`Supporting Information

Title: A Whole-Cell Screen for Adjunctive and Direct Antimicrobials Active Against Carbapenem-Resistant *Enterobacteriaceae*

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1. **Materials and Instrumentation:**

Reagents and solvents were purchased from commercial suppliers (Fisher Scientific {Hampton, New Hampshire, USA}, Sigma-Aldrich {St. Louis, Missouri, USA} and TCI America {Portland, OR, USA}) and used without further purification unless otherwise stated. ^{1}H NMR spectra were recorded at ambient temperature on a Varian Mercury NMR spectrometer (Palo Alto, California, USA) operating at 400 MHz in the solvent indicated with the signal of the residual solvent ($CHCl_3$ δ 7.26 ppm or DMSO-d_6 δ 2.50 ppm) as internal standard. Data are reported as follows: chemical shift, multiplicity (s = singlet, d = doublet, dd = doublet of doublets, t = triplet, q = quartet, m = multiplet), coupling constant (Hz) and integration. Thin-layer chromatography (TLC) was performed with silica gel 60 F_{254} pre-coated plates and visualized with exposure to UV light (254 nm) or by cerium ammonium molybdate (CAM) followed by heating. ESI-MS were recorded on a Agilent 1260 Infinity spectrometer (Santa Clara, CA).

2. **List of compound libraries screened:**

Library Name	**Supplier**
ChemDiv Targeted Diversity Library	ChemDiv
Enamine 2	Enamine
Enamine2a	Enamine
ActiMolTimTec1	Biomol-TimTec
Asinex 1	Asinex
Bionet (Ryan Scientific) 2	Bionet
ChemBridge3	ChemBridge
ChemDiv1 (Combilab and International)	ChemDiv
ChemDiv3	ChemDiv
ChemDiv4	ChemDiv
ChemDiv Targeted Diversity Library	ChemDiv
Enamine 2	Enamine
IFLab1	IFLab
Life Chemicals 1	Life Chemicals
Maybridge4	Maybridge
Maybridge5	Maybridge

3. **Cheminformatics:**

a) Permeation and Efflux Multiparameter Optimization (PEMPO):

Multiparameter optimization (MPO) scoring was first reported by Wager et al. to rank order compounds that would be effective as CNS drugs to penetrate the blood brain barrier.[1] The authors also suggested that compounds with a higher MPO scores demonstrate better *in vitro* ADME and safety characteristics.

We developed a variation of MPO scoring known as PEMPO (Permeation and Efflux Multiparameter Optimization), focused on identifying compounds with physicochemical properties ideal for permeating the lipopolysaccharide layer (LPS) of a Gram-negative bacterial cell, as well as avoiding extracellular efflux.

PEMPO scores were determined using the physicochemical properties below with optimal values shown in parentheses and suboptimal in brackets, outside the parentheses. A property within the optimal range received a score of 1, whereas suboptimal values were scored as a linear function from the undesired value to the optimal. Undesirable values, defined as values outside of suboptimal, received a score of 0. Physicochemical properties and desirable ranges are as follows: isoelectric point [4.0-6.0 (6.1-8.7) 8.8-10], total polar surface area [60-99 (100-200 $Å^2$) 201-240], number of hydrogen bond donors [0-1 (2-6) 7-9], number of hydrogen bond acceptors [0-5 (6-11) 12-19], partition coefficient clogP [5-3.1 (≤3)], and distribution coefficient $clogD_{7.4}$ [3-0.3 (≤0.2)].

Isoelectric point was chosen because compounds with zwitterionic character at physiological pH (7.4) exhibit charge that can be desirable for entry into porin proteins on the LPS as well as an uncharged state to aid with absorption in the gut, essential for bioavailability. Compounds that can exist in both a charged, and uncharged state are observed with a pKa close to 7.4. The remainder of the physicochemical properties were chosen to reflect the polarity of each compound, awarding higher scores to more hydrophilic molecules.

b) PEMPO scoring of known antibacterial compounds:

PEMPO scoring of known antibacterial compounds was performed across two property calculation platforms to reveal any discrepancies between scoring when properties were calculated between different engines.

Compound name	PEMPO Score[a]	PEMPO Score[b]
Chloramphenicol	4.60	4.71
Loracarbef	5.47	5.64
Ertapenem	5.00	5.00
Imipenem	6.00	6.00
Meropenem	6.00	6.00
R-115685	5.57	4.54

Cefetamet	5.00	4.98
Ceftibuten	5.00	4.59
Cefaclor	5.44	5.61
Cefadroxil	5.71	5.71
Cefamandole	5.00	4.97
Cefazolin	5.00	4.00
Cefdinir	5.00	4.71
Cefditoren	5.07	5.00
Cefixime	5.00	3.92
Cefmetazole	5.00	4.25
Cefoperazone	4.58	3.25
Cefotaxime	5.00	4.20
Cefotetan	4.58	3.63
Cefoxitin	5.00	4.96
Cefpodoxime	5.00	4.75
Cefprozil	5.72	5.72
Ceftizoxime	5.00	4.98
Ceftriaxone	4.66	3.50
Cefuroxime	5.00	4.88
Cephalexin	5.54	5.71
Cephalothin	4.83	5.00
Cephradine	5.63	5.80
T-91825	6.00	4.75
Cefepime	5.00	4.98
Cefpirome	6.00	5.88
Ceftazidime	5.00	3.75
Ceftobiprole	6.00	4.63
Iclaprim	4.25	4.36
Trimethoprim	4.68	4.86
Nalidixic Acid	4.19	4.20
ABT-492	4.79	5.00
Ciprofloxacin	5.32	5.32
Clinafloxacin	5.67	5.67
Danofloxacin	4.60	4.60
Difloxacin	4.09	4.24
DX-619	5.90	5.90
Enoxacin	5.63	5.64
Fleroxacin	4.46	4.46
Garenoxacin	4.96	5.47
Gatifloxacin	5.55	5.55
Gemifloxacin	6.00	6.00

