General Experimental Methods

Organic Synthesis

All reactions were carried out under an argon atmosphere. Anhydrous 1,4-dioxane, dimethylsulfoxide, tetrahydrofuran, acetonitrile and dichloromethane were purchased as Sure Seal™ bottles from Sigma-Aldridge. Potassium phosphate tribasic was purchased from Sigma-Aldrich and ground to a fine powder just prior to use. Sodium iodide powder was purchased from Alfa Aesar and ground to a fine powder just prior to use. Copper(I) iodide (99.999%) powder was purchased from Sigma-Aldrich. 6-bromoindole were purchased from Chem-Impex International. All other reagents were commercially available and used without further purification. Flash chromatography was performed using a Teledyne ISCO CombiFlash Rf system. NMR spectra were recorded on Bruker DRX-300 and DRX-400 instruments. The following abbreviations were used to describe multiplicities: s = singlet; d = doublet; t = triplet; q = quartet; p = pentet; m = multiplet; br = broad. High-resolution mass spectrometry (HRMS) was performed by the Columbia University Mass Spectroscopy Core Facility with a JOEL HX110 mass spectrometer by means of fast atom bombardment (FAB).

Spectroscopic Measurements

Absorbance and fluorescence measurements and spectra were obtained with a Tecan Infinite 200 spectrofluorometer. Measurements were performed in quartz cuvettes with a 1-cm pathlength.

Live Cell Imaging

HEK 293T and HeLA cells were cultured in DMEM w/ glutamine (Gibco #11995) with 10% v/v fetal bovine serum and 1% v/v Pen/Strep. All cells were maintained under 5% CO₂ at 37°C. For live cell protein labeling, cells were plated in 8-well chambered #1 borosilicate coverglass (Thermo, Nunc 155411) 24 h before transfection with expression plasmids for eDHFR-H2B¹ and EGFP-H2B or PM-eDHFR² and EGFP-H2B (0.4 ug DNA per well) using Fugene HD (Roche). 24 h after transfection, 300 uL of media containing 1 uM of the TMP-probe (made by adding 0.3 uL of a 1 mM stock of the TMP-probe in DMF to the media) was added to the wells. Cells were incubated with the staining solution for 10 min at 37°C, followed by quickly washing with fresh media twice before imaging.
Figure S1. UV absorbance spectra of compound 1b after varying durations of exposure to 254 nm light.

Figure S2. Overlay of (A) absorbance and (B) fluorescence spectra for the 1b photoproduct and authentic oxazine 2. Fluorescence excitation = 594 nm.
Figure S3. Reverse-phase HPLC analysis of the azido-acyl oxazine photoreaction.
Figure S4. Photoactivation of the azido-acetyl oxazine 1a in live HeLa cells.

Scheme S1. Preparation of iodoindolines for diaryl ether coupling reactions. Conditions: (a) NaI, CuI (10 mol %), N,N'-dimethylethylenediamine (20 mol %), 1,4-dioxane, 110 °C, 22 hrs. (b) NaBH₃CN, AcOH, rt. (c) NaBH₄, AcOH, rt. (d) ethyl 4-bromobutyrate, NaI, DIPEA, 60 °C. (e) Pd/C, H₂, MeOH, rt. Compounds were synthesized according to previously published protocols and methods.
**General procedure for the Copper(I) promoted coupling of phenols and aryl iodides.**

According to the published protocol by Maiti and Buchwald\(^4\), an oven dried, sealable glass vessel was charged with a magnetic stirbar, the phenol (2.40 mmol), potassium phosphate (4.00 mmol, 849 mg), copper(I) iodide (0.20 mmol, 38 mg), 2-picolinic acid (0.40 mmol, 49 mg), and the aryl iodide, if a solid (2.00 mmol). The vessel was then fitted with a rubber septum, evacuated under vacuum and backfilled with argon. This process was repeated 3 times. The vessel was then charged with DMSO (4.0 mL), or if the aryl iodide is a liquid, the vessel was charged with the aryl iodide as a solution in DMSO. The rubber septum was removed and the reaction vessel was immediately sealed tightly with a Teflon screw cap. The reaction was then heated to 85 °C for 16-24 hours. After cooling to room temperature, the reaction was diluted with 10 mL of water and extracted with ethyl acetate (25 mL, 4x). The combined organic layers were washed with brine and dried over Na\(_2\)SO\(_4\), then concentrated in vacuo to a crude residue. Purification by flash chromatography (hexanes/ethyl acetate) afforded the diaryl ethers as colorless oils, which were stored at -20 °C under inert atmosphere.

