H), 4.06 (bs, 1H), 2.88 (bs, 1H), 2.67 (bs, 1H), 2.13 − 2.02 (m, 3H),

1.71 (bs, 1H), 1.45 (bs, 1H), 0.97 (bs, 1H), 0.32 (bs, 1H). TOF ES+ MS: (M + H) 501.2,

(M + Na) 523.2. HPLC t_R = 5.49 min, > 95%.

1-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)azetidine-3-carboxamide **(CCG-224002).** The methyl ester **125** (100 mg, 0.26 mmol) was dissolved in EtOH (5 mL) and 10% aq. NaOH (3 mL) was added and the reaction was stirred at RT for 24 h. At this time, the solvent was removed *in vacuo* and the white residue was dissolved in H_2O and acidified with conc. HCl to pH<1 at 0 °C. The resulting precipitate was collected over a filter and dried via aspirator then high vacuum to afford the desired carboxylic acid as a white powder. No further purification necessary. Yield: 95 mg, 0.26 mmol, 99%.

The carboxylic acid (50 mg, 0.14 mmol), amine **27** (100 μL, 0.16 mmol), EDC·HCl (31 mg, 0.16 mmol), HOBT (25 mg, 0.16 mmol), and TEA (76 μL, 0.54 mmol) were dissolved in DCM (5 mL) and stirred at RT for 15 h, at which time the DCM was removed *in vacuo* and the residue was taken up in EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic extract was dried ($MgSO_4$) and concentrated, and the residue purified via silica flash chromatography (10 g silica, 1% to 20% methanolic ammonia:EtOAc) to give a colorless oil. Yield: 42 mg, 0.09 mmol, 65%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.53 (d, *J* = 5.2 Hz, 2H), 7.67 (d, *J* = 8.0 Hz, 1H), 7.34 − 7.28 (m, 2H), 7.22 (d, *J* = 8.4 Hz, 2H), 7.19 − 7.15 (m, 1H), 7.13 (s, 2H), 7.03 (d, *J* = 8.4 Hz, 2H), 6.83 (s, 1H), 5.73 (s, 2H), 5.67 (s, 1H), 4.62 (s, 1H), 4.31 (d, *J* = 71.6 Hz, 3H), 3.61 (q, *J* = 6.6 Hz, 2H), 3.22 (p, *J* = 7.4 Hz, 2H), 2.87 (t, *J* = 7.0 Hz, 2H). TOF ES+ MS: (M + H) 473.2, (M + Na) 495.2. HPLC t_R = 5.60 min, >95% purity.

2-(1-(4-Chlorobenzyl)-1H-indole-2-carbonyl)-N-(2-(pyridin-4-yl)ethyl)-2-azaspiro[3.3]heptane-6-carboxamide **(CCG-224220).** In a dry round bottom flask under

argon at RT, naphthalene (220 mg, 1.74 mmol) was dissolved in anhydrous THF (10 mL) containing hexane-washed sodium (100 mg, 4.35 mmol). The mixture was sonicated until the reaction became a dark green homogenous solution and no solid material was visible. This entire solution was then added dropwise to tosyl amide **123** (41 mg, 0.10 mmol), dissolved in anhydrous THF (1 mL), which caused the reaction to turn turbid brown. This was allowed to stir for 5 min. After this time, the reaction was diluted with EtOAc and a few drops of MeOH and washed with H_2O (1X), 10% Na_2CO_3 (3X), and brine (1X), then dried ($MgSO_4$) and concentrated. The residue of amine **124** was used directly in the subsequent reaction.

The indole carboxylic acid **4a** (29 mg, 0.10 mmol), spiro amine **124** (*approx.* 30 mg, 0.10 mmol), HATU (49 mg, 0.12 mmol), and TEA (42 µL, 0.30 mmol) were dissolved in DMF (2 mL) and stirred at RT for 18 h, at which time the reaction was diluted with EtOAc and washed with H_2O (3X), 10% aq. Na_2CO_3 (3X), and brine (1X). The organic extract was dried ($MgSO_4$) and concentrated, and the residue purified via silica flash chromatography (4 g silica, 1% to 20% methanolic ammonia:EtOAc) to give a colorless oil. Yield: 18 mg, 0.04 mmol, 35%. ^{1}H NMR (500 MHz, Chloroform-d) δ 8.50 (d, J = 4.5 Hz, 2H), 7.65 (d, J = 7.7 Hz, 1H), 7.28 (d, J = 16.1 Hz, 3H), 7.19 − 7.13 (m, 2H), 7.11 (d, J = 5.2 Hz, 2H), 6.99 (d, J = 7.6 Hz, 2H), 6.80 (s, 1H), 5.71 (s, 2H), 5.58 (bs, 1H), 4.28 (bd, J = 32.5 Hz, 2H), 4.06 (s, 2H), 3.52 (q, J = 6.7 Hz, 2H), 2.82 (t, J = 7.0 Hz, 2H), 2.74 (bs, 1H), 2.49 − 2.29 (m, 4H). TOF ES+ MS: (M + H) 513.2, (M + Na) 535.2. HPLC t_R = 5.65 min, > 95%.

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