Grepafloxacin	5.32	5.32
Levofloxacin	4.77	4.77
Lomefloxacin	5.32	5.32
Moxifloxacin	5.55	5.55
Nadifloxacin	4.40	4.53
Norfloxacin	5.32	5.32
Pefloxacin	4.60	4.60
Rufloxacin	4.57	5.20
Sitafloxacin	5.67	5.67
Sparfloxacin	5.97	5.97
Temafloxacin	5.01	5.21
Trovafloxacin	5.98	5.99
Azithromycin	4.75	4.01
Aztreonam	4.97	3.80
Fosfomycin	3.92	4.16
Faropenem	4.51	4.68
Doripenem	6.00	6.00
Amoxicillin	5.66	5.66
Ampicillin	5.50	5.66
Carbenicillin	5.00	5.00
Mezlocillin	5.00	4.57
Ticarcillin	5.00	5.00
Azlocillin	5.00	5.00
Piperacillin	5.00	4.88
Sulfabenzamide	4.24	4.77
Sulfacetamide	4.40	4.77
Sulfachlorpyridazine	4.90	5.15
Sulfadiazine	4.99	5.24
Sulfadimethoxine	5.00	5.16
Sulfaguanidine	5.00	5.00
Sulfamerazine	4.99	5.24
Sulfameter	5.24	5.24
Sulfamethazine	4.99	5.10
Sulfamethizole	4.92	5.16
Sulfamethoxazole	4.54	5.03
Sulfamethoxypyridazine	5.21	5.21
Sulfamonomethoxine	5.36	5.42
Sulfanitran	4.41	4.08
Sulfaphenazole	4.30	4.86
Sulfapyridine	4.35	4.75
Sulfaquinoxaline	4.69	5.22

Sulfathioazole	4.40	5.06
Sulfisoxazole	4.51	5.00
Demeclocycline	5.00	4.67
Doxycycline	5.07	4.73
Meclocycline	5.00	4.67
Methacycline	5.03	4.69
Minocycline	5.69	5.69
Oxytetracycline	4.63	4.29
PTK-0796	6.00	5.67
Tetracycline	5.03	4.70
Tigecycline	5.52	4.94
Chlortetracycline	5.00	4.67
Average	5.08	4.97

[A]Physicochemical properties calculated by ChemAxon, ChemDraw

[b]Physicochemical properties calculated by Pipeline Pilot, ACD/Labs

c) PEMPO and MPO scoring for selected confirmed hit compounds:

Physicochemical properties were calculated by ChemAxon, ChemDraw.

SMILES	Compound ID	Cluster	MPO Score	PEMPO Score
n1(CC(=O)Nc2onc(c3cccc(Cl)c3)c2)ncc(COc(cccc4)c45)c15	KP10	1	4.489	2.794
n(CC(=O)NCC1COc(c2O1)cccc2)(c(c3ccc(cc3)C)cc4C(OCC)=O)c4C	KP11	1	3.544	2.774
N(C(C)C(=O)Nc(cc1)ccc1OCC)(N=Nc2c3c4c(s2)CCCC4)C3=O	KP16	2	3.492	2.547
c1(C(c(cccn2)c2)N([H])c3nc(C)cc(C)n3)c(C)c(sc1NC(c4ccccc4)=O)C	KP26	2	2.946	2.911
c1(cc(sc1NC(c2ccccc2)=O)C)C(c3ccccn3)N([H])c4ccccn4	KP27	2	3.228	1.870
c1(C(c2cccc(OC)c2)N([H])c3nc(C)cc(C)n3)c4c(sc1NC(c5ccccc5)=O)CCCC4	KP31	2	2.524	2.860
c12c(NC(=O)NC1=O)[nH]c(c3ccc(c4c3)cccc4)c2C5c6c(NC5=O)cccc6	KP44	2	2.950	3.037
N1(CCc2onc(c(ccc(c34)OCO3)c4)n2)C(=O)Nc(c5C1=O)cc(c6c5)OCO6	KP52	3	3.501	3.626
S(=O)(=O)(N(CC1)CCC1C(=O)N2CCc(c3C2)cccc3)c4c(C)noc4\C=C/c(cc5)ccc5OC	KP7	3	4.302	2.583
n1(nc(c(C([H])([H])c2ccccc2)c1O)C)c(nc(c3C(=O)N4CCc(c5C4)cccc5)C)s3	KP37	5	3.618	1.607
n1(nc(c(C([H])([H])c2ccccc2)c1O)C)c(nc(c3C(=O)Nc4cccc(Cl)c4)C)s3	KP40	5	2.967	2.198
S(=O)(=O)(c1c(C)cc(c(C)c1)OCC)N2CCc(cc(c3n4)ccc(OC)c3)c24	KP34	7	3.870	1.274
S(=O)(=O)(c(ccc1c2OC(N1CC(=O)NCc(cc3)ccc3OC)=O)c2)N4CCC(CC4)C	KP9	7	4.454	4.424
c1(C(c2ccccc2)NC(COc(cc3)ccc3C)=O)nnc(c4ccccc4)o1	KP6	9	4.021	2.480
o1c(c(ccc2c3c(C)c([nH]2)C)c3)nnc1SC(C)C(=O)Nc(ccc(c45)OCCO4)c5	KP18	10	3.237	4.262
c1(nnc(SCC(=O)NC(C)c2[nH]c(c3n2)cccc3)o1)c4cc(c5n4C)cccc5	KP19	10	4.208	4.750
c1(Cc2ccccc2)nc(c3n1CC(O)COc(cc4)ccc4F)cccc3	KP29	11	3.850	1.431
c1(C2)n(CCN2Cc3c(OC)ccc(c3)NC(=O)c4ccc(cc4)OC)c5c(cccc5)n1	KP33	11	3.853	2.151
n1(CC2CC)c(c(O2)ccc3Cl)c3cc1C(=O)N4CCC(CC4)C(OCC)=O	KP50	11	4.879	1.347
n1(CC2CC)c(c(O2)ccc3Cl)c3cc1C(NCCCOC(C)C)=O	KP51	11	4.933	2.834
n1(Cc(cc2)ccc2OC)c3c(nc1CCC(=O)Nc(ccc(c4C)C)c4)cccn3	KP8	11	3.574	2.644
c(nc(C)cc1NCc(cccn2)c2)(c3c4ccc(cc4)F)n1nc3C(F)(F)F	KP32	12	4.058	1.700