![Chemical structure](image)

**5-(1-ethylindolin-6-ylxy)-2-methylaniline (5).** Following the general procedure, 3-amino-4-methyl phenol S04 (1.13 g, 9.18 mmol) and 1-ethyl-6-iodoindoline 4 (2.09 g, 7.65 mmol) were coupled to provide S1 (1.66 g, 81%) as a colorless oil. \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta\) 7.00 (dd, \(J = 7.9, 1.4\) Hz, 2H), 6.41 (dd, \(J = 8.1, 2.4\) Hz, 1H), 6.38 (d, \(J = 2.4\) Hz, 1H), 6.30 (dd, \(J = 7.8, 2.1\) Hz, 1H), 6.22 (d, \(J = 2.1\) Hz, 1H), 3.62 (br s, 2H), 3.41 (t, \(J = 8.2\) Hz, 2H), 3.12 (q, \(J = 7.2\) Hz, 2H), 2.96 (t, \(J = 8.2\) Hz, 2H), 2.16 (s, 3H), 1.20 (t, \(J = 7.2\) Hz, 3H). \(^{13}\)C NMR (300 MHz, CDCl\(_3\)) \(\delta\) 157.82, 157.57, 154.15, 145.96, 131.45, 125.42, 124.89, 117.02, 108.94, 107.99, 105.44, 99.58, 53.16, 43.28, 28.32, 17.06, 12.22. HRMS (FAB+) Calcd. For C\(_{17}\)H\(_{20}\)N\(_2\)O\(_2\) [M\(^+\)]: 268.1576; found 268.1580.
ethyl 4-(6-(3-amino-4-methylphenoxy)indolin-1-yl)butanoate (S05). Following the general procedure, 3-amino-4-methyl phenol S04 (880 mg, 7.15 mmol) and S03 (2.14 g, 5.96 mmol) were coupled to provide XX (1.69 g, 80%) as a colorless oil. $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 6.98 (dd, $J = 7.8$ Hz, 3.1 Hz, 2H), 6.39 (dd, $J = 7.8$, 2.3 Hz, 1H), 6.38 (s, 1H), 6.28 (dd, $J = 7.8$, 2.1 Hz, 1H), 6.19 (d, $J = 2.1$ Hz, 1H), 6.14 (q, $J = 7.1$ Hz, 2H), 3.57 (br s, 2H), 3.42 (t, $J = 8.3$ Hz, 2H), 3.08 (t, $J = 7.1$ Hz, 2H), 2.96 (t, $J = 8.2$ Hz, 2H), 2.41 (t, $J = 7.4$ Hz, 2H), 2.15 (s, 3H), 1.94 (p, $J = 7.3$ Hz, 2H), 1.27 (t, $J = 7.1$ Hz, 3H). $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 173.74, 157.86, 157.46, 154.29, 145.90, 131.44, 125.00, 124.91, 117.10, 108.99, 107.96, 105.49, 99.27, 60.79, 53.99, 48.76, 32.20, 28.34, 23.22, 17.05, 14.61. HRMS (FAB+) Calcd. For C$_{21}$H$_{26}$N$_2$O$_3$ $^+$ [M$^+$]: 354.1943; found 354.1946.

4-bromo-5-(5-bromo-1-ethylindolin-6-yloxy)-2-methylaniline (6). Compound 5 (1.58 g, 5.91 mmol) was dissolved in dichloromethane (20 mL) and cooled to 0°C in an ice bath. The solution was then treated with N-bromosuccinimide (2.16 g, 12.1 mmol) in small portions over a period of 10 minutes. After stirring at 0°C for 30 minutes, the reaction showed completion by TLC and was treated with 25 mL of sat. aq. NaHCO$_3$. The organic layer was separated from the aqueous, and the aqueous layer extracted twice with 25 mL of dichloromethane. The combined organic layers were washed with brine and dried over Na$_2$SO$_4$, then concentrated in vacuo to a red-brown oil. Purified by flash chromatography to yield 1.91 g of 6 as a beige solid (89%). $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.26 (s, 1H), 7.23 (s, 1H), 6.14 (s, 1H), 6.08 (s, 1H), 3.59 (br s, 2H), 3.41 (t, $J = 8.3$ Hz, 2H), 3.05 (q, $J = 7.2$ Hz, 2H), 2.97 (t, $J = 8.3$ Hz, 2H), 2.13 (s, 3H), 1.14 (t, $J = 7.2$ Hz, 3H). $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 153.39, 153.15, 152.91, 145.25, 134.71,
General procedure for the Copper(I) promoted coupling and cyclization of di-aryl bromides with amides and carbamates.