c(N(CCOc1ccccc1)S(=O)(=O)c2ccc(cc2)C)(nn(c34)c(C)cc(C)n3)n4	KP42	12	3.915	2.199
N1(Cc(cc2)ccc2C(NCCc(cc3)ccc3C)=O)c4c(cccc4)N(C(=O)C1=O)CC=C	KP13	14	3.393	2.528
N(CC(=O)Nc1cccc(C)c1C)(c2c3cccc2)C(=O)C=C3C(=O)NCc4ccccc4	KP43	15	3.435	3.025
N1(C)C(=O)CSc(ccc(c2)NC(=O)Nc3cc(C)ccc3C)c12	KP14	16	4.735	2.215
c12c(ncnc1N(C)C)n(cc2c3ccccc3)c4ccc(cc4)F	KP28	17	3.752	0.554
n1(c2ccccc2Cl)cc(c3c1ncnc3NC4CC4)c5ccccc5	KP30	17	3.759	1.000
n1c(SCc2ccccc2)nc(cc1N(CC3)CCC3C(N)=O)c4ccc(cc4)C	KP45	18	3.350	1.500
c1(scc(c2ccccc2)n1)NC(=O)Nc(ccc(c3Cl)F)c3	KP21	20	3.500	1.333
n1c(c(cc2)ccc2NC(=O)c3ccccc3C)onc1c4ccccc4	KP22	21	3.750	2.201
n1c(c(cccc2NC(=O)C3CC3)c2)onc1c4ccccc4	KP23	21	4.127	2.569
n1c(c2ccc(cc2)Cl)onc1N([H])C(COc(cc3)ccc3OC)=O	KP35	22	4.739	2.700
C1(NC(=O)CC2c3cc(F)cc(F)c3)=C2C(NC(SCc(cc4)ccc4F)=N1)=O	KP17	23	3.845	2.330
N1=C(SC([H])([H])c2ccccc2)C3=C(N(CCc4ccccc4)C1=O)CCC3	KP36	23	3.668	0.369
N1(N(C(C)C)C=N2)C2=NC(CSc(cc3)ccc3NC(=O)Nc4cccc(Cl)c4)=CC1=O	KP39	24	3.315	3.980
c1(nnc(NC(CSc(cc2)ccc2Cl)=O)s1)S(=O)(=O)N3CCC(CC3)C	KP49	25	4.788	3.249
n1(nc(s2)COc3ccccc3)c2nnc1CSc4ccccc4	KP47	26	4.923	1.361
n12c(nnc1c3ccccc3Cl)sc(c4ccc(c5n4)cccc5)n2	KP48	27	3.988	0.667
[nH](c(ccc(c1)CNC(=O)Nc(cc2)ccc2Cl)c1c3C)c3C	KP15	28	3.629	2.017
C(=O)(c1cccc(c1)Nc2cnc(c3n2)cccc3)Nc(cc4)ccc4CC	KP5	28	3.455	2.840
c1(NC(=O)Nc(cc2)ccc2OC)sc(c3n1)cc(c(C)c3)C	KP20	29	3.594	1.673

4. Primers used in construction of carbapenemase-expressing *E. coli* strains:

Primer Name	Sequence (5'-3')
NDM-1R	GTTGGCGGGTGTCGGGGCTGGCTTAATCAGCGCAGCTTGTCGGCC
NDM-1F	GATAACAATTTCACACAGGAAACAGCTATGGAATTGCCCAATATTATGCAC
KPC-2and3F	GATAACAATTTCACACAGGAAACAGCTATGTCACTGTATCGCCGTCTAGT
KPC-2and3R	GTTGGCGGGTGTCGGGGCTGGCTTAACTTACTGCCCGTTGACGCCC
pUC18R	TTAAGCCAGCCCCGACACCCGCC
pUC18F	AGCTGTTTCCTGTGTGAAATTGTTATCCGCTCAC

5. Activity of identified compounds against *Escherichia coli strains* harboring various carbapenemase enzymes

b) DH5α

Carbapenemase	Measurement	KP9	KP11	KP19	KP40	KP56	Meropenem
None	**MIC (μg/mL)**	>128	>128	>128	>128	>128	0.13
	FIC	>1	>1	>1	>1	>1	N/A
KPC2	**MIC**	>128	>128	>128	>128	>128	2
	FIC	>1	>1	>1	>1	>1	N/A
KPC3	**MIC**	>128	>128	>128	>128	>128	2
	FIC	>1	>1	>1	>1	>1	N/A
NDM1	**MIC**	>128	>128	>128	>128	>128	16
	FIC	>1	>1	>1	>1	>1	N/A

b) *tolC* mutant

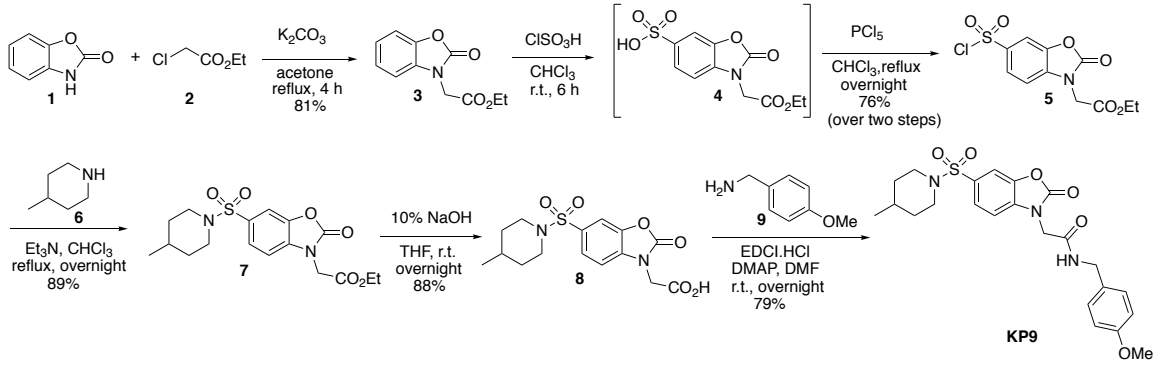
Carbapenemase	Measurement	KP9	KP11	KP19	KP40	KP56	Meropenem
None	**MIC (μg/mL)**	>128	>128	>128	16-32	>128	0.063
	FIC	>1	>1	>1	>1	>1	N/A
KPC2	**MIC**	>128	>128	>128	32-128	>128	2
	FIC	>1	>1	>1	0.25	>1	N/A
KPC3	**MIC**	>128	>128	>128	64-128	>128	4
	FIC	>1	>1	>1	0.25	>1	N/A
NDM1	**MIC**	>128	>128	>128	32-128	>128	>16
	FIC	>1	>1	>1	>1	>1	N/A

c) *lptD* mutant

Carbapenemase	Measurement	KP9	KP11	KP19	KP40	KP56	Meropenem
None	**MIC (μg/mL)**	>128	>128	>128	>128	>128	0.03
	FIC	>1	>1	>1	>1	>1	N/A
KPC2	**MIC**	>128	>128	>128	>128	>128	1
	FIC	>1	>1	>1	>1	>1	N/A
KPC3	**MIC**	>128	>128	>128	>128	>128	0.5
	FIC	>1	>1	>1	>1	>1	N/A
NDM1	**MIC**	>128	>128	>128	>128	>128	8
	FIC	>1	>1	>1	>1	>1	N/A

6. Synthesis:

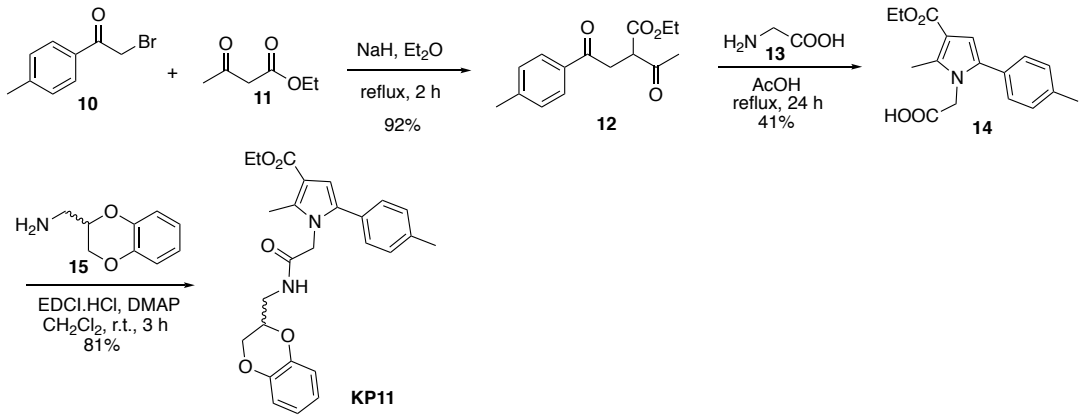
a) Synthesis of **KP9** was accomplished using sulfonylation and coupling reactions as the keys steps from commercially available starting materials (Scheme 1).



Scheme 1: Synthesis of **KP9**

N-Alkylation of benzoxazolinone **1** with ethyl chloroacetate **2** followed by sulfonylation of the resultant *N*-alkylated compound **3** gave corresponding sulfonic acid derivative **4**. Compound **4** was converted into chlorosulfonyl **5** by treatment with PCl_5. Coupling of **5** with 4-methylpiperidine **6** afforded sulfonamide **7**, which upon hydrolysis and subsequent coupling with 4-methoxy benzylamine **9** using EDCI/DMAP yielded target compound **KP9**.

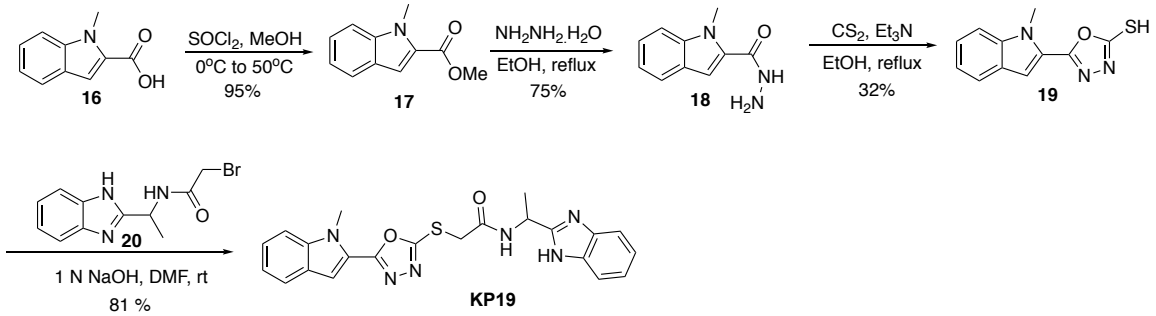
b) Synthesis of **KP11** was achieved *via* alkylation/condensation/coupling approaches from readily available starting materials (Scheme 2).



Scheme 2: Synthesis of **KP11**

α-Alkylation of ethyl acetoacetate **11** with 2-bromo-4'-methyl-acetophenone **10** *via* enolate afforded compound **12**. Condensation of **12** with glycine **13** followed by intramolecular cyclization-dehydration-aromatization in one-pot provided pyrrole derivative **14**. Finally, coupling of **14** with 2-aminomethyl-1,4-benzodioxane **15** using EDCI/DMAP gave desired compound **KP11**.

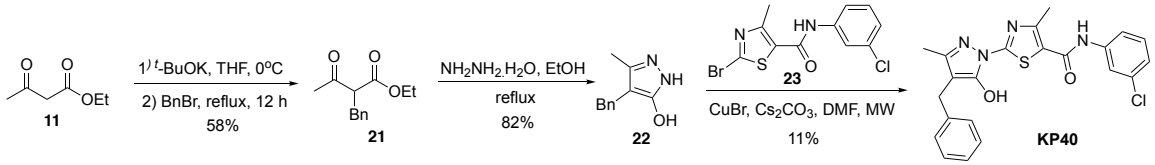
c) **KP19** was synthesized by utilizing condensation and coupling reactions from easily prepared starting materials (Scheme 3).



Scheme 3: Synthesis of **KP19**

Esterification of 1-methylindole-2-carboxylic acid **16** followed by condensation of the resultant ester **17** with hydrazine hydrate afforded compound **18**. Heating the mixture of **18** and carbon disulfide in ethanol provided 1,3,4-oxadiazole thiol derivative **19** *via* condensation-intramolecular cyclization-aromatization cascade reactions. Base-catalyzed coupling of **19** with α-bromo-amido compound **20** gave **KP19**.

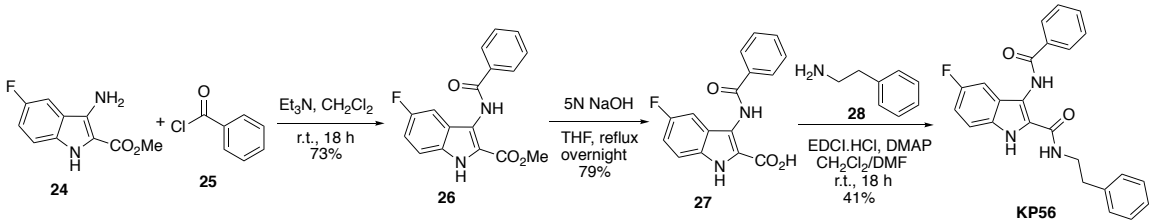
d) Synthesis of **KP40** was accomplished through benzylation/condensation/coupling approaches starting from ethyl acetoacetate (Scheme 4).