According to the published protocol by Klapars et al\textsuperscript{5}, an oven dried, sealable glass vessel was charged with a magnetic stir bar, the di-bromoaryl ether (1.00 mmol), the amide or carbamate (1.20-1.50 mmol), potassium carbonate (3.00 mmol, 415 mg), copper(I) iodide (0.10 mmol, 19 mg), and N,N’-DMED (0.20 mmol, 17.6 mg). The vessel was then fitted with a rubber septum, evacuated under vacuum and backfilled with argon. This process was repeated 3 times. The vessel was then charged with toluene (1.2 mL). The rubber septum was removed and the reaction vessel was immediately sealed tightly with a Teflon screw cap. The reaction was then heated to 110 °C for 24 hours. After cooling to room temperature, the reaction was diluted with 10 mL of water and extracted with ethyl acetate (25 mL, 4x). The combined organic layers were washed with brine and dried over Na\textsubscript{2}SO\textsubscript{4}, then concentrated in vacuo to a crude residue. Purification by flash chromatography (hexanes/ethyl acetate) afforded the cyclized acyl-leuco oxazines as colorless amorphous solids, which were stored protected from light at -20 °C under inert atmosphere.

![Chemical Structure](image)

\[1-(8\text{-}amino\text{-}1\text{-}ethyl\text{-}7\text{-}methyl\text{-}2,3\text{-}dihydropyrrolo[3,2\text{-}b]phenoxazin\text{-}5(1\text{H})\text{-}y1)ethan\text{-}1\text{-}one\] (7a).

Following the general procedure, 6 (319 mg, 0.75 mmol) and acetamide (53 mg, 0.90 mmol) were
coupled to provide 7a (218 mg, 90%) as a colorless amorphous solid. $^1$H NMR (400 MHz, CDCl$_3$) δ 7.13 (s, 2H), 6.44 (s, 1H), 6.21 (s, 1H), 3.64 (br s, 2H), 3.38 (t, $J$ = 8.2 Hz, 2H), 3.12 (q, $J$ = 7.2 Hz, 2H), 2.95 (t, $J$ = 8.2 Hz, 2H), 2.29 (s, 3H), 2.16 (s, 3H), 0.91 (t, $J$ = 7.2 Hz, 3H). $^{13}$C NMR (300 MHz, CDCl$_3$) δ 170.13, 151.69, 151.49, 150.86, 143.48, 126.63, 124.94, 121.44, 121.13, 119.88, 117.15, 103.06, 96.20, 53.01, 43.40, 28.38, 23.27, 17.37, 12.10. HRMS (FAB+) Calcd. For C$_{19}$H$_{21}$N$_3$O$_2$+ [M$^+$]: 323.1634; found 323.1644.

![Chemical structure of tert-butyl 8-amino-1-ethyl-7-methyl-2,3-dihydropyrrolo[3,2-b]phenoxazine-5(1H)-carboxylate (7b).]

**tert-butyl 8-amino-1-ethyl-7-methyl-2,3-dihydropyrrolo[3,2-b]phenoxazine-5(1H)-carboxylate (7b).** Following the general procedure, 6 (319 mg, 0.75 mmol) and t-butyl carbamate (106 mg, 0.90 mmol) were coupled to provide 7b (204 mg, 71%) as a colorless amorphous solid. $^1$H NMR (400 MHz, CDCl$_3$) δ 7.20 (s, 2H), 6.37 (s, 1H), 6.16 (s, 1H), 3.57 (s, 2H), 3.35 (t, $J$ = 7.8 Hz, 2H), 3.11 (q, $J$ = 7.1 Hz, 2H), 2.93 (t, $J$ = 7.6 Hz, 2H), 2.14 (s, 3H), 1.55 (s, 9H), 1.19 (t, $J$ = 7.2 Hz, 3H). $^{13}$C NMR (75 MHz, CDCl$_3$) δ 153.29, 150.82, 150.77, 149.98, 142.72, 126.79, 124.72, 121.23, 120.67, 119.25, 116.98, 102.83, 96.03, 81.71, 53.11, 43.58, 28.68, 28.44, 17.31, 12.21. HRMS (FAB+) Calcd. For C$_{22}$H$_{27}$N$_3$O$_3$+ [M$^+$]: 381.2052; found 381.2058.