Scheme 4: Synthesis of **KP40**

α-Benzylation of ethyl acetoacetate **11** followed by condensation of the corresponding benzylated compound **21** with hydrazine hydrate gave hydroxyl-pyrazole derivative **22** *via* condensation-intramolecular cyclization-aromatization cascade reactions. Cu(I)-catalyzed coupling of **22** with bromo-thiazole **23** under microwave irradiation provided **KP40**.

e) **KP56** was prepared by simple coupling reactions from commercially available starting materials (Scheme 5).



Scheme 5: Synthesis of **KP56**

Coupling of methyl-3-amino-5-fluoro-1*H*-indole-2-carboxylate **24** with benzoyl chloride **25** gave corresponding amide **26**. Base-catalyzed hydrolysis of **26** and subsequent coupling of the resultant acid **27** with 2-phenethylamine **28** using EDCI/DMAP afforded **KP56**.

6. **Experimental Details and Compound Characterization:**

Ethyl 2-(2-oxobenzo[*d*]oxazol-3(2*H*)-yl)acetate 3:[1] To a solution of 2-benzoxazolinone **1** (1.10 g, 8.16 mmol) in acetone (10 mL) was added K_2CO_3 (2.26 g, 16.32 mmol) and ethyl chloroacetate **2** (1.3 mL, 12.24 mmol). The resulting mixture was refluxed for 4 hours. After cooling, reaction mixture was filtered, diluted with EtOAc and washed with H_2O. The organic layer was dried over anhydrous Na_2SO_4 and concentrated. Crude product was purified by flash chromatography (EtOAc:hexanes = 1:3 to 1:2). Yield: 1.70 g (94%). R_f = 0.39 in EtOAc : hexanes = 1:3. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.24-7.11 (m, 3H), 6.90 (d, *J* = 8.0 Hz, 1H), 4.56 (s, 2H), 4.26 (q, *J* = 8.0 Hz, 2H), 1.28 (t, *J* = 8.0 Hz, 3H). ESI-MS: $[M+H]^+$ 222.1; found 222.1

Ethyl 2-(6-(chlorosulfonyl)-2-oxobenzo[*d*]oxazol-3(2*H*)-yl)acetate 5:[2] Solution of ethyl 2-(2-oxobenzo[*d*]oxazol-3(2*H*)-yl)acetate **3** (1.00 g, 4.52 mmol) in $CHCl_3$ (10 mL) was cooled in ice-water bath, chlorosulfonic acid (361 µL, 5.42 mmol) was added dropwise at 0 °C and stirred at room temperature for 6 hours. To this was then added PCl_5 (1.13 g, 5.44 mmol) at 0 °C and refluxed overnight. After cooling to room temperature, reaction mixture was diluted with CH_2Cl_2 and washed with brine. The organic layer was dried over anhydrous Na_2SO_4 and concentrated. Crude product obtained was washed with hexanes and used for the next reaction without further purification. Yield: 1.14 g (86%). R_f = 0.5 in EtOAc:hexanes = 1:1. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.97 (d, *J* = 8.0 Hz, 1H), 7.91 (s, 1H), 7.09 (d, *J* = 8.0 Hz, 1H), 4.64 (s, 2H), 4.28 ((q, *J* = 8.0 Hz, 2H), 1.32 (t, *J* = 8.0 Hz, 3H).

Ethyl 2-(6-((4-methylpiperidine-1-yl)sulfonyl)-2-oxobenzo[*d*]oxazol-3(2*H*)-yl)acetate 7: To a solution of ethyl 2-(6-(chlorosulfonyl)-2-oxobenzo[*d*]oxazol-3(2*H*)-yl)acetate **5** (1.10 g, 3.78 mmol) and 4-methylpiperidine **6** (537 µL, 4.54 mmol) in $CHCl_3$ (10 mL) was added Et_3N (1.6 mL, 11.34 mmol) and refluxed overnight. After cooling to room temperature, reaction mixture was diluted with CH_2Cl_2, washed with saturated NH_4Cl and

brine. The organic layer was dried over anhydrous Na_2SO_4 and concentrated. Crude product was purified by flash chromatography (EtOAc:hexanes = 1:2 to 1:1). Yield: 1.24 g (89%). R_f = 0.41 in EtOAc : hexanes = 1:1. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.65 (d, *J* = 8.0 Hz, 1H), 7.63 (s, 1H), 7.02 (d, *J* = 8.0 Hz, 2H), 4.61 (s, 2H), 4.29 (q, *J* = 8.0 Hz, 2H), 3.76 (d, *J* = 12.0 Hz, 2H), 2.26 (t, *J* = 12.0 Hz, 2H), 1.69 (d, *J* = 8.0 Hz, 2H), 1.33-0.91 (m, 5H), 0.93 (d, *J* = 8.0 Hz, 3H).

Ethyl 2-(6-((4-methylpiperidine-1-yl)sulfonyl)-2-oxobenzo[*d*]oxazol-3(2*H*)-yl)acetic acid 8: To a solution of ethyl 2-(6-((4-methylpiperidine-1-yl)sulfonyl)-2-oxobenzo[*d*]oxazol-3(2*H*)-yl)acetate **7** (0.60 g, 1.64 mmol) in THF (10 mL) was added 10% aq. NaOH solution (5 mL) and stirred at room temperature overnight. Reaction mixture was diluted with H_2O and washed with Et_2O. The aqueous layer was acidified (pH between 1-2) with 1:1 HCl (aq.) at 0 °C. Solid that comes out was filtered and dried to give the desired product. Yield: 0.51 g (88%). ^{1}H NMR (DMSO-d_6, 400 MHz) δ 10.07 (s, 1H, -OH), 7.02 (d, *J* = 8.0 Hz, 1H), 6.96 (s, 1H), 6.48 (d, *J* = 8.0 Hz, 1H), 3.90 (s, 2H), 3.49 (d, *J* = 12.0 Hz, 2H), 2.14-2.07 (m, 2H), 1.63 (d, *J* = 8.0 Hz, 2H), 1.26 (m, 1H), 1.15-1.05 (m, 2H), 0.84 (d, *J* = 8.0 Hz, 3H).