![Chemical structure of benzyl 8-amino-1-ethyl-7-methyl-2,3-dihydropyrrolo[3,2-b]phenoxazine-5(1H)-carboxylate (7c).]

**benzyl 8-amino-1-ethyl-7-methyl-2,3-dihydropyrrolo[3,2-b]phenoxazine-5(1H)-carboxylate (7c).** Following the general procedure, 6 (319 mg, 0.75 mmol) and benzyl carbamate (136 mg, 0.90 mmol)
were coupled to provide 7c (199 mg, 64%) as a colorless amorphous solid. $^1$H NMR (400 MHz, CDCl$_3$) δ 7.47 – 7.33 (m, 5H), 7.25 (d, $J = 4.3$ Hz, 2H), 6.39 (s, 1H), 6.20 (s, 1H), 5.30 (s, 2H), 3.60 (br s, 2H), 3.37 (t, $J = 8.2$ Hz, 2H), 3.12 (q, $J = 7.2$ Hz, 2H), 2.94 (t, $J = 8.2$ Hz, 2H), 2.13 (s, 3H), 1.21 (t, $J = 7.2$ Hz, 3H). $^{13}$C NMR (75 MHz, CDCl$_3$) δ 154.31, 151.17, 150.91, 150.06, 143.24, 136.70, 128.90, 128.52, 128.43, 128.22, 126.62, 124.94, 120.12, 118.80, 117.17, 102.89, 96.05, 68.17, 53.06, 43.50, 28.42, 17.30, 12.20. HRMS (FAB+) Calcd. For C$_{25}$H$_{25}$N$_3$O$_3$ $^+ \text{[M]}$: 415.1896; found 415.1907.

![methyl 8-amino-1-ethyl-7-methyl-2,3-dihydropyrrolo[3,2-b]phenoxazine-5(1H)-carboxylate (7d).](image1)

Following the general procedure, 6 (319 mg, 0.75 mmol) and methyl carbamate (68 mg, 0.90 mmol) were coupled to provide 7d (168 mg, 66%) as a colorless amorphous solid. $^1$H NMR (400 MHz, CDCl$_3$) δ 7.20 (d, $J = 5.7$ Hz, 2H), 6.39 (s, 1H), 6.17 (s, 1H), 3.83 (s, 3H), 3.60 (br s, 2H), 3.37 (t, $J = 8.1$ Hz, 2H), 3.11 (q, $J = 7.2$ Hz, 2H), 2.95 (t, $J = 8.1$ Hz, 2H), 2.15 (s, 3H), 1.19 (t, $J = 7.2$ Hz, 3H). $^{13}$C NMR (75 MHz, CDCl$_3$) δ 155.07, 151.16, 150.92, 150.11, 143.14, 126.48, 124.95, 120.97, 120.30, 118.82, 117.22, 102.88, 96.03, 53.54, 53.05, 43.47, 28.43, 17.37, 12.13. HRMS (FAB+) Calcd. For C$_{19}$H$_{25}$N$_3$O$_3$ $^+ \text{[M]}$: 339.1583; found 339.1578.

![8-amino-1-ethyl-7-methyl-2,3-dihydropyrrolo[3,2-b]phenoxazine-5(1H)-carbaldehyde (7e).](image2)

Following the general procedure, 6 (319 mg, 0.75 mmol) and formamide (51 mg, 1.13 mmol) were
coupled to provide 7e (137 mg, 59%) as a colorless amorphous solid. \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta\) 8.52 (s, 1H), 7.64 (d, \(J = 5.3\) Hz, 1H), 6.80 (d, \(J = 6.5\) Hz, 1H), 6.38 (d, \(J = 5.7\) Hz, 1H), 6.16 (d, \(J = 6.4\) Hz, 1H), 3.54 (br s, 2H), 3.36 (t, \(J = 7.8\) Hz, 2H), 3.11 (q, \(J = 7.2\) Hz, 2H), 2.93 (t, \(J = 7.8\) Hz, 2H), 2.13 (s, 3H), 1.17 (t, \(J = 7.2\) Hz, 3H). \(^{13}\)C NMR (75 MHz, CDCl\(_3\)) \(\delta\) 160.05, 151.49, 148.70, 147.81, 143.74, 125.75, 124.78, 120.01, 119.26, 117.44, 114.78, 102.78, 96.00, 52.90, 43.33, 28.38, 17.30, 12.02. HRMS (FAB+) Calcd. For C\(_{18}\)H\(_{19}\)N\(_3\)O\(_2\)\(^+\) [M\(^+\)]: 309.1477; found 309.1473.

![Chemical structure of ethyl 4-(5-acetyl-8-amino-7-methyl-2,3-dihydropyrrolo[3,2-b]phenoxazin-1(5\(H\))-yl)butanoate (S07).](image)

**ethyl 4-(5-acetyl-8-amino-7-methyl-2,3-dihydropyrrolo[3,2-b]phenoxazin-1(5\(H\))-yl)butanoate (S07).**

Compound **S05** (1.58 g, 5.91 mmol) was dissolved in dichloromethane (20 mL) and cooled to 0°C in an ice bath. The solution was then treated with N-bromosuccinimide (1.62 g, 9.08 mmol) in small portions over a period of 10 minutes. After stirring at 0°C for 30 minutes, the reaction showed completion by TLC and was treated with 25 mL of sat. aq. NaHCO\(_3\). The organic layer was separated from the aqueous, and the aqueous layer extracted twice with 25 mL of dichloromethane. The combined organic layers were washed with brine and dried over Na\(_2\)SO\(_4\), then concentrated in vacuo to a red-brown oil. Purified by flash chromatography to yield 1.98 g of the dibromide as an amber oil (87%). Following the general procedure, the dibromide (512 mg, 1.00 mmol) and acetamide (71 mg, 1.20 mmol) were coupled to provide **S07** (310 mg, 76%) as a colorless amorphous solid. \(^1\)H NMR (400 MHz, CDCl\(_3\)) \(\delta\) 7.08 (m, 2H), 6.39 (s, 1H), 6.17 (s, 1H), 4.13 (q, \(J = 7.1\) Hz, 2H), 3.69 (s, 2H), 3.35 (t, \(J = 8.3\) Hz, 2H), 3.05 (t, \(J = 7.0\) Hz, 2H), 2.92 (t, \(J = 8.2\) Hz, 2H), 2.39 (t, \(J = 7.3\) Hz, 2H), 2.25 (s, 3H), 2.11 (s, 3H), 1.91 (p, \(J = 7.2\) Hz, 2H), 1.25 (t, \(J = 7.1\) Hz, 3H). \(^{13}\)C NMR (75 MHz, CDCl\(_3\)) \(\delta\) 173.68, 170.10, 151.68, 151.65, 150.78, 143.72, 126.51, 124.48,
General procedure for conversion of anilines to aryl azides.

Imidazole sulfonyl azide · HCl (172 mg, 0.821 mmol) prepared by the reported procedure was dissolved in anhydrous dichloromethane (6 mL) and treated with triethylamine (0.32 mL, 2.30 mmol). To this mixture, the aniline (0.657 mmol) was added followed by copper (II) sulfate pentahydrate (~1 mg, 1.0 mol %). The reaction was stirred at room temperature for 6 hours and monitored for consumption of starting material. If the reaction did not reach completion, it was supplemented with additional triethylamine and imidazole sulfonyl azide · HCl. Upon completion, the reaction was filtered through celite, then diluted with 10 mL of water and extracted with dichloromethane (25 mL, 4x). The combined organic layers were washed with brine and dried over Na₂SO₄, then concentrated in vacuo to a crude residue. Purification by flash chromatography (hexanes/ethyl acetate) afforded the azido-acyl oxazines as colorless amorphous solids, which were stored protected from light at -20 °C under inert atmosphere.