KP9: To a solution of acid **8** (0.17 g, 0.49 mmol) in DMF (5 mL) was added 4-methoxybenzylamine **9** (70 µL, 0.54 mmol), DMAP (6 mg, 0.05 mmol) and EDCI·HCl (0.19, 1.03 mmol) and stirred at room temperature overnight. Reaction mixture was diluted with EtOAc, washed with saturated NH_4Cl, H_2O and brine. The organic layer was dried over anhydrous Na_2SO_4 and concentrated. Crude product was purified by flash chromatography (EtOAc:hexanes = 1:1 to 1:0). Yield 0.18 g (79%). R_f = 0.38 in EtOAc. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.20 (d, *J* = 8.0 Hz, 1H), 7.18 (s, 1H), 7.14 (d, *J* = 8.0 Hz, 2H), 6.82 (d, *J* = 8.0 Hz, 2H), 6.52 (d, *J* = 8.0 Hz, 1H), 5.16 (br t, 1H, -NH), 4.42 (d, *J* = 8.0 Hz, 2H), 3.92 (d, *J* = 8.0 Hz, 2H), 3.77 (s, 3H), 3.64 (d, *J* = 8.0 Hz, 2H), 2.22 (t, *J* = 12 Hz, 2H), 1.65 (d, *J* = 12.0 Hz, 2H), 1.26-1.25 (m, 3H), 0.90 (d, *J* = 8.0 Hz, 3H). ESI-MS: $[M+K-SO_2]^+$ 448.2; found 448.2.

Ethyl 2-acetyl-4-oxo-4-(*p*-tolyl)butanoate 12:[3] Ethyl acetoacetate **11** (715 µL, 5.63 mmol) was added dropwise to a stirred suspension of NaH (0.23 g, 5.63 mmol) in Et_2O (10 mL) at room temperature under Ar atmosphere. After 10 min, a solution of 2-bromo-4'-methylacetophenone **10** (1.00 g, 4.69 mmol) in Et_2O (5 mL) was added dropwise and the resulting mixture was refluxed for 2 hours. After cooling, reaction mixture was filtered, washed twice with Et_2O and the filtrate was concentrated. Crude product was purified by flash chromatography (EtOAc : hexanes = 1:4 to 1:2). Yield: 1.13 g (92%). R_f = 0.38 in EtOAc:hexanes = 1:4. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.86 (d, *J* = 8.0 Hz, 2H), 7.25 (d, *J* = 8.0 Hz, 2H), 4.21 (q, *J* = 7.2 Hz, 2H), 4.18 (dd, *J* = 8.8 Hz, 5.2 Hz, 1H), 3.67 (dd, *J* = 8.8 Hz, 5.2 Hz, 1H), 3.51 (dd, *J* = 8.8 Hz, 5.2 Hz, 1H), 2.43 (s, 3H), 2.40 (s, 3H), 1.28 (t, *J* = 7.2 Hz, 3H). ESI-MS: $[M+H]^+$ 263.1; found 263.1.

2-(3-(Ethoxycarbonyl)-2-methyl-5-(*p*-tolyl)-1*H*-pyrrol-1-yl)acetic acid 14:[4] To a solution of ethyl 2-acetyl-4-oxo-4-(*p*-tolyl)butanoate **12** (0.90 g, 3.43 mmol) in AcOH (10 mL) was added glycine **13** (0.28 g, 3.78 mmol) and refluxed for 24 hours. After cooling to room temperature, reaction mixture was diluted with Et_2O and extracted with sat. Na_2CO_3. Aqueous layer was acidified (pH between 1-2) with 1:1 HCl (aq.) at 0 °C. Solid that comes out was filtered and dried to give the desired product. Yield: 0.418 g (40%). ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.22-7.17 (m, 4H), 6.58 (s, 1H), 4.62 (s, 2H), 4.27 (q, *J* = 8.0 Hz, 2H), 2.55 (s, 3H), 2.39 (s, 3H), 1.34 (t, *J* = 8.0 Hz, 3H).

KP11: To a solution of 2-(3-(ethoxycarbonyl)-2-methyl-5-(*p*-tolyl)-1*H*-pyrrol-1-yl)acetic acid **14** (0.15 g, 0.5 mmol) in CH_2Cl_2 (5 mL) was added 2-aminomethyl-1,4-benzodioxane **15** (79 µL, 0.55 mmol), DMAP (6 mg, 0.05 mmol) and EDCI·HCl (0.20 g, 1.05 mmol) and stirred at room temperature overnight. Reaction mixture was diluted with CH_2Cl_2, washed with saturated NH_4Cl, H_2O and brine. The organic layer was dried over anhydrous Na_2SO_4 and concentrated. Crude product was purified by flash chromatography

(EtOAc:hexanes = 1:2 to 1:1). Yield: 0.18 g (81%). R_f = 0.5 in EtOAc:hexanes = 1:1. ^{1}H NMR= ($CDCl_3$, 400 MHz) δ 7.16 (d, *J* = 8.0 Hz, 2H), 7.04 (d, *J* = 4.0 Hz, 2H), 6.87-6.85 (m, 4H), 6.64 (s, 1H), 5.72 (t, *J* = 8.0 Hz, 1H, -NHCO), 4.58 (d, *J* = 8.0 Hz, 2H), 4.32 (d, *J* = 8.0 Hz, 2H), 4.20-4.14 (m, 2H), 3.68-3.58 (m, 2H), 3.45-3.39 (m, 1H), 2.56 (s, 3H), 2.31 (s, 3H), 1.37 (t, *J* = 8 Hz, 3H). ESI-MS: $[M+H]^+$ 449.2; found 449.1.

Methyl 1-methyl-1*H*-indole-2-carboxylate 17: Thionyl chloride (0.29 mL, 4 mmol) was added dropwise over a period of 10 min to the solution of 1-methyl-1*H*-indole-2-carboxylic acid **16** (350 mg, 2 mmol) in MeOH (4 mL) at 0 °C. The reaction mixture was stirred at 50 °C overnight. After adding water (2.5 mL), immediately solid comes out. Solid was collected by filtration and washed several times with MeOH to give the desired compound. Yield: 179 mg (95%). R_f = 0.75 in EtOAc:hexanes = 1:5. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.67 (d, *J* = 8.0 Hz, 1H), 7.37 (m, 2H), 7.29 (s, 1H), 7.15 (t, 8.0 Hz, 1H), 4.08 (s, 3H), 3.91 (s, 3H). ESI-MS: $[M+H]^+$ 190.1; found 190.1.