Following the general procedure, 7a (167 mg, 0.516 mmol) was converted to 1a (94 mg, 52%) and recovered as a colorless light-sensitive amorphous solid. ¹H NMR (400 MHz, CDCl₃) δ 7.32 (s, 1H), 7.06 (s, 1H), 6.88 (s, 1H), 6.22 (s, 1H), 3.41 (t, 2H), 3.15 (q, J = 7.2 Hz, 2H), 2.97 (t, J = 8.2 Hz, 2H), 2.30 (s,
1H NMR (400 MHz, CDCl$_3$) $\delta$ 7.32 (s, 1H), 7.19 (s, 1H), 6.80 (s, 1H), 6.16 (s, 1H), 3.38 (t, $J = 8.2$ Hz, 2H), 3.13 (q, $J = 7.2$ Hz, 2H), 2.94 (t, $J = 8.2$ Hz, 2H), 2.18 (s, 3H), 1.54 (s, 9H), 1.20 (t, $J = 7.2$ Hz, 3H). $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 173.72, 152.82, 151.23, 150.44, 135.74, 127.47, 126.35, 124.79, 124.06, 121.22, 118.47, 106.21, 95.59, 82.37, 53.91, 43.87, 28.60, 28.45, 17.15, 12.64. HRMS (FAB+) Calcd. For C$_{22}$H$_{25}$N$_5$O$_3^+$ [M$^+$]: 407.1957; found 407.1963.

**tert-butyl 8-azido-1-ethyl-7-methyl-2,3-dihydropyrrolo[3,2-b]phenoxazine-5(1H)-carboxylate (1b).**

Following the general procedure, 7b (208 mg, 0.545 mmol) was converted to 1b (146 mg, 66%) and recovered as a colorless light-sensitive amorphous solid. $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.32 (s, 1H), 7.19 (s, 1H), 6.80 (s, 1H), 6.16 (s, 1H), 3.38 (t, $J = 8.2$ Hz, 2H), 3.13 (q, $J = 7.2$ Hz, 2H), 2.94 (t, $J = 8.2$ Hz, 2H), 2.18 (s, 3H), 1.54 (s, 9H), 1.20 (t, $J = 7.2$ Hz, 3H). $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 170.00, 151.77, 151.48, 150.55, 136.63, 127.52, 127.01, 125.50, 124.47, 120.97, 119.11, 106.52, 96.08, 52.91, 43.26, 28.33, 23.29, 17.21, 12.07. HRMS (FAB+) Calcd. For C$_{19}$H$_{19}$N$_5$O$_2^+$ [M$^+$]: 349.1539; found 349.1539.

**ethyl 4-(5-acetyl-8-azido-7-methyl-2,3-dihydropyrrolo[3,2-b]phenoxazin-1(5H)-yl)butanoate (S08).**

Following the general procedure, S07 (305 mg, 0.745 mmol) was converted to S08 (184 mg, 57%) and recovered as a colorless light-sensitive amorphous solid. $^1$H NMR (400 MHz, CDCl$_3$) $\delta$ 7.29 (s, 1H), 7.04 (s, 1H), 6.85 (s, 1H), 6.20 (s, 1H), 4.16 (q, $J = 7.1$ Hz, 2H), 3.42 (t, $J = 8.3$ Hz, 2H), 3.10 (t, $J = 7.1$ Hz, 2H), 2.97 (t, $J = 8.3$ Hz, 2H), 2.42 (t, $J = 7.2$ Hz, 2H), 2.29 (s, 3H), 2.19 (s, 3H), 1.94 (p, $J = 7.2$ Hz, 2H), 1.27 (t, $J = 7.1$ Hz, 3H). $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 173.66, 169.98, 151.98, 151.47, 150.50, 136.61,
127.49, 126.94, 125.10, 124.44, 121.00, 119.09, 106.50, 95.84, 60.86, 53.80, 48.65, 32.01, 28.39, 23.30, 22.99, 17.25, 14.64. HRMS (FAB+) Calcd. For C_{23}H_{25}N_{5}O_{4}^{+} [M^{+}]: 435.1907; found 435.1896.