1-Methyl-1*H*-indole-2-carbohydrazide 18: Hydrazine Hydrate (0.49 mL, 10 mmol) was added to a solution of methyl 1-methyl-1*H*-indole-2-carboxylate **17** (189 mg, 1 mmol) in EtOH (5 mL), and the reaction mixture was refluxed overnight. Reaction mixture was allowed to cool to room temperature. Solid comes out was collected by filtration, washed with EtOH and dried to give the desired compound. Yield: 141 mg (75%). ^{1}H NMR (DMSO-d_6, 400 MHz) δ 9. 75 (s, 1H), 7.59 (d, *J* = 8.8 Hz, 1H), 7.50 (d, *J* = 8.0 Hz, 1H), 7.25 (t, *J* = 8.8 Hz, 2H), 7.07 (t, *J* = 8.0 Hz, 1H), 6.98 (s, 1H), 4. 48 (s, 2H), 3.96 (s, 3H). ESI-MS: $[M+H]^+$ 190.1; found 190.1.

5-(1-Methyl-1*H*-indol-2-yl)-1,3,4-oxadiazole-2-thiol 19: Carbon disulfide (0.39 mL, 6.6 mmol) and NEt_3 (0. 469 mL, 3.3 mmol) were added to a solution of methyl 1-methyl-1*H*-indole-2-carbohydrazide **18** (567 mg, 3 mmol) in EtOH (5 mL), and the reaction mixture was refluxed overnight. Reaction was allowed to

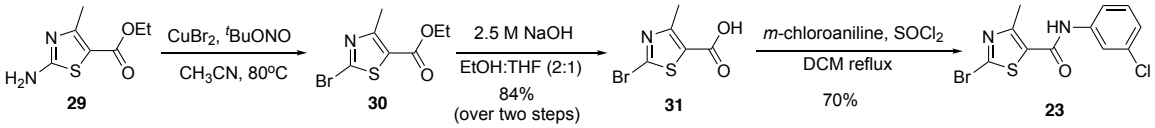
cool to room temperature. Solid comes out was collected by filtration, washed with EtOH and dried to give the desired compound. Yield: 221 mg (32%). ^{1}H NMR ($CDCl_3$, 400 MHz) δ 11. 10 (br s, 1H), 7.70 (d, *J* = 8.0 Hz, 1H), 7.40-7.39 (m, 2H), 7.29-7.26 (m, 1H), 7.21-7.17 (m, 1H), 4. 06 (s, 3H). ESI-MS: $[M+H]^+$ 232.1; found 232.1.

KP19: To a solution of 5-(1-methyl-1*H*-indol-2-yl)-1,3,4-oxadiazole-2-thiol **19** (36 mg, 0.13 mmol) in DMF (1 mL) was added 1N NaOH (0.11 mL, 0.11 mmol) and *N*-(1-(1*H*-benzo[*d*]imidazol-2-yl)ethyl)-2-bromoacetamide **20**[5,6] (25 mg, 0.11 mmol) at room temperature. The reaction mixture was stirred at room temperature overnight and extracted with EtOAc (3 times). Combined organic layers were dried over anhydrous Na_2SO_4 and concentrated to give a pure product. Yield: 42 mg (81%). R_f = 0.2 in EtOAc:hexanes = 1:1. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.67 (d, *J* = 8.0 Hz, 1H), 7.62 (br s, 2H), 7.42-7.38 (m, 2H), 7.22-7.18 (m, 4H), 5.42 (q, *J* = 7.2 Hz, 1H), 4.18 (s, 3H), 3.197 (s, 2H), 1.82 (d, *J* = 6.8 Hz, 3H). ESI-MS: $[M+H]^+$ 433.1; found 433.1.

Ethyl 2-benzyl-3-oxobutanoate 21:[7] To a suspension of potassium *tert*-butoxide (5.4 g, 48 mmol) in THF (100 mL) was added ethyl acetoacetate **11** (5.66 mL, 44 mmol) slowly with *tert*-butanol (0.42 mL, 4.4 mmol) at 0 °C. During the time, the reaction solution was clear and then benzyl bromide (5 mL, 42 mmol) was added. The reaction mixture was stirred at 70 °C overnight. The reaction mixture was quenched with water (0.13 mL / mmol) and then sat. $NaHCO_3$ was added and organic layer was extracted with ether (3 times). Combined organic layers were dried over anhydrous Na_2SO_4 and concentrated. Crude product was purified by flash chromatography (ether:hexanes = 1:19). Yield: 5.61g (58%). ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.28-7.16 (m, 5H), 4.14 (q, *J* = 7.2 Hz, 2H), 3.77 (t, *J* = 8.0 Hz, 1H), 3.15 (d, *J* = 8.0 Hz, 2H), 2. 17 (s, 3H), 1.19 (t, *J* = 7.2 Hz, 3H).

4-Benzyl-3-methyl-1*H*-pyrazol-5-ol 22: To a solution of hydrazine (1.94 mL, 40 mmol) in EtOH (9 mL) was added ethyl 2-benzyl-3-oxobutanoate **21** (2.11g, 9.6 mmol) and the reaction mixture was refluxed for 2 hours. The reaction was allowed to cool to room temperature. Solid comes out was collected by filtration and dried to give the desired product. Yield: 1.48 g (82%). ^{1}H NMR (DMSO-d_6, 400 MHz) δ 7.23-7.11 (m, 5H), 3.52 (s, 2H), 1.98 (s, 3H). ESI-MS: $[M+H]^+$ 189.1; found 189.1.

Synthesis of 2-Bromo-*N*-(3-chlorophenyl)-4-methylthiazole-5-carboxamide 23:



Ethyl 2-bromo-4-methylthiazole-5-carboxylate 30: Ethyl 2-amino-4-methylthiazole-5-carboxylate **29**[7,8] (2.2 g, 12 mmol) in CH_3CN (25 mL) was added into a two-neck round bottom flask, flushed with Ar atmosphere and stirred at 60 °C. In parallel, $CuBr_2$ (2.8 g, 21.6 mmol) and *tert*-BuONO (2.6 mL, 20 mmol) in CH_3CN (20 mL) was prepared under argon atmosphere. The resulting solution was added slowly to the reaction mixture, and kept at 80 °C for 1 hour. Afterwards, the reaction mixture was evaporated and the residue was dissolved in CH_2Cl_2, washed twice with a 1M HCl, and then washed with 1M NH_4Cl. Organic layer was dried over anhydrous Na_2SO_4 and concentrated. Crude product was used for the next reaction without further purification. Yield: 2.0 g (67%). ^{1}H NMR ($CDCl_3$, 400 MHz) δ 4.32 (q, J = 6.8 Hz, 2H), 2.70 (s, 3H), 1.35 (t, J = 7.2 Hz, 3H). ESI-MS: $[M+H]^+$ 249.9; found 249.9.

2-Bromo-4-methylthiazole-5-carboxylic acid 31: To a solution of ethyl 2-bromo-4-methylthiazole-5-carboxylate **30** (500 mg, 2 mmol) in EtOH:THF (2:1, 12 mL) was added 2.5M NaOH (5 mL) solution and the reaction mixture was stirred at 50 °C overnight. After cooling, the reaction mixture was acidified and extracted with ethyl acetate. Organic layer was dried over anhydrous Na_2SO_4

and concentrated. Crude product was used for the next reaction without further purification. Yield: quant. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 2.73 (s, 3H).

2-Bromo-*N*-(3-chlorophenyl)-4-methylthiazole-5-carboxamide 23: 2-Bromo-4-methylthiazole-5-carboxylic acid **30** (222 mg, 1 mmol) in thionyl chloride (5 mL) was refluxed for 1 h under argon atmosphere. After completion of reaction (monitored by TLC), thionyl chloride was evaporated, then the solution of 3-chloroaniline in CH_2Cl_2:pyridine (2:1, 9 mL) was added into the reaction mixture and stirred overnight. The reaction mixture was diluted with EtOAc and washed with 1M HCl. Organic layer was dried over anhydrous Na_2SO_4 and concentrated. Crude product was used for the next reaction without further purification. Yield: 230 mg (70%) ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.65 (s, 1H), 7.45 (br s, 1H), 7.37 (d, *J* = 8.0 Hz, 1H), 7.26-30 (m, 1H), 7.15 (d, *J* = 8.0 Hz, 1H), 2.69 (s, 3H). ESI-MS: $[M+H]^+$ 330.9; found 330.9.

KP40: 4-Benzyl-3-methyl-1*H*-pyrazol-5-ol **22** (112 mg, 0.6 mmol), 2-bromo-*N*-(3-chlorophenyl)-4-methylthiazole-5-carboxamide **23** (190 mg, 0.6 mmol), CuBr (0.1 mmol), and DMF (2 mL) was added to a 15 mL vial. The vial was sealed with a septum and placed in Anton Paar Monowave 300. After irradiation at 170 °C for 90 min and subsequent cooling, the reaction mixture was diluted with sat. NH_4Cl and extracted with ethyl acetate. Organic layer was dried over anhydrous Na_2SO_4 and concentrated. Crude product was purified by flash chromatography (EtOAc:hexanes = 1:2 to 1:1). Yield: 30 mg (11 %). R_f = 0.3 in EtOAc : hexanes = 1:2. ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.36-7.7 (m, 3H), 7.09-7.23 (m, 6H), 3.76 (s, 2H), 2.61 (s, 3H), 2.17 (s, 3H). ESI-MS: $[M+H]^+$ 439.1; found 439.1.

Methyl 3-benzamido-5-fluoro-1*H*-indole-2-carboxylate 26: To a solution of methyl 3-amino-5-fluoro-1*H*-indole-2-carboxylate **24** (0.16 g, 0.77 mmol) in CH_2Cl_2 (5 mL) was added Et_3N (117 µL, 0.85 mmol) and benzoyl chloride **25** (97 µL, 0.85 mmol). The resulting mixture was stirred at room

temperature for 18 hours. The reaction mixture was diluted with CH_2Cl_2, washed with 1N aq. HCl and H_2O. The organic layer was dried over anhydrous Na_2SO_4 and concentrated. Recrystallization (EtOAc) yielded the desired product. Yield: 0.18 g (73%). R_f = 0.35 in EtOAc:hexanes = 1:1. ^{1}H NMR (DMSO-d_6, 400 MHz) δ 11.87 (s, 1H, -NH), 10.02 (s, 1H, -NHCO), 7.92-7.90 (m, 2H), 7.49-7.44 (m, 3H), 7.34-7.32 (m, 2H), 7.09-7.06 (m, 1H), 3.74 (s, 3H).

3-Benzamido-5-fluoro-1*H*-indole-2-carboxylic acid 27: To a solution of methyl 3-benzamido-5-fluoro-1*H*-indole-2-carboxylate **26** (0.14 g, 0.45 mmol) in THF (5 mL) was added 5N NaOH (3 mL) and refluxed overnight. The reaction mixture was acidified (pH between 5-6) using 1:1 HCl (aq.) and extracted with EtOAc. The organic layer was dried over anhydrous Na_2SO_4 and concentrated to give the desired product. Yield: 0.11 g (79%). ^{1}H NMR (DMSO-d_6, 400 MHz) δ 11.82 (s, 1H, -NH), 10.13 (s, 1H, -NHCO), 8.02-8.00 (m, 2H), 7.60-7.40 (m, 5H), 7.15-7.14 (m, 1H).

KP56: To a solution of 3-benzamido-5-fluoro-1*H*-indole-2-carboxylic acid **27** (90 mg, 0.31 mmol) in CH_2Cl_2:DMF (4:1, 5 mL) was added 2-phenylethan-1-amine **28** (44 µL, 0.34 mmol), DMAP (4 mg, 0.03 mmol) and EDCI·HCl (0.13, 0.65 mmol) and stirred at room temperature overnight. Reaction mixture was diluted with CH_2Cl_2, washed with sat. NH_4Cl, H_2O and brine. The organic layer was dried over anhydrous Na_2SO_4 and concentrated. Crude product was purified by flash chromatography (EtOAc:hexanes = 1:4 to 1:1). Yield: 51 mg (41%). R_f = 0.52 in EtOAc:hexanes = 1:1. ^{1}H NMR (DMSO-d_6, 400 MHz) δ 11.64 (s, 1H, -NH), 10.60 (s, 1H, -NHCO), 8.17-8.15 (m, 1H), 8.06-8.04 (m, 2H), 7.66-7.59 (m, 3H), 7.51-7.45 (m, 2H), 7.24-7.23 (m, 4H), 7.16-7.14 (m, 1H), 3.57 (t, *J* = 5.6 Hz, 2H), 2.84 (t, *J* = 5.6 Hz, 2H). ESI-MS: $[M+H]^+$ 402.1; found 402.1.

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