4-(8-imino-7-methyl-2,3-dihydropyrrolo[3,2-b]phenoxazin-1(8H)-yl)butanoic acid · HCl salt (S09). Compound S07 (25 g, 0.0574 mmol) was heated to 60 °C for 16 hours in a mixture of THF (3 mL) and 0.5N HCl (12 mL). The aqueous layer was extracted with 25 mL of a 4:1 mixture of ethyl acetate/hexanes, and then extracted with dichloromethane (3x 25 mL). The dichloromethane layers were concentrated under reduced pressure to yield S10 as a deep blue residue (15 mg, 71%). \(^1\)H NMR (400 MHz, MeOD) \(\delta\) 7.49 (s, 1H), 7.37 (s, 1H), 6.76 (s, 1H), 6.69 (s, 1H), 4.11 (t, \(J = 6.0\) Hz, 2H), 3.72 (t, \(J = 6.0\) Hz, 2H), 3.31 (m, 2H), 2.49 (t, \(J = 6.8\) Hz, 2H), 2.30 (s, 3H), 2.07 (p, \(J = 7.2\) Hz, 2H). \(^{13}\)C NMR (75 MHz, CDCl$_3$) \(\delta\) 175.27, 161.27, 158.18, 151.52, 147.62, 139.25, 136.91, 132.64, 132.05, 127.20, 126.70, 96.93, 91.14, 54.00, 46.86, 30.47, 25.99, 22.32, 16.36.

TMP-1a. Compound S08 (25 mg, 0.0574 mmol) was subjected to hydrolysis conditions as above (THF/0.5N HCl) for 5 hours at 60 °C, then purified by flash chromatography (DCM/MeOH) to yield 8 mg of the carboxylic acid (35%). Then, TMP-(PEG)$_3$-NH$_2$ (1.2 mg, 1.7 umol), the carboxylic acid of S08 (1.0
mg, 2.3 umol), HCTU (4.5 mg, 11 umol), triethylamine (1.0 uL, 7.2 umol) and dimethylformamide (0.2 mL) were combined in an amber vial, which was then purged with argon and sealed. Stirred at room temperature for 75 minutes, then purified directly by reverse phase HPLC to yield 0.8 mg of **TMP-1a** (48%). $^1$H NMR (400 MHz, MeOD) $\delta$ 7.39 (s, 1H), 7.24 (s, 1H), 7.14 (s, 1H), 6.97 (s, 1H), 6.58 (s, 2H), 6.30 (s, 1H), 3.93 (t, $J$ = 6.1 Hz, 2H), 3.82 (s, 6H), 3.67 (s, 2H), 3.63 - 3.59 (m, 4H), 3.59-3.52 (m, 4H), 3.52 - 3.46 (m, 4H), 3.43 (t, $J$ = 8.4 Hz, 2H), 3.27 (t, $J$ = 6.8 Hz, 4H), 3.11 (t, $J$ = 6.9 Hz, 2H), 2.96 (t, $J$ = 8.3 Hz, 2H), 2.29 (s, 3H), 2.29 (p, $J$ = 8.1 Hz, 4H), 2.20 (s, 3H), 1.94 (p, $J$ = 7.6 Hz, 2H), 1.86 - 1.69 (m, 8H). HRMS (FAB+) Calcd. For C$_{49}$H$_{66}$N$_{11}$O$_{10}^+$ [M$^+$]: 968.4989; found 968.5004.

**TMP-2.** TMP-(PEG)$_3$-NH$_2$ (1.2 mg, 1.7 umol), S09 (1.0 mg, 2.2 umol), HCTU (4.5 mg, 11 umol), triethylamine (1.0 uL, 7.2 umol) and dimethylformamide were combined in an amber vial, which was then purged with argon and sealed. Stirred at room temperature for 75 minutes, then purified directly by reverse phase HPLC to yield 1.2 mg of **TMP-2** (69%). $^1$H NMR (400 MHz, MeOD) $\delta$ 7.62 (s, 1H), 7.49 (s, 1H), 7.25 (s, 1H), 6.85 (s, 1H), 6.82 (s, 1H), 6.57 (s, 2H), 4.12 (t, $J$ = 6.9 Hz, 2H), 3.93 (t, $J$ = 6.1 Hz, 2H), 3.81 (s, 6H), 3.73 (t, $J$ = 7.1 Hz, 2H), 3.67 (s, 2H), 3.65 - 3.60 (m, 4H), 3.60 - 3.55 (m, 4H), 3.53 - 3.47 (m, 4H), 3.36 (m, 2H), 3.26 (dt, $J$ = 9.4, 6.9 Hz, 4H), 2.34 (s, 3H), 2.34 (t, $J$ = 6.8 Hz, 2H), 2.27 (t, $J$ = 7.3 Hz, 2H), 2.10 (p, $J$ = 7.2 Hz, 2H), 1.87 - 1.67 (m, 8H). HRMS (FAB+) Calcd. For C$_{47}$H$_{64}$N$_9$O$_9^+$ [M$^+$]: 898.4822; found 898.4857.
References: