Supporting Information

Targeting of fumarate hydratase from *Mycobacterium tuberculosis* using allosteric inhibitors with a dimeric-binding mode

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Table of Contents

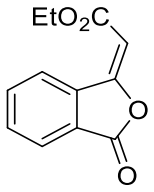
Synthetic Chemistry S2

Biochemical Assay Dose-response Curves S26

X-ray Crystallography S33

Synthetic Chemistry

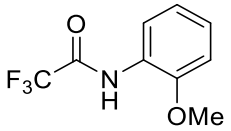
Ethyl-(E)-2-(3-oxoisobenzofuran-1(3H)-ylidene)acetate (18). [1, 2]



A solution of (carbethoxymethylene)triphenylphosphorane (4.70 g, 13.5 mmol) in chloroform (12.5 mL) was added dropwise to a solution of phthalic anhydride **17** (2.00 g, 13.5 mmol) in chloroform (12.5 mL). The reaction mixture was heated under reflux for 3 hours, then concentrated *in vacuo*. Purification by flash column chromatography (5% ethyl acetate in petroleum ether) afforded **18** (1.97 g, 67% yield).

^{1}H NMR (500 MHz, $CDCl_3$) 9.05 (1H, d, J = 8.0 Hz), 7.96 (1H, dt, J = 7.5, 1.0 Hz), 7.84-7.78 (1H, m), 7.70 (1H, td, J = 7.5, 0.9 Hz), 6.15 (1H, s), 4.30 (2H, q, J = 7.2 Hz), 1.36 (3H, t, J = 7.2 Hz); ^{13}C NMR (125 MHz, $CDCl_3$) 165.9, 165.7, 158.0, 136.3, 135.4, 132.6, 128.4, 126.7, 125.5, 102.6, 61.1, 14.4; ^{1}H NMR spectroscopic data consistent with literature. [2]

2,2,2-Trifluoro-N-(2-methoxyphenyl)acetamide (20). [3]

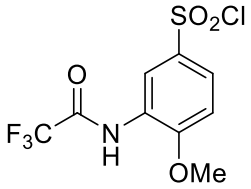


Trifluoroacetic anhydride (0.55 mL, 3.9 mmol) was added dropwise at 0 °C to a mixture of o-anisidine **19** (0.37 mL, 3.3 mmol), pyridine (0.39 mL, 4.9 mmol) and DCM (4 mL). The reaction mixture was warmed to room temperature and stirred over 3 days. Water (20 mL) was added dropwise at 0 °C to the reaction mixture. The product was extracted into DCM (3 x 20 mL). The combined organic extracts were dried ($MgSO_4$) and concentrated *in vacuo*.

Purification by flash column chromatography (0 – 20% ethyl acetate in petroleum ether) afforded **20** (0.713 g, 99% yield).

LCMS (ESI-): m/z 218.1 $[M - H]^-$, rt 2.14 minutes, >99%; ^{1}H NMR (400 MHz, $CDCl_3$) 8.57 (1H, br s), 8.32 (1H, d, J = 8.0 Hz), 7.17 (1H, t, J = 8.0 Hz), 7.01 (1H, t, J = 7.7 Hz), 6.94 (1H, d, J = 8.2 Hz), 3.93 (3H, s); ^{13}C NMR (100 MHz, $CDCl_3$) 154.5 (q, J = 37 Hz), 148.4, 126.1, 125.2, 121.4, 120.3, 115.8 (q, J = 288 Hz), 110.3, 56.0; ^{1}H NMR spectroscopic data consistent with literature.[3]

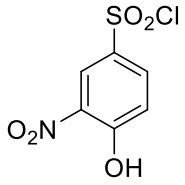
4-Methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride (21).[3]



Chlorosulfonic acid (0.41 mL, 6.2 mmol) was added dropwise at 0 °C to a solution of 2,2,2-trifluoro-N-(2-methoxyphenyl)acetamide **20** (0.683 g, 3.12 mmol) in DCM (6 mL). The reaction mixture was warmed to room temperature and stirred over 16 hours. Water (20 mL) was added dropwise at 0 °C to the reaction mixture. The product was extracted into DCM (3 x 20 mL). The combined organic extracts were dried ($MgSO_4$) and concentrated *in vacuo* to afford **21** (0.767 g, 77% yield).

^{1}H NMR (400 MHz, $CDCl_3$) 9.05 (1H, s), 8.59 (1H, br s), 7.90 (1H, d, J = 8.8 Hz), 7.11 (1H, d, J = 8.9 Hz), 4.09 (3H, s); ^{13}C NMR (100 MHz, $CDCl_3$) 154.9 (q, J = 38 Hz), 153.1, 137.1, 126.1, 125.9, 118.9, 115.5 (q, J = 289 Hz), 110.5, 57.1; ^{1}H NMR spectroscopic data consistent with literature.[3]

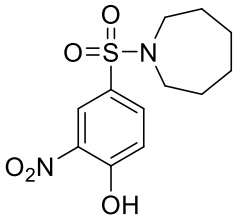
4-Hydroxy-3-nitrobenzenesulfonyl chloride (23).



Chlorosulfonic acid (0.96 mL, 14 mmol) was added dropwise at 0 °C to a solution of 2-nitrophenol **22** (1.00 g, 7.19 mmol) in chloroform (5 mL). The reaction mixture was heated under reflux for 90 minutes. Water (15 mL) was added dropwise at 0 °C to the reaction mixture. The product was extracted into DCM (3 x 25 mL). The combined organic extracts were dried ($MgSO_4$) and concentrated *in vacuo* to afford **23** (1.33 g, 78% yield).

LCMS (ESI-): m/z 236.0 $[M - H]^-$, rt 1.89 minutes, >99%; ^1H NMR (400 MHz, $CDCl_3$) 11.12 (1H, s), 8.84 (1H, d, J = 2.4 Hz), 8.21 (1H, dd, J = 9.0, 2.4 Hz), 7.42 (1H, d, J = 9.0 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) 159.5, 136.2, 134.9, 132.9, 126.0, 122.4; ν_{max}/cm^{-1} 3249 (br, O-H), 3088, 1615, 1578, 1539 (N=O), 1328 (N=O).

4-(Azepan-1-ylsulfonyl)-2-nitrophenol (24).

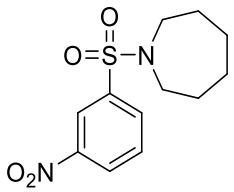


Hexamethyleneimine (0.108 mL, 0.963 mmol) and *N*,*N*-diisopropylethylamine (0.305 mL, 1.75 mmol) were added to a solution of 4-hydroxy-3-nitrobenzenesulfonyl chloride **23** (0.208 g, 0.875 mmol) in DCM (2 mL). The reaction mixture was stirred over 15 hours, then water (10 mL) and aqueous HCl (37.5% w/v, 5 mL) were added. The product was extracted into DCM (3 x 25 mL). The combined organic extracts were dried ($MgSO_4$) and concentrated *in*

vacuo. Purification by flash column chromatography (0 – 40% ethyl acetate in petroleum ether) afforded **24** (0.190 g, 64% yield).

LCMS (ESI+): m/z 301.2 $[M + H]^+$, (ESI-): m/z 299.1 $[M - H]^-$, rt 2.11 minutes, 88%; ^{1}H NMR (400 MHz, $CDCl_3$) 10.85 (1H, s), 8.56 (1H, d, J = 2.4 Hz), 7.96 (1H, dd, J = 8.9, 2.2 Hz), 7.29 (1H, d, J= 8.7 Hz), 3.29 (4H, t, J = 5.9 Hz), 1.79-1.69 (4H, m), 1.65-1.58 (4H, m); ^{13}C NMR (100 MHz, $CDCl_3$) 157.5, 135.2, 133.1, 132.5, 124.8, 121.4, 48.5, 29.3, 27.0; ν_{max}/cm^{-1} 2939, 1615, 1583, 1528 (N=O), 1330 (N=O); HRMS (ESI)+: m/z calculated for $[C_{12}H_{16}N_2O_5S + Na]^+$ = 323.0672, observed 323.0661.

1-((3-Nitrophenyl)sulfonyl)azepane (27).

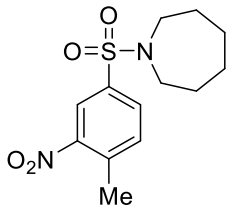


Hexamethyleneimine (0.303 mL, 2.71 mmol) was added dropwise at 0 °C to a suspension of sodium hydride (60% in mineral oil, 0.271 g, 6.77 mmol) in DMF (3 mL). The reaction mixture was stirred at 0 °C over 20 minutes. A solution of 3-nitrobenzenesulfonyl chloride **25** (0.500 g, 2.26 mmol) in DMF (2 mL) was added dropwise at 0 °C to the reaction mixture. The reaction mixture was warmed to room temperature and stirred over 90 min. Water (25 mL) was added dropwise at 0 °C to the reaction mixture, followed by ethyl acetate (25 mL) with the resultant aqueous layer discarded. The organic layer was washed with water (2 x 25 mL) and brine (25 mL), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (0 – 20% ethyl acetate in petroleum ether) afforded **27** (0.267 g, 42% yield).

^{1}H NMR (400 MHz, $CDCl_3$) 8.61 (1H, t, J = 1.9 Hz), 8.40 (1H, ddd, J = 8.2, 2.2, 1.0 Hz), 8.12 (1H, ddd, J = 7.8, 1.7, 1.1 Hz), 7.73 (1H, t, J = 8.0 Hz), 3.31 (4H, t, J = 5.9 Hz), 1.80-

1.68 (4H, m), 1.65-1.55 (4H, m); ^{13}C NMR (100 MHz, $CDCl_3$) 148.5, 142.0, 132.5, 130.5, 126.8, 122.1, 48.6, 29.3, 26.9; ν_{max}/cm^{-1} 3101, 2937, 2858, 1608, 1523 (N=O), 1338 (N=O); HRMS (ESI)+: m/z calculated for $[C_{12}H_{16}N_2O_4S + H]^+$ = 285.0904, observed 285.0897.

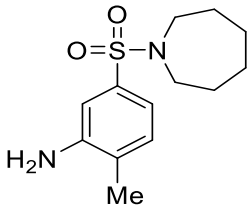
1-((4-Methyl-3-nitrophenyl)sulfonyl)azepane (28).



Hexamethyleneimine (0.285 mL, 2.55 mmol) was added dropwise at 0 °C to a suspension of sodium hydride (60% in mineral oil, 0.110 g, 2.76 mmol) in DMF (3 mL). The reaction mixture was stirred at 0 °C over 20 minutes. A solution of 4-methyl-3-nitrobenzenesulfonyl chloride **26** (0.500 g, 2.12 mmol) in DMF (2 mL) was added dropwise at 0 °C to the reaction mixture. The reaction mixture was warmed to room temperature and stirred over 1 hour. Water (25 mL) was added dropwise at 0 °C to the reaction mixture, followed by ethyl acetate (25 mL) with the resultant aqueous layer discarded. The organic layer was washed with water (2 x 25 mL) and brine (25 mL), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (0 – 20% ethyl acetate in petroleum ether) afforded **28** (0.453 g, 72% yield).

LCMS (ESI+): m/z 299.2 $[M + H]^+$, rt 2.15 minutes, >99%; ^{1}H NMR (400 MHz, $CDCl_3$) 8.35 (1H, d, J = 1.9 Hz), 7.89 (1H, dd, J = 8.0, 1.9 Hz), 7.50 (1H, d, J = 8.1 Hz), 3.30 (4H, t, J = 5.9 Hz), 2.67 (3H, s), 1.81-1.68 (4H, m), 1.65-1.54 (4H, m); ^{13}C NMR (100 MHz, $CDCl_3$) 149.3, 139.3, 137.9, 133.9, 130.8, 123.4, 48.5, 29.3, 27.0, 20.6; ν_{max}/cm^{-1} 2934, 2861, 1608, 1523 (N=O), 1338 (N=O); HRMS (ESI)+: m/z calculated for $[C_{13}H_{18}N_2O_4S + H]^+$ = 299.1060, observed 299.1065.

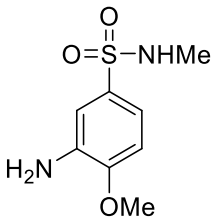
5-(Azepan-1-ylsulfonyl)-2-methylaniline (29).



Sodium borohydride (83 mg, 2.2 mmol) was added portionwise at 0 °C to a suspension of $NiCl_2$ (95 mg, 0.74 mmol) in methanol (2 mL). The reaction mixture was warmed to room temperature and stirred over 30 minutes. 1-((4-Methyl-3-nitrophenyl)sulfonyl)azepane **28** (0.439 g, 1.47 mmol) was added at 0 °C to the reaction mixture, followed by further methanol (8 mL) and sodium borohydride (0.278 g, 7.36 mmol). The reaction mixture was warmed to room temperature and stirred over 45 minutes. Water (10 mL) was added at 0 °C and the mixture filtered through celite, eluted with methanol (10 mL) and water (15 mL). The filtrate was concentrated *in vacuo* to remove methanol, then extracted into ethyl acetate (3 x 25 mL). The combined organic extracts were washed (brine), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (20 – 50% ethyl acetate in petroleum ether) afforded **29** (0.347 g, 88% yield).

LCMS (ESI+): m/z 269.2 $[M + H]^+$, rt 1.90 minutes, >99%; ^1H NMR (400 MHz, $CDCl_3$) 7.13 (1H, d, J = 8.4 Hz), 7.10-7.04 (2H, m), 3.80 (2H, br s), 3.24 (4H, t, J = 5.9 Hz), 2.19 (3H, s), 1.77-1.65 (4H, m), 1.63-1.53 (4H, m); ^{13}C NMR (100 MHz, $CDCl_3$) 145.2, 137.9, 131.0, 126.7, 116.9, 112.8, 48.4, 29.3, 27.1, 17.6; ν_{max}/cm^{-1} 3490 (N-H), 3377 (N-H), 2930, 2858, 1625, 1574; HRMS (ESI)+: m/z calculated for $[C_{13}H_{20}N_2O_2S + H]^+$ = 269.1318, observed 269.1321.

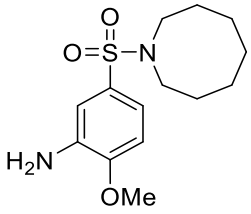
3-Amino-4-methoxy-N-methylbenzenesulfonamide (31a).



Methylamine (2 M in THF, 0.33 mL, 0.67 mmol) was added at 0 °C to a solution of 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.106 g, 0.334 mmol) in THF (2 mL). The reaction mixture was warmed to room temperature and stirred overnight. Further methylamine (2 M in THF, 2.0 mL, 4.0 mmol) was added, and the reaction mixture heated under reflux for 1 hour. Further methylamine (2 M in THF, 1.0 mL, 2.0 mmol) was added at room temperature, and the reaction mixture heated under reflux for 30 minutes. The reaction mixture was concentrated *in vacuo*, then ethanol (5 mL), water (5 mL) and aqueous HCl (37.5% w/v, 5 mL) were added. The reaction mixture was heated under reflux for 3 hours 30 minutes. The reaction mixture was adjusted to pH 14 by the dropwise addition of aqueous NaOH (10% w/v) at room temperature, then concentrated *in vacuo* to remove ethanol. The product was extracted into ethyl acetate (3 x 25 mL). The combined organic extracts were dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (25 – 75% ethyl acetate in petroleum ether) afforded **31a** (59 mg, 82% yield).

LCMS (ESI+): m/z 217.2 $[M + H]^+$, rt 1.23 minutes, >99%; ^{1}H NMR (400 MHz, CD_3CN) 7.12-7.06 (2H, m), 6.92 (1H, d, J = 8.0 Hz), 5.28-5.07 (1H, m), 4.39 (2H, br s), 3.88 (3H, s), 2.46 (3H, d, J = 5.3 Hz); ^{13}C NMR (100 MHz, CD_3CN) 150.8, 138.8, 131.8, 117.8, 112.5, 110.7, 56.5, 29.6; ν_{max}/cm^{-1} 3388 (N-H), 3306 (N-H), 3043, 2919, 2840, 1590, 1509; HRMS (ESI)+: m/z calculated for $[C_8H_{12}N_2O_3S + Na]^+$ = 239.0461, observed 239.0463.

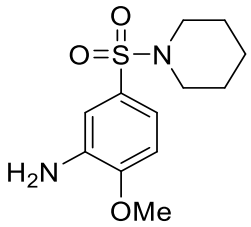
5-(Azocan-1-ylsulfonyl)-2-methoxyaniline (31b).



Heptamethyleneimine (95 µL, 0.76 mmol) was added dropwise at 0 °C to a suspension of sodium hydride (60% in mineral oil, 76 mg, 1.9 mmol) in DMF (2 mL). The reaction mixture was stirred at 0 °C over 20 minutes. A solution of 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.200 g, 0.630 mmol) in DMF (1 mL) was added dropwise at 0 °C to the reaction mixture. The reaction mixture was warmed to room temperature and stirred over 2 hours. Ethanol (5 mL) was added dropwise at 0 °C to the reaction mixture, followed by water (5 mL) and aqueous HCl (37.5% w/v, 5 mL). The reaction mixture was heated under reflux for 17 hours. The reaction mixture was adjusted to pH 9 by the dropwise addition of aqueous NaOH (10% w/v) at room temperature, then concentrated *in vacuo* to remove ethanol. The mixture was diluted with ethyl acetate (25 mL), and the resultant aqueous layer discarded. The organic layer was washed with water (2 x 25 mL) and brine (25 mL), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (0 – 50% ethyl acetate in petroleum ether) afforded **31b** (0.130 g, 69% yield).

LCMS (ESI+): m/z 299.2 $[M + H]^+$, rt 1.96 minutes, >99%; ^{1}H NMR (400 MHz, $CDCl_3$) 7.16 (1H, dd, J = 8.4, 2.2 Hz), 7.09 (1H, d, J = 2.2 Hz), 6.81 (1H, d, J = 8.3 Hz), 3.97 (2H, br s), 3.89 (3H, s), 3.11 (4H, t, J = 5.8 Hz), 1.75-1.57 (10H, m); ^{13}C NMR (100 MHz, $CDCl_3$) 150.1, 136.7, 130.8, 118.2, 112.9, 109.7, 55.8, 48.8, 28.0, 26.8, 25.3; ν_{max}/cm^{-1} 3488 (N-H), 3383 (N-H), 2914, 2851, 1729, 1611, 1577, 1512; HRMS (ESI)+: m/z calculated for $[C_{14}H_{22}N_2O_3S + H]^+$ = 299.1424, observed 299.1429.

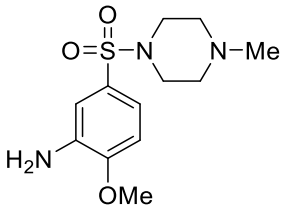
2-Methoxy-5-(piperidin-1-ylsulfonyl)aniline (31c).



Piperidine (62 µL, 0.63 mmol) was added dropwise at 0 °C to a suspension of sodium hydride (60% in mineral oil, 76 mg, 1.9 mmol) in DMF (1 mL). The reaction mixture was stirred at 0 °C over 20 minutes. A solution of 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.200 g, 0.630 mmol) in DMF (2 mL) was added dropwise at 0 °C to the reaction mixture. The reaction mixture was warmed to room temperature and stirred over 20 hours. Ethanol (5 mL) was added dropwise at 0 °C to the reaction mixture, followed by water (5 mL) and aqueous HCl (37.5% w/v, 5 mL). The reaction mixture was heated under reflux for 9 hours. The reaction mixture was adjusted to pH 7 by the dropwise addition of aqueous NaOH (10% w/v) at room temperature, then concentrated *in vacuo*. Water (15 mL) was added to the crude residue. The product was extracted into DCM (3 x 20 mL). The combined organic extracts were dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (20 – 50% ethyl acetate in petroleum ether) afforded **31c** (0.104 g, 61% yield).

LCMS (ESI+): m/z 271.2 $[M + H]^+$, rt 1.86 minutes, >99%; ^{1}H NMR (500 MHz, $CDCl_3$) 7.13 (1H, dd, J = 8.4, 2.1 Hz), 7.05 (1H, d, J = 2.2 Hz), 6.83 (1H, d, J = 8.4 Hz), 3.97 (2H, br s), 3.91 (3H, s), 2.95 (4H, t, J = 5.4 Hz), 1.63 (4H, quin, J = 5.7 Hz), 1.45-1.36 (2H, m); ^{13}C NMR (125 MHz, $CDCl_3$) 150.3, 136.6, 128.2, 118.9, 113.3, 109.7, 55.9, 47.1, 25.3, 23.7; ν_{max}/cm^{-1} 3475 (N-H), 3372 (N-H), 2939, 2851, 1616, 1581, 1510; HRMS (ESI)+: m/z calculated for $[C_{12}H_{18}N_2O_3S + Na]^+$ = 293.0930, observed 293.0935.

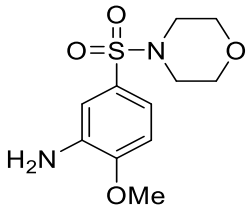
2-Methoxy-5-((4-methylpiperazin-1-yl)sulfonyl)aniline (31d).



1-Methylpiperazine (84 µL, 0.76 mmol) was added dropwise at 0 °C to a suspension of sodium hydride (60% in mineral oil, 76 mg, 1.9 mmol) in DMF (2 mL). The reaction mixture was stirred at 0 °C over 20 minutes. A solution of 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.200 g, 0.630 mmol) in DMF (1 mL) was added dropwise at 0 °C to the reaction mixture. The reaction mixture was warmed to room temperature and stirred over 2 hours. Ethanol (5 mL) was added dropwise at 0 °C to the reaction mixture, followed by water (5 mL) and aqueous HCl (37.5% w/v, 5 mL). The reaction mixture was heated under reflux for 18 hours. The reaction mixture was adjusted to pH 9 by the dropwise addition of aqueous NaOH (10% w/v) at room temperature, then concentrated *in vacuo* to remove ethanol. The mixture was diluted with ethyl acetate (25 mL), and the resultant aqueous layer discarded. The organic layer was washed with water (2 x 25 mL) and brine (25 mL), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (0 – 10% methanol in DCM) afforded **31d** (0.120 g, 65% yield).

LCMS (ESI+): m/z 286.3 $[M + H]^+$, rt 0.47 minutes, 97%; ^1H NMR (500 MHz, $(CD_3)_2SO$) 6.99-6.95 (2H, m), 6.88 (1H, dd, J = 8.5, 2.3 Hz), 5.23 (2H, s), 3.84 (3H, s), 2.82 (4H, br s), 2.34 (4H, t, J = 4.4 Hz), 2.13 (3H, s); ^{13}C NMR (125 MHz, $(CD_3)_2SO$) 149.5, 138.3, 126.3, 116.1, 111.5, 109.9, 55.6, 53.6, 45.7, 45.3; ν_{max}/cm^{-1} 3481 (N-H), 3376 (N-H), 2919, 2842, 2795, 1609, 1576, 1516; HRMS (ESI)+: m/z calculated for $[C_{12}H_{19}N_3O_3S + H]^+$ = 286.1220, observed 286.1225.

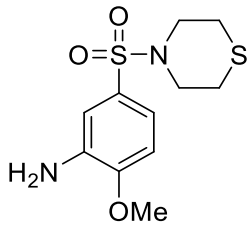
2-Methoxy-5-(morpholinosulfonyl)aniline (31e).



Morpholine (66 µL, 0.76 mmol) was added dropwise at 0 °C to a suspension of sodium hydride (60% in mineral oil, 76 mg, 1.9 mmol) in DMF (2 mL). The reaction mixture was stirred at 0 °C over 20 minutes. A solution of 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.200 g, 0.630 mmol) in DMF (1 mL) was added dropwise at 0 °C to the reaction mixture. The reaction mixture was warmed to room temperature and stirred over 2 hours. Ethanol (5 mL) was added dropwise at 0 °C to the reaction mixture, followed by water (5 mL) and aqueous HCl (37.5% w/v, 5 mL). The reaction mixture was heated under reflux for 18 hours. The reaction mixture was adjusted to pH 9 by the dropwise addition of aqueous NaOH (10% w/v) at room temperature, then concentrated *in vacuo* to remove ethanol. The mixture was diluted with ethyl acetate (25 mL), and the resultant aqueous layer discarded. The organic layer was washed with water (2 x 25 mL) and brine (25 mL), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (20 – 60% ethyl acetate in petroleum ether) afforded **31e** (0.117 g, 68% yield).

LCMS (ESI+): m/z 273.2 $[M + H]^+$, rt 1.40 minutes, >99%; ^{1}H NMR (500 MHz, $(CD_3)_2SO$) 7.01-6.96 (2H, m), 6.89 (1H, dd, J = 8.3, 2.3 Hz), 5.25 (2H, s), 3.85 (3H, s), 3.62 (4H, t, J = 4.8 Hz), 2.80 (4H, t, J = 4.7 Hz); ^{13}C NMR (125 MHz, $(CD_3)_2SO$) 149.6, 138.4, 125.9, 116.2, 111.5, 109.9, 65.3, 55.6, 45.9; ν_{max}/cm^{-1} 3486 (N-H), 3390 (N-H), 2924, 2864, 1691, 1611, 1507; HRMS (ESI)+: m/z calculated for $[C_{11}H_{16}N_2O_4S + Na]^+$ = 295.0723, observed 295.0719.

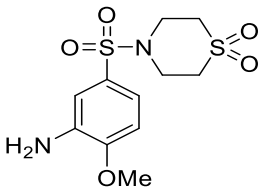
2-Methoxy-5-(thiomorpholinosulfonyl)aniline (31f).



Thiomorpholine (76 µL, 0.76 mmol) was added dropwise at 0 °C to a suspension of sodium hydride (60% in mineral oil, 76 mg, 1.9 mmol) in DMF (2 mL). The reaction mixture was stirred at 0 °C over 20 minutes. A solution of 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.200 g, 0.630 mmol) in DMF (1 mL) was added dropwise at 0 °C to the reaction mixture. The reaction mixture was warmed to room temperature and stirred over 2 hours. Ethanol (5 mL) was added dropwise at 0 °C to the reaction mixture, followed by water (5 mL) and aqueous HCl (37.5% w/v, 5 mL). The reaction mixture was heated under reflux for 17 hours. The reaction mixture was adjusted to pH 9 by the dropwise addition of aqueous NaOH (10% w/v) at room temperature, then concentrated *in vacuo* to remove ethanol. The mixture was diluted with ethyl acetate (50 mL), washed with water (3 x 25 mL) and brine (25 mL), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (0 – 50% ethyl acetate in petroleum ether) afforded **31f** (0.126 g, 67% yield).

LCMS (ESI+): m/z 289.2 $[M + H]^+$, rt 1.66 minutes, 96%; ^{1}H NMR (400 MHz, $CDCl_3$) 7.11 (1H, dd, J = 8.4, 2.2 Hz), 7.02 (1H, d, J = 2.2 Hz), 6.83 (1H, d, J = 8.4 Hz), 4.02 (2H, br s), 3.91 (3H, s), 3.31 (4H, t, J = 4.9 Hz), 2.69 (4H, t, J = 5.2 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) 150.5, 137.0, 128.6, 118.5, 112.8, 109.8, 55.9, 48.1, 27.5; ν_{max}/cm^{-1} 3489 (N-H), 3389 (N-H), 2970, 2914, 2852, 1730, 1611, 1577, 1512; HRMS (ESI)+: m/z calculated for $[C_{11}H_{16}N_2O_3S_2 + H]^+$ = 289.0675, observed 289.0688.

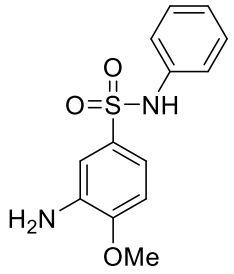
4-((3-Amino-4-methoxyphenyl)sulfonyl)thiomorpholine 1,1-dioxide (31g).



Triethylamine (0.66 mL, 4.7 mmol) and DCM (20 mL) were added to a mixture of thiomorpholine 1,1-dioxide (0.153 g, 1.13 mmol), 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.300 g, 0.944 mmol) and DMAP (35 mg, 0.28 mmol). The reaction mixture was stirred over 30 minutes, then concentrated *in vacuo*. Ethanol (30 mL), water (30 mL) and aqueous HCl (37.5% w/v, 30 mL) were added, and the reaction mixture heated under reflux for 3 hours. The reaction mixture was concentrated *in vacuo* to remove ethanol, then adjusted to pH 14 by the dropwise addition of aqueous NaOH (10% w/v). The product was extracted into DCM (3 x 100 mL). The combined organic extracts were washed (brine), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (20 – 70% ethyl acetate in petroleum ether, 0 – 7% methanol in DCM) afforded **31g** (0.250 g, 80% yield).

LCMS (ESI+): m/z 321.2 $[M + H]^+$, rt 1.44 minutes, 97%; ^{1}H NMR (400 MHz, $(CD_3)_2SO$) 7.03-6.92 (3H, m), 5.27 (2H, s), 3.85 (3H, s), 3.42-3.30 (4H, m), 3.22 (4H, t, J = 5.0 Hz); ^{13}C NMR (100 MHz, $(CD_3)_2SO$) 150.1, 138.9, 127.4, 116.2, 111.0, 110.4, 55.9, 50.2, 45.3; ν_{max}/cm^{-1} 3458 (N-H), 3369 (N-H), 2907, 2849, 1616, 1579, 1510; HRMS (ESI)+: m/z calculated for $[C_{11}H_{16}N_2O_5S_2 + Na]^+$ = 343.0393, observed 343.0394.

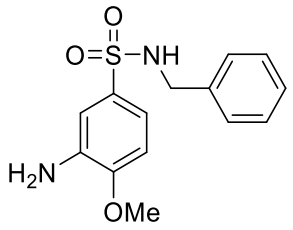
3-Amino-4-methoxy-N-phenylbenzenesulfonamide (31h).



Aniline (43 µL, 0.47 mmol) was added dropwise at 0 °C to a suspension of sodium hydride (60% in mineral oil, 57 mg, 1.4 mmol) in DMF (1 mL). The reaction mixture was stirred at 0 °C over 20 minutes. A solution of 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.150 g, 0.472 mmol) in DMF (2 mL) was added dropwise at 0 °C to the reaction mixture. The reaction mixture was warmed to room temperature and stirred over 1 hour. Ethanol (5 mL) was added dropwise at 0 °C to the reaction mixture, followed by water (5 mL) and aqueous HCl (37.5% w/v, 5 mL). The reaction mixture was heated under reflux for 10 hours. The reaction mixture was adjusted to pH 9 by the dropwise addition of aqueous NaOH (10% w/v) at room temperature, then concentrated *in vacuo* to remove ethanol. The product was extracted into DCM (3 x 25 mL). The combined organic extracts were washed (brine), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (0 – 75% ethyl acetate in petroleum ether) afforded **31h** (50 mg, 37% yield).

LCMS (ESI+): m/z 279.2 $[M + H]^+$, (ESI-): m/z 277.1 $[M - H]^-$, rt 1.77 minutes, 97%; ^1H NMR (400 MHz, $CDCl_3$) 7.25-7.19 (2H, m), 7.16 (1H, dd, J = 8.4, 2.3 Hz), 7.12-7.04 (4H, m), 6.82 (1H, s), 6.72 (1H, d, J = 8.5 Hz), 3.85 (3H, s), 3.54 (1H, br s); ^{13}C NMR (100 MHz, $CDCl_3$) 150.7, 136.9, 136.7, 131.0, 129.4, 125.2, 121.6, 118.7, 112.7, 109.6, 55.8; ν_{max}/cm^{-1} 3380 (N-H), 3250 (N-H), 1615, 1598, 1508; HRMS (ESI)+: m/z calculated for $[C_{13}H_{14}N_2O_3S + H]^+$ = 279.0798, observed 279.0796.

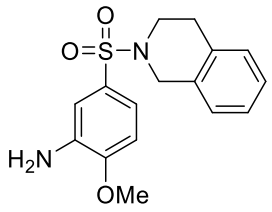
3-Amino-N-benzyl-4-methoxybenzenesulfonamide (31i).



Benzylamine (52 µL, 0.47 mmol) was added dropwise at 0 °C to a suspension of sodium hydride (60% in mineral oil, 57 mg, 1.4 mmol) in DMF (1 mL). The reaction mixture was stirred at 0 °C over 20 minutes. A solution of 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.150 g, 0.472 mmol) in DMF (2 mL) was added dropwise at 0 °C to the reaction mixture. The reaction mixture was warmed to room temperature and stirred over 1 hour. Ethanol (5 mL) was added dropwise at 0 °C to the reaction mixture, followed by water (5 mL) and aqueous HCl (37.5% w/v, 5 mL). The reaction mixture was heated under reflux for 10 hours. The reaction mixture was adjusted to pH 7 by the dropwise addition of aqueous NaOH (10% w/v) at room temperature, then concentrated *in vacuo* to remove ethanol. The product was extracted into DCM (3 x 25 mL). The combined organic extracts were washed (brine), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (0 – 10% methanol in DCM) afforded **31i** (89 mg, 59% yield).

LCMS (ESI+): m/z 293.2 $[M + H]^+$, (ESI-): m/z 291.1 $[M - H]^-$, rt 1.80 minutes, 92%; ^{1}H NMR (400 MHz, $CDCl_3$) 7.31-7.18 (6H, m), 7.16 (1H, d, J = 2.3 Hz), 6.82 (1H, d, J = 8.5 Hz), 4.59 (1H, t, J = 6.2 Hz), 4.08 (2H, d, J = 6.3 Hz), 3.91 (3H, s); ^{13}C NMR (100 MHz, $CDCl_3$) 150.5, 137.0, 136.6, 131.6, 128.8, 128.1, 128.0, 118.4, 112.6, 109.8, 55.9, 47.5; ^{1}H NMR spectroscopic data consistent with literature.[4]

5-((3,4-Dihydroisoquinolin-2(1H)-yl)sulfonyl)-2-methoxyaniline (31j).

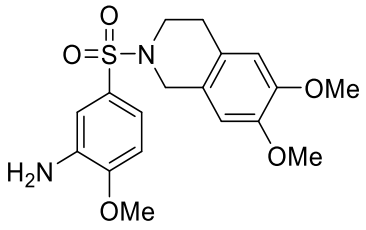


1,2,3,4-Tetrahydroisoquinoline (95 µL, 0.76 mmol) was added dropwise at 0 °C to a suspension of sodium hydride (60% in mineral oil, 76 mg, 1.9 mmol) in DMF (2 mL). The reaction mixture was stirred at 0 °C over 20 minutes. A solution of 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.200 g, 0.630 mmol) in DMF (1 mL) was added dropwise at 0 °C to the reaction mixture. The reaction mixture was warmed to room temperature and stirred over 1 hour. Ethanol (5 mL) was added dropwise at 0 °C to the reaction mixture, followed by water (5 mL) and aqueous HCl (37.5% w/v, 5 mL). The reaction mixture was heated under reflux for 24 hours, then further ethanol (10 mL), water (10 mL) and aqueous HCl (37.5% w/v, 10 mL) were added. The reaction mixture was heated under reflux for 24 hours. The reaction mixture was adjusted to pH 10 by the dropwise addition of aqueous NaOH (10% w/v) at room temperature, then concentrated *in vacuo* to remove ethanol. The mixture was diluted with ethyl acetate (50 mL), and the resultant aqueous layer discarded. The organic layer was washed with water (2 x 50 mL) and brine (50 mL), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (0 – 30% ethyl acetate in petroleum ether) afforded **31j** (0.146 g, 73% yield).

LCMS (ESI+): m/z 319.2 $[M + H]^+$, rt 1.94 minutes, >99%; ^{1}H NMR (400 MHz, $(CD_3)_2SO$) 7.18-7.09 (4H, m), 7.07 (1H, d, J = 1.9 Hz), 7.02-6.94 (2H, m), 5.23 (2H, s), 4.10 (2H, s), 3.83 (3H, s), 3.20 (2H, t, J = 5.9 Hz), 2.86 (2H, t, J = 5.9 Hz); ^{13}C NMR (100 MHz, $(CD_3)_2SO$) 149.5, 138.4, 133.0, 131.7, 128.7, 127.1, 126.6, 126.4, 126.1, 116.1, 111.4, 110.0,

55.6, 47.3, 43.6, 28.2; ν_{max}/cm^{-1} 3457 (N-H), 3363 (N-H), 1624, 1583, 1505; HRMS (ESI)+: m/z calculated for $[C_{16}H_{18}N_2O_3S + H]^+$ = 319.1111, observed 319.1122.

5-((6,7-Dimethoxy-3,4-dihydroisoquinolin-2(1H)-yl)sulfonyl)-2-methoxyaniline (31k).

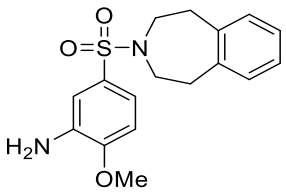


6,7-Dimethoxy-1,2,3,4-tetrahydroisoquinoline hydrochloride (0.174 g, 0.756 mmol) was added portionwise at 0 °C to a suspension of sodium hydride (60% in mineral oil, 0.126 g, 3.15 mmol) in DMF (3 mL). The reaction mixture was stirred at 0 °C over 20 minutes. A solution of 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.200 g, 0.630 mmol) in DMF (2 mL) was added dropwise at 0 °C to the reaction mixture. The reaction mixture was warmed to room temperature and stirred over 1 hour. Ethanol (15 mL) was added dropwise at 0 °C to the reaction mixture, followed by water (15 mL) and aqueous HCl (37.5% w/v, 15 mL). The reaction mixture was heated under reflux for 20 hours. The reaction mixture was concentrated *in vacuo* to remove ethanol, then adjusted to pH 14 by the dropwise addition of aqueous NaOH (10% w/v). The mixture was diluted with ethyl acetate (50 mL), and the resultant aqueous layer discarded. The organic layer was washed with water (2 x 50 mL) and brine (50 mL), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (0 – 100% ethyl acetate in petroleum ether) afforded **31k** (0.194 g, 81% yield).

LCMS (ESI+): m/z 379.3 $[M + H]^+$, rt 1.80 minutes, >99%; ^1H NMR (400 MHz, $CDCl_3$) 7.21 (1H, dd, J = 8.4, 2.2 Hz), 7.13 (1H, d, J = 2.2 Hz), 6.84 (1H, d, J = 8.4 Hz), 6.55 (1H, s), 6.50 (1H, s), 4.15 (2H, s), 4.01 (2H, br s), 3.90 (3H, s), 3.82 (3H, s), 3.81 (3H, s), 3.31 (2H, t, J = 5.9 Hz), 2.84 (2H, t, J = 5.8 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) 150.5, 147.9, 147.8, 136.8,

128.1, 125.2, 123.8, 118.9, 113.2, 111.4, 109.8, 109.2, 56.1, 56.0, 55.9, 47.5, 44.0, 28.7; ν_{max}/cm^{-1} 3483 (N-H), 3365 (N-H), 2966, 2930, 2842, 1661, 1611, 1578, 1512; HRMS (ESI)+: m/z calculated for $[C_{18}H_{22}N_2O_5S + Na]^+ = 401.1142$, observed 401.1124.

2-Methoxy-5-((1,2,4,5-tetrahydro-3H-benzo[d]azepin-3-yl)sulfonyl)aniline (31l).

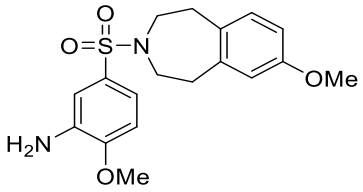


A solution of 2,3,4,5-tetrahydro-1H-benzo[d]azepine (0.167 g, 1.13 mmol) in DMF (1 mL) was added dropwise at 0 °C to a suspension of sodium hydride (60% in mineral oil, 0.113 g, 2.83 mmol) in DMF (2 mL). The reaction mixture was stirred at 0 °C over 20 minutes. A solution of 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.300 g, 0.944 mmol) in DMF (2 mL) was added dropwise at 0 °C to the reaction mixture. The reaction mixture was warmed to room temperature and stirred over 30 minutes. Ethanol (30 mL) was added dropwise at 0 °C to the reaction mixture, followed by water (30 mL) and aqueous HCl (37.5% w/v, 30 mL). The reaction mixture was heated under reflux for 15 hours. The reaction mixture was concentrated *in vacuo* to remove ethanol, then adjusted to pH 8 by the dropwise addition of aqueous NaOH (10% w/v). The mixture was diluted with ethyl acetate (100 mL), and the resultant aqueous layer discarded. The organic layer was washed with water (2 x 100 mL) and brine (100 mL), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (0 – 50% ethyl acetate in petroleum ether) afforded **31l** (0.131 g, 42% yield).

LCMS (ESI+): m/z 333.2 $[M + H]^+$, rt 2.07 minutes, >99%; ^{1}H NMR (500 MHz, $(CD_3)_2SO$) 7.10 (4H, s), 7.00-6.96 (1H, m), 6.93-6.88 (2H, m), 5.17 (2H, s), 3.80 (3H, s), 3.17-3.06 (4H, m), 2.96-2.87 (4H, m); ^{13}C NMR (125 MHz, $(CD_3)_2SO$) 149.3, 140.5, 138.3, 129.1, 129.0,

126.5, 115.5, 110.9, 109.9, 55.5, 48.2, 35.6; ν_{max}/cm^{-1} 3459 (N-H), 3367 (N-H), 2906, 2850, 1615, 1579, 1509; HRMS (ESI)+: m/z calculated for $[C_{17}H_{20}N_2O_3S + Na]^+$ = 355.1087, observed 355.1085.

2-Methoxy-5-((7-methoxy-1,2,4,5-tetrahydro-3H-benzo[d]azepin-3-yl)sulfonyl)aniline (31m).

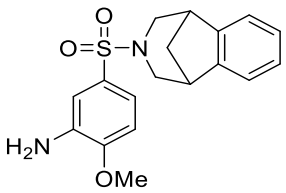


Triethylamine (0.29 mL, 2.1 mmol) and DCM (10 mL) were added to a mixture of 7-methoxy-2,3,4,5-tetrahydro-1H-benzo[d]azepine hydrochloride (96 mg, 0.45 mmol), 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.130 g, 0.409 mmol) and DMAP (10 mg, 0.082 mmol). The reaction mixture was stirred over 1 hour, then concentrated *in vacuo.* Ethanol (30 mL), water (30 mL) and aqueous HCl (37.5% w/v, 30 mL) were added, and the reaction mixture heated under reflux for 3 hours. The reaction mixture was adjusted to pH 10 by the dropwise addition of Na_2CO_3 solution at 0 °C and extracted into DCM (3 x 100 mL). The combined organic extracts were washed (brine), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (20 – 50% ethyl acetate in petroleum ether) afforded **31m** (0.111 g, 75% yield).

LCMS (ESI+): m/z 363.3 $[M + H]^+$, rt 2.01 minutes, >99%; ^{1}H NMR (500 MHz, $(CD_3)_2SO$) 7.00 (1H, d, J = 8.3 Hz), 6.97 (1H, d, J = 1.9 Hz), 6.93-6.87 (2H, m), 6.69 (1H, d, J = 2.7 Hz), 6.64 (1H, dd, J = 8.3, 2.7 Hz), 5.17 (2H, s), 3.80 (3H, s), 3.67 (3H, s), 3.16-3.02 (4H, m), 2.91-2.77 (4H, m); ^{13}C NMR (125 MHz, $(CD_3)_2SO$) 157.8, 149.3, 141.8, 138.4, 132.5, 130.2, 129.0, 115.6, 115.0, 111.2, 110.9, 109.9, 55.6, 55.0, 48.7, 48.2, 35.8, 34.7; ν_{max}/cm^{-1} 3474 (N-

H), 3443, 3367 (N-H), 2944, 2906, 2845, 1613, 1579, 1506; HRMS (ESI)+: m/z calculated for $[C_{18}H_{22}N_2O_4S + Na]^+$ = 385.1192, observed 385.1192.

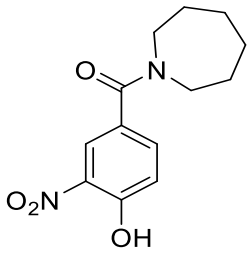
2-Methoxy-5-((1,2,4,5-tetrahydro-3H-1,5-methanobenzo[d]azepin-3-yl)sulfonyl)aniline (31n).



Triethylamine (0.33 mL, 2.4 mmol) and DCM (10 mL) were added to a mixture of 2,3,4,5-tetrahydro-1H-1,5-methano-3-benzazepine hydrochloride (0.102 g, 0.519 mmol), 4-methoxy-3-(2,2,2-trifluoroacetamido)benzenesulfonyl chloride **21** (0.150 g, 0.472 mmol) and DMAP (12 mg, 0.094 mmol). The reaction mixture was stirred over 1 hour, then concentrated *in vacuo*. Ethanol (30 mL), water (30 mL) and aqueous HCl (37.5% w/v, 30 mL) were added, and the reaction mixture heated under reflux for 16 hours. The reaction mixture was adjusted to pH 10 by the dropwise addition of Na_2CO_3 solution at 0 °C and extracted into DCM (3 x 100 mL). The combined organic extracts were washed (brine), dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (0 – 50% ethyl acetate in petroleum ether) afforded **31n** (0.118 g, 65% yield).

LCMS (ESI+): m/z 345.2 $[M + H]^+$, rt 2.05 minutes, >99%; ^{1}H NMR (500 MHz, $(CD_3)_2SO$) 7.25-7.14 (4H, m), 6.88 (1H, d, J = 8.5 Hz), 6.84 (1H, d, J = 2.3 Hz), 6.68 (1H, dd, J = 8.3, 2.3 Hz), 5.14 (2H, s), 3.83 (3H, s), 3.46-3.38 (2H, m), 3.23-3.16 (2H, m), 2.74 (2H, dd, J = 10.7, 1.3 Hz), 2.14-2.04 (1H, m), 1.50 (1H, d, J = 10.8 Hz); ^{13}C NMR (125 MHz, $(CD_3)_2SO$) 149.2, 144.4, 138.1, 128.0, 126.9, 122.4, 115.6, 111.2, 109.8, 55.6, 49.3, 41.5 (1 peak missing); ν_{max}/cm^{-1} 3455 (N-H), 3364 (N-H), 2950, 2854, 1733, 1621, 1578, 1509; HRMS (ESI)+: m/z calculated for $[C_{18}H_{20}N_2O_3S + Na]^+$ = 367.1087, observed 367.1092.

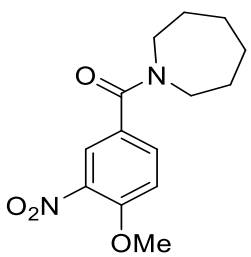
Azepan-1-yl(4-hydroxy-3-nitrophenyl)methanone (33).



T3P® (50 wt. % in DMF, 3.3 mL, 5.5 mmol) and *N*,*N*-diisopropylethylamine (0.95 mL, 5.5 mmol) were added to a solution of 4-hydroxy-3-nitrobenzoic acid **32** (0.500 g, 2.73 mmol) and hexamethyleneimine (0.61 mL, 5.5 mmol) in DMF (2 mL). The reaction mixture was stirred over 1 day, then diluted with water (15 mL), adjusted to pH 2 and extracted into DCM (3 x 20 mL). The combined organic extracts were dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (0 – 10% methanol in DCM) afforded **33** (0.174 g, 24% yield).

LCMS (ESI+): m/z 265.3 $[M + H]^+$, (ESI-): m/z 263.2 $[M - H]^-$, rt 1.84 minutes, >99%; ^1H NMR (400 MHz, CD_3CN) 10.34 (1H, br s), 8.10 (1H, d, J = 2.0 Hz), 7.65 (1H, dd, J = 8.7, 2.2 Hz), 7.20 (1H, d, J = 8.6 Hz), 3.58 (2H, t, J = 5.7 Hz), 3.37 (2H, t, J = 5.4 Hz), 1.83-1.71 (2H, m), 1.68-1.51 (6H, m); ^{13}C NMR (100 MHz, CD_3CN) 169.3, 155.8, 136.8, 134.5, 130.8, 124.4, 120.9, 50.5, 46.9, 30.0, 28.4, 28.0, 27.0; ν_{max}/cm^{-1} 2927, 2857, 1623 (C=O), 1531 (N=O), 1350 (N=O); HRMS (ESI)+: m/z calculated for $[C_{13}H_{16}N_2O_4 + H]^+$ = 265.1183, observed 265.1184.

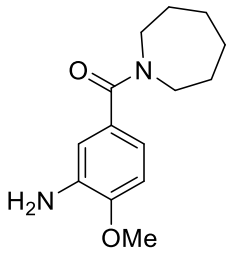
Azepan-1-yl(4-methoxy-3-nitrophenyl)methanone (34).



Dimethyl sulfate (0.115 mL, 1.21 mmol) was added to a suspension of azepan-1-yl(4-hydroxy-3-nitrophenyl)methanone **33** (0.160 g, 0.605 mmol) and potassium carbonate (0.167 g, 1.21 mmol) in acetone (5 mL). The reaction mixture was heated under reflux for 2 hours. The reaction mixture was diluted with water (15 mL) at 0 °C, then concentrated *in vacuo* to remove acetone. The product was extracted into DCM (3 x 20 mL). The combined organic extracts were dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (40 – 100% ethyl acetate in petroleum ether) afforded **34** (0.129 g, 77% yield).

LCMS (ESI+): m/z 279.3 $[M + H]^+$, rt 1.90 minutes, >99%; ^1H NMR (400 MHz, CD_3CN) 7.83 (1H, d, J = 2.1 Hz), 7.62 (1H, dd, J = 8.6, 2.1 Hz), 7.27 (1H, d, J = 8.7 Hz), 3.96 (3H, s), 3.58 (2H, t, J = 5.7 Hz), 3.37 (2H, t, J = 5.5 Hz), 1.83-1.71 (2H, m), 1.67-1.50 (6H, m); ^{13}C NMR (100 MHz, CD_3CN) 169.4, 153.9, 140.1, 133.6, 130.7, 124.7, 114.9, 57.6, 50.5, 46.8, 30.0, 28.5, 28.0, 27.0; ν_{max}/cm^{-1} 2928, 2854, 1615 (C=O), 1530 (N=O), 1350 (N=O); HRMS (ESI)+: m/z calculated for $[C_{14}H_{18}N_2O_4 + H]^+$ = 279.1339, observed 279.1345.

(3-Amino-4-methoxyphenyl)(azepan-1-yl)methanone (35).



Sodium borohydride (22 mg, 0.59 mmol) was added portionwise at 0 °C to a suspension of $NiCl_2 \cdot 6H_2O$ (47 mg, 0.20 mmol) in methanol (1 mL). The reaction mixture was warmed to room temperature and stirred over 20 minutes. A solution of azepan-1-yl(4-methoxy-3-nitrophenyl)methanone **34** (0.109 g, 0.392 mmol) in methanol (1 mL) was added at 0 °C to the reaction mixture, followed by sodium borohydride (52 mg, 1.4 mmol). The reaction

mixture was warmed to room temperature and stirred over 1 hour, then further sodium borohydride (30 mg, 0.78 mmol) was added at 0 °C. The reaction mixture was warmed to room temperature and stirred over 30 minutes. Water (15 mL) was added at 0 °C, and the reaction mixture filtered through celite. The filtrate was extracted into DCM (3 x 20 mL). The combined organic extracts were dried ($MgSO_4$) and concentrated *in vacuo*. Purification by flash column chromatography (20 – 100% ethyl acetate in petroleum ether) afforded **35** (62 mg, 64% yield).

LCMS (ESI+): m/z 249.3 $[M + H]^+$, rt 1.61 minutes, >99%; ^1H NMR (500 MHz, CD_3CN) 6.80 (1H, d, J = 8.1 Hz), 6.65 (1H, d, J = 2.0 Hz), 6.62 (1H, dd, J = 8.1, 2.1 Hz), 4.16 (2H, br s), 3.83 (3H, s), 3.61-3.48 (2H, m), 3.44-3.29 (2H, m), 1.82-1.67 (2H, m), 1.66-1.48 (6H, m); ^{13}C NMR (125 MHz, CD_3CN) 172.2, 148.3, 138.0, 131.5, 116.6, 113.3, 110.8, 56.2, 50.4, 46.6, 30.2, 28.5, 28.1, 27.0; ν_{max}/cm^{-1} 3466 (N-H), 3330 (N-H), 2922, 2853, 1608 (C=O), 1584, 1516; HRMS (ESI)+: m/z calculated for $[C_{14}H_{20}N_2O_2 + H]^+$ = 249.1598, observed 249.1600.

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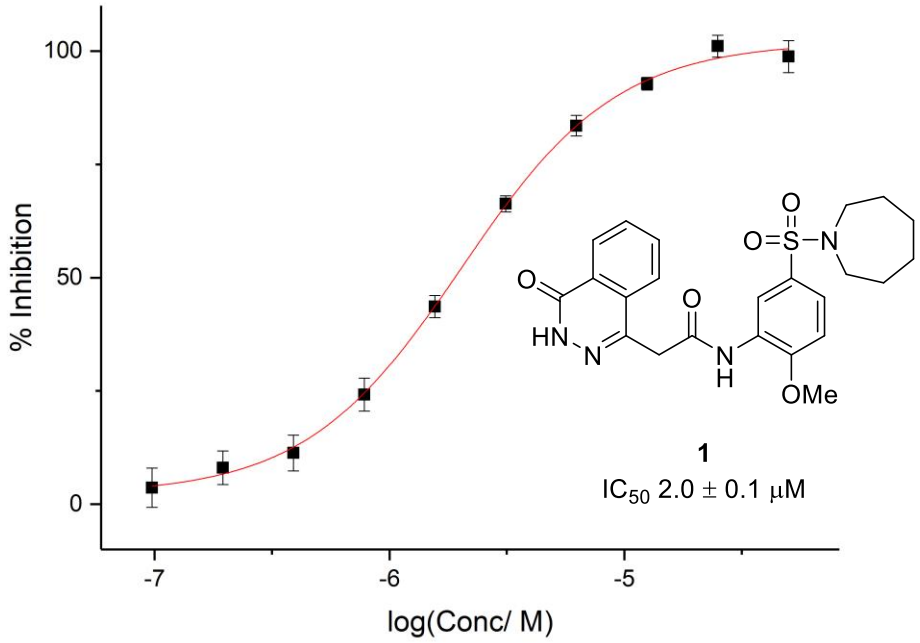
Biochemical Assay Dose-response Curves

Figure S1: Dose-response curves for **1** and **15g**, with data points representing an average of replicates (n = 6) and error bars indicating standard errors of the mean.



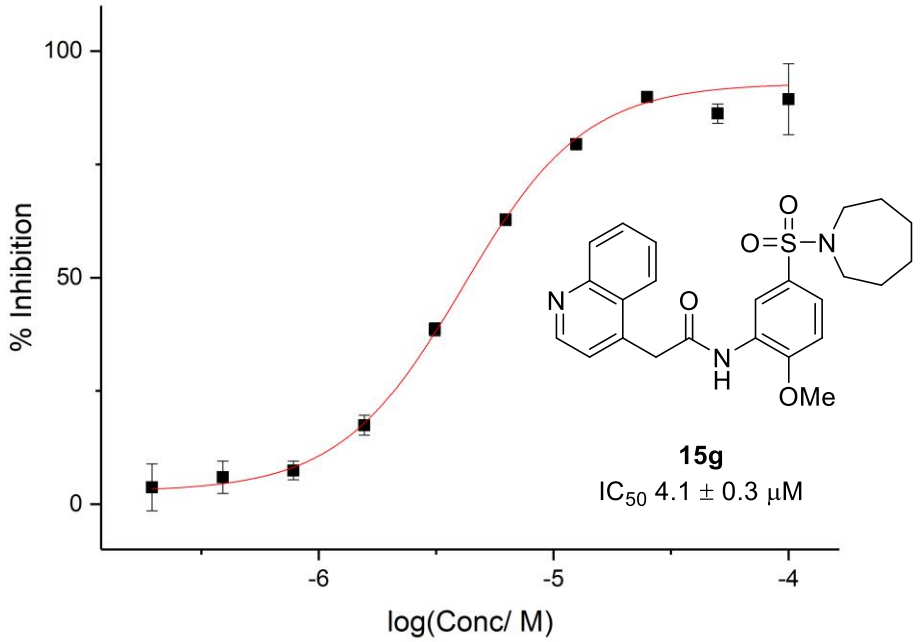
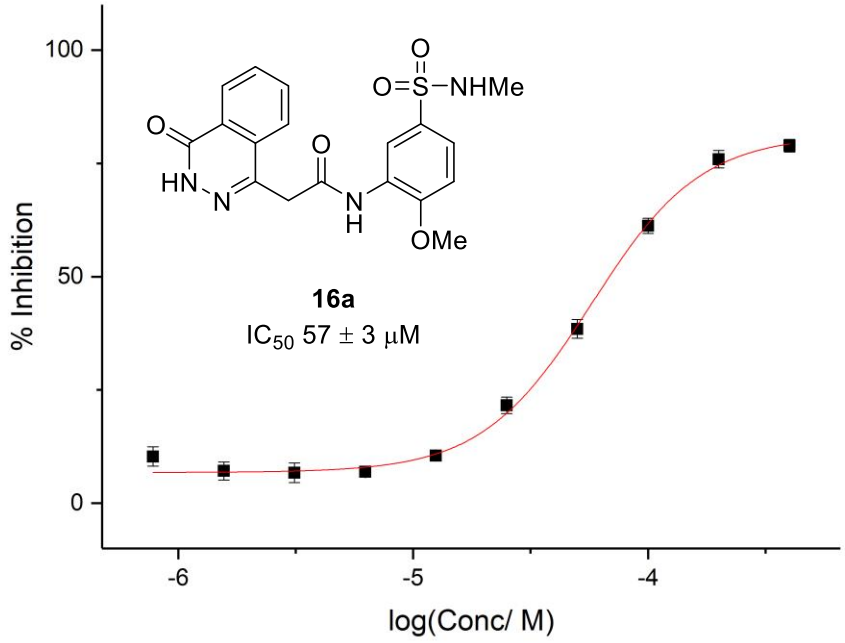


Figure S2: Dose-response curves for **16a** and **16b**, with data points representing an average of replicates (n = 6) and error bars indicating standard errors of the mean.



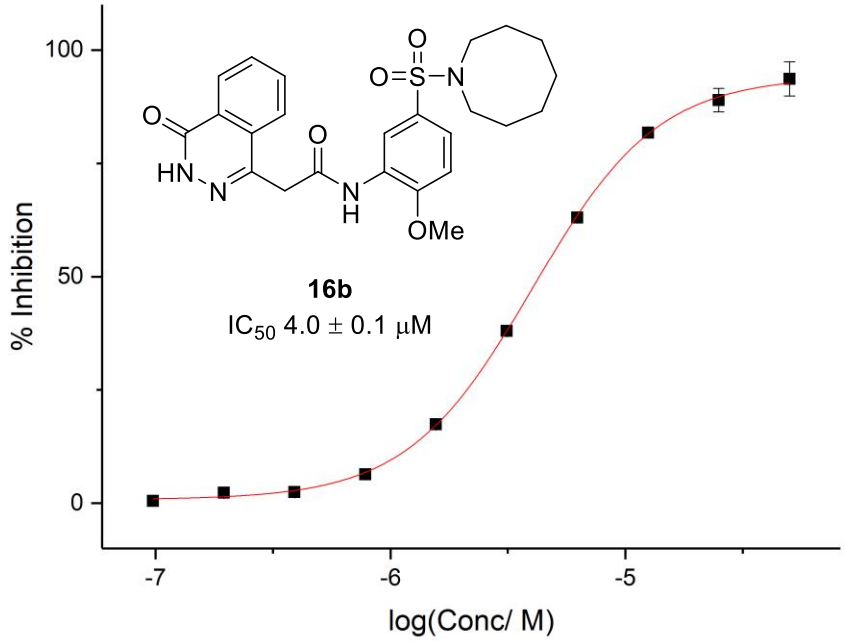
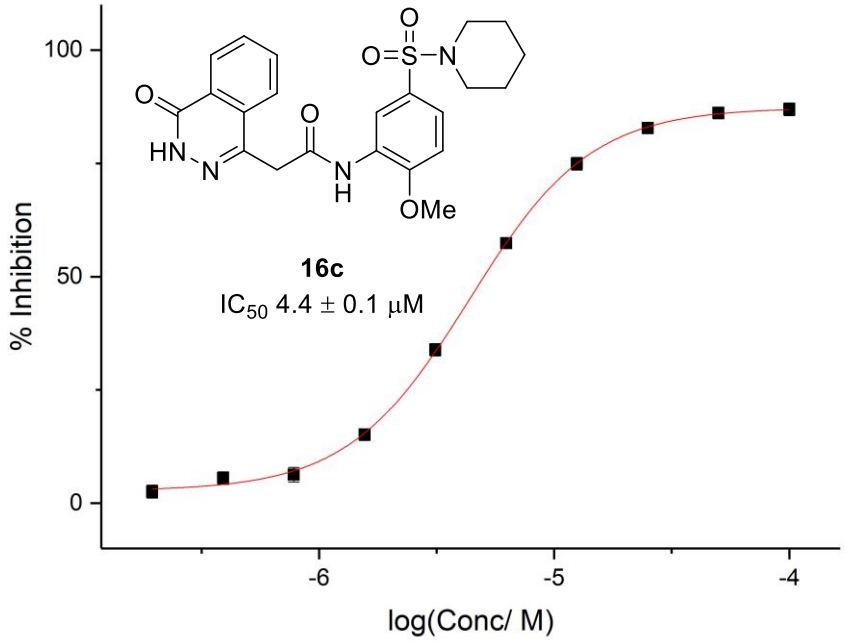


Figure S3: Dose-response curves for **16c** and **16d**, with data points representing an average of replicates (n = 6) and error bars indicating standard errors of the mean.



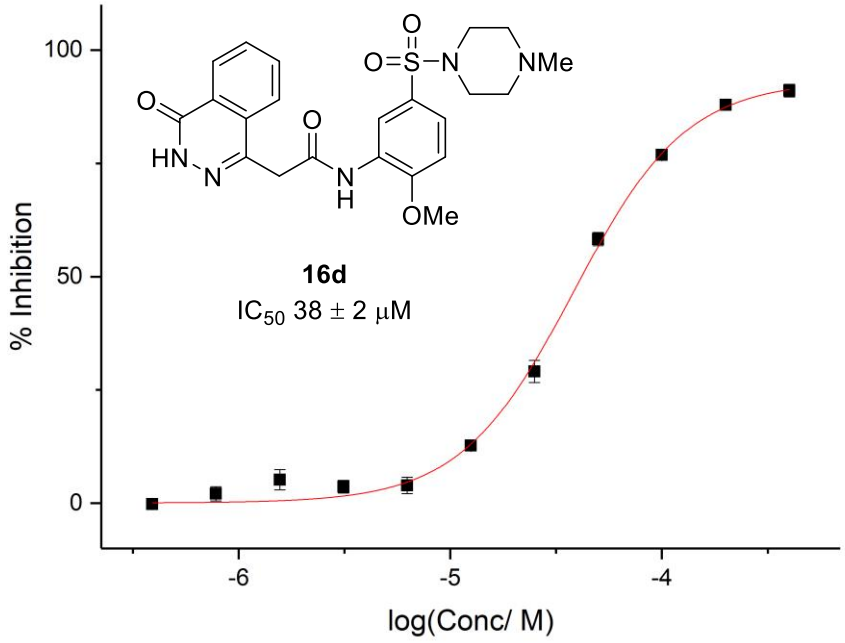
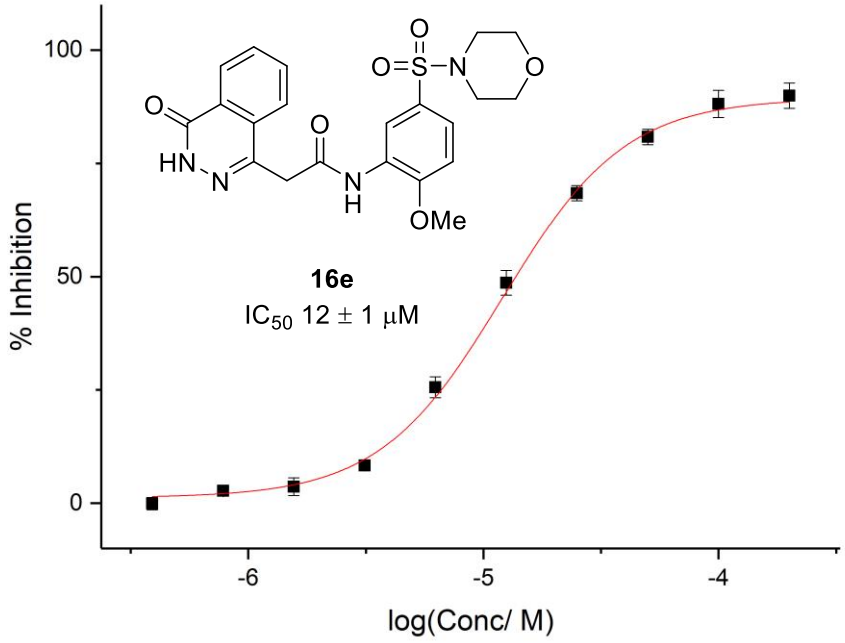


Figure S4: Dose-response curves for **16e** and **16f**, with data points representing an average of replicates (n = 6) and error bars indicating standard errors of the mean.



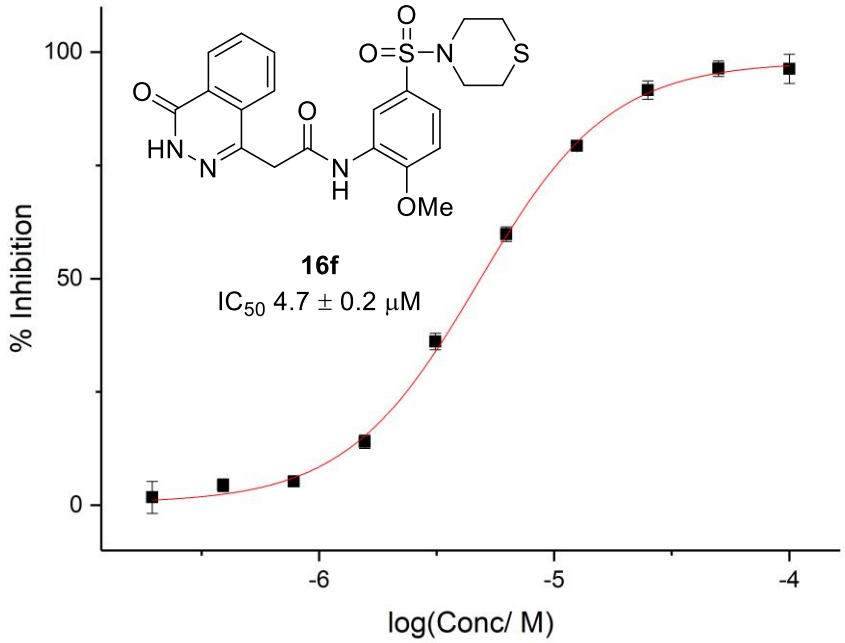
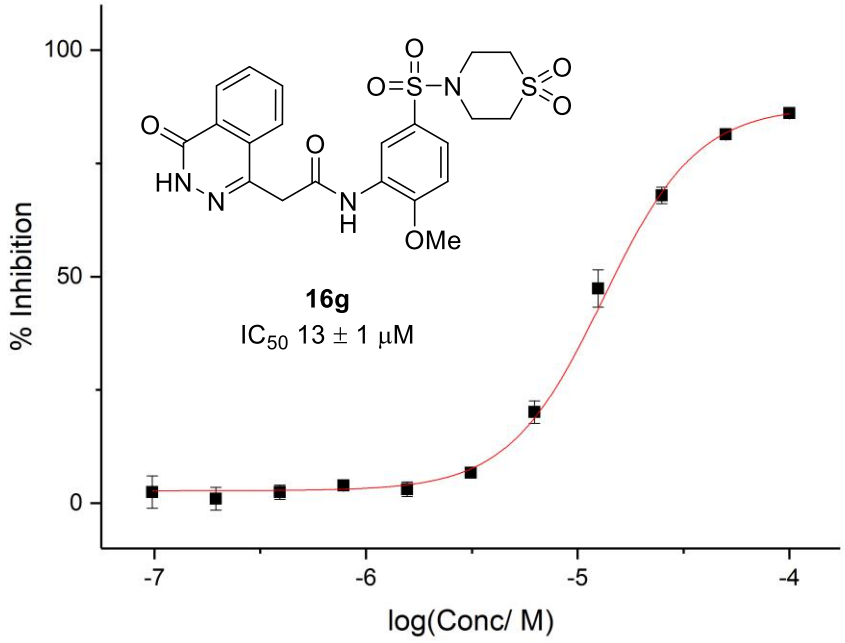


Figure S5: Dose-response curves for **16g** and **16h**, with data points representing an average of replicates (n = 6) and error bars indicating standard errors of the mean.



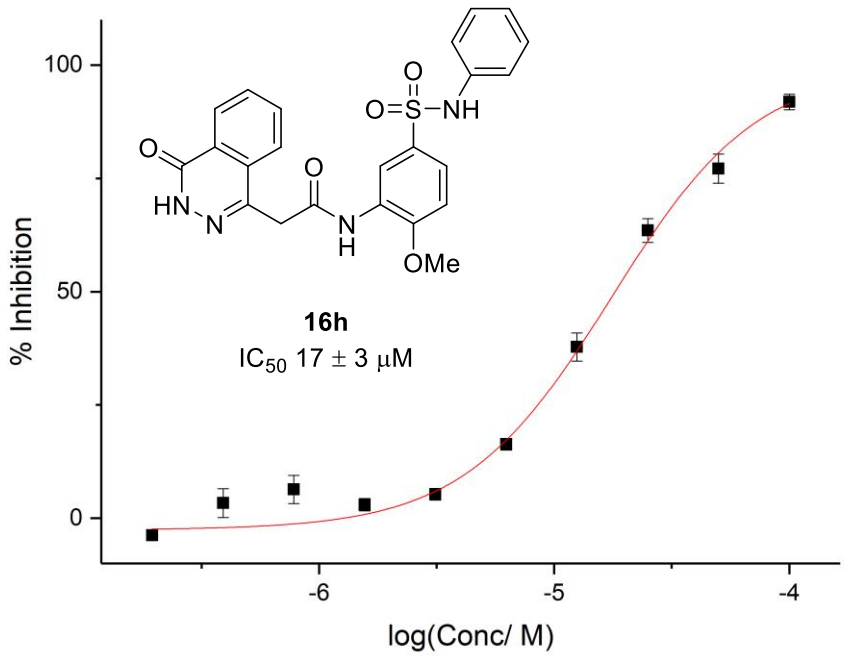
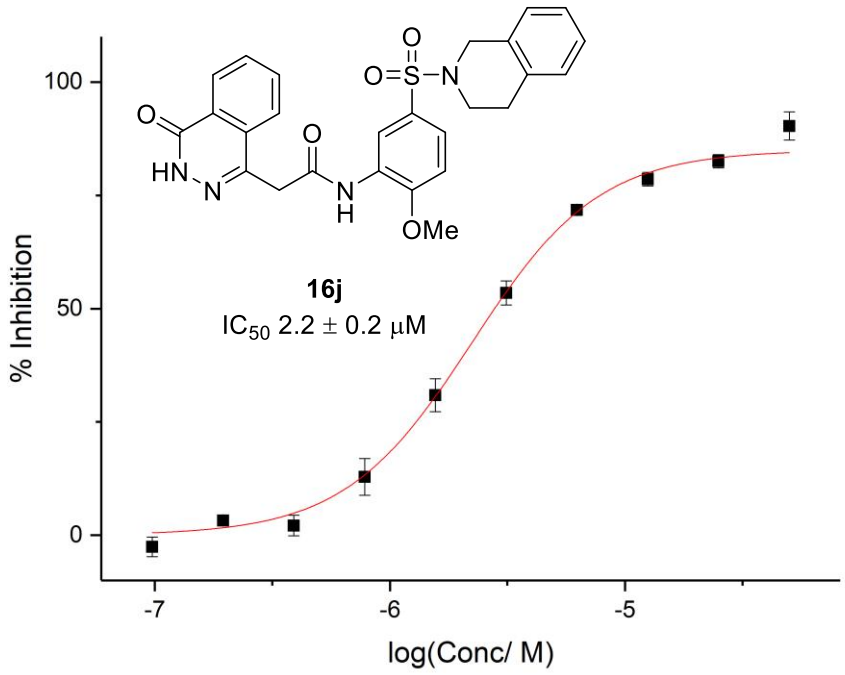


Figure S6: Dose-response curves for **16j** and **16k**, with data points representing an average of replicates (n = 6) and error bars indicating standard errors of the mean.



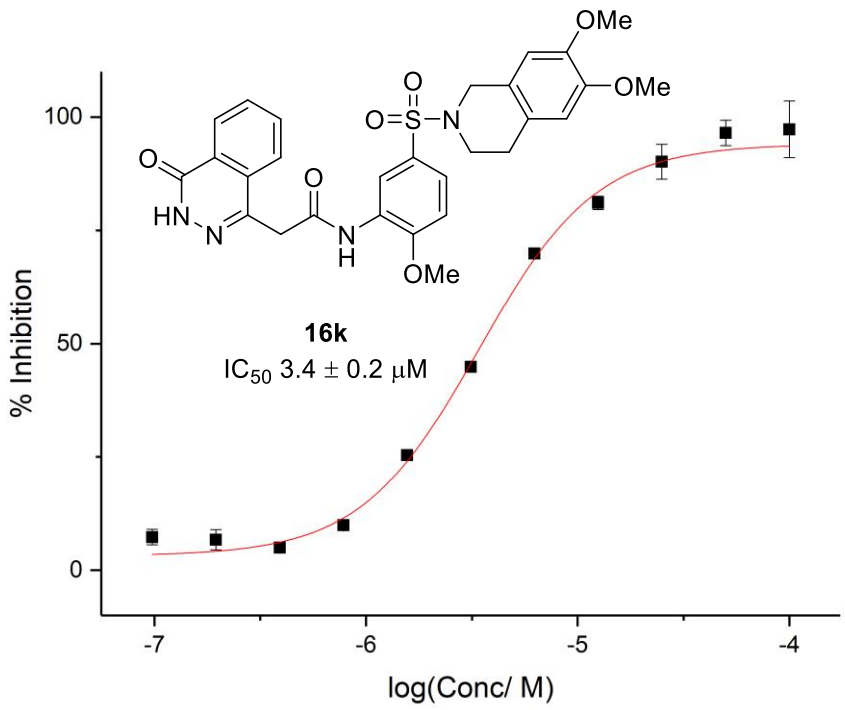
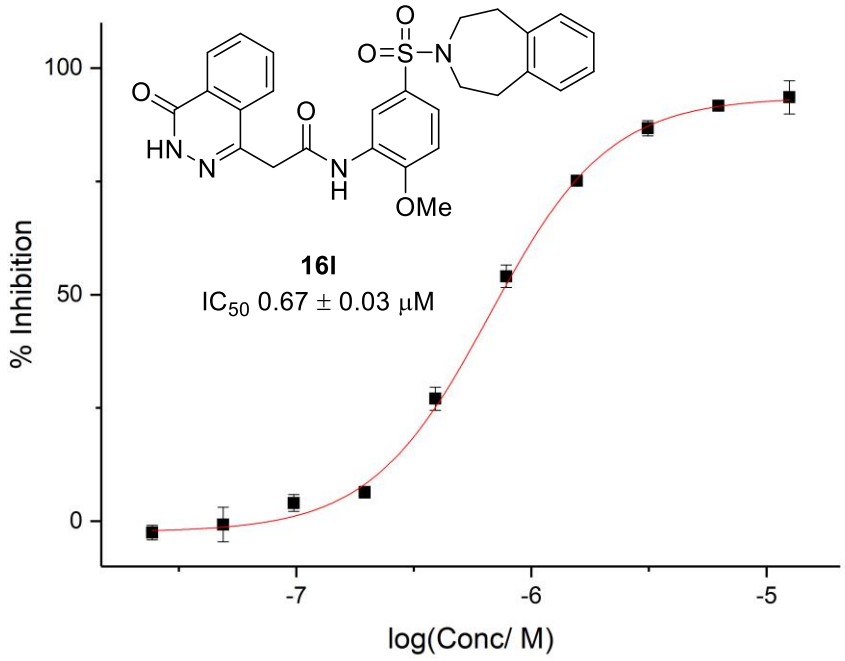
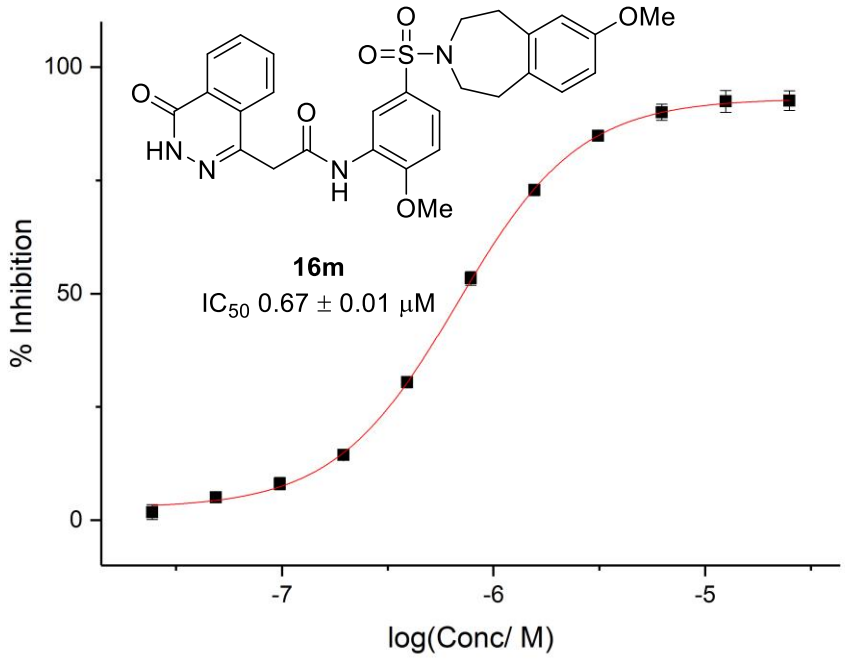


Figure S7: Dose-response curves for **16l** and **16m**, with data points representing an average of replicates (n = 6) and error bars indicating standard errors of the mean.





X-ray Crystallography

Figure S8: Stereo views of the X-ray crystal structure of *Mtb* fumarase in complex with **1** (PDB code 5F91, subunit A = white, subunit B = green, subunit C = cyan, subunit D = yellow, **1** = lilac), illustrating the interactions (yellow dashed lines) of **1** in the allosteric site with a focus on a) the whole allosteric site and b) the interactions of the methoxy group of **1**.

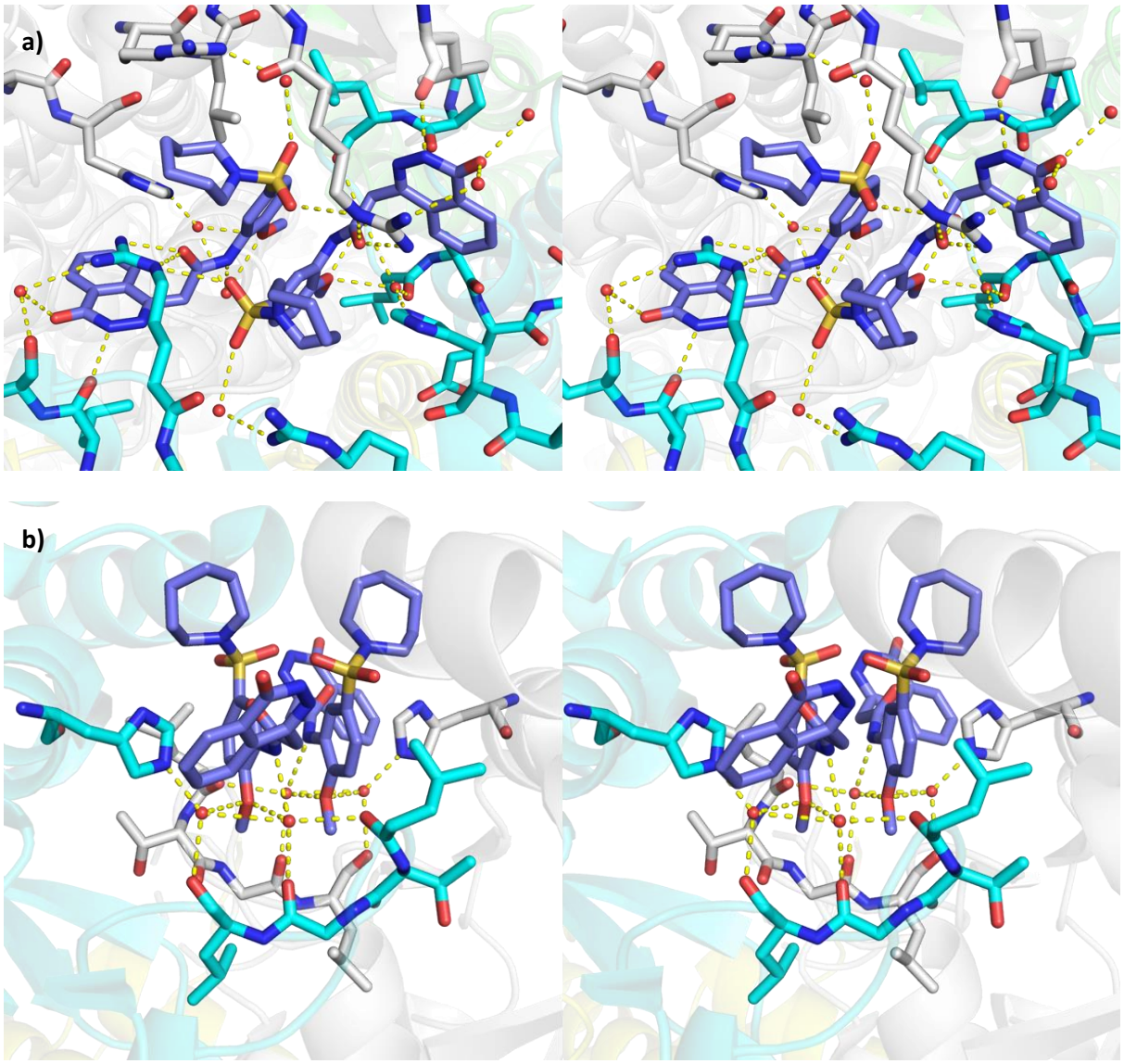


Figure S9: a) X-ray crystal structure of *Mtb* fumarase in complex with **16h** (PDB code 6S7S, subunit A = white, subunit B = green, subunit C = cyan, subunit D = yellow, **16h** = orange), illustrating the interactions (yellow dashed lines) of the ligand in the allosteric site. b) Overlay of the X-ray crystal structure of *Mtb* fumarase in complex with **1** (PDB code 5F91, subunit A = white, subunits B to D not shown, **1** = lilac) with the structure in complex with **16a** (PDB code 6S7K, subunit A = cyan, subunits B to D not shown, **16a** = light pink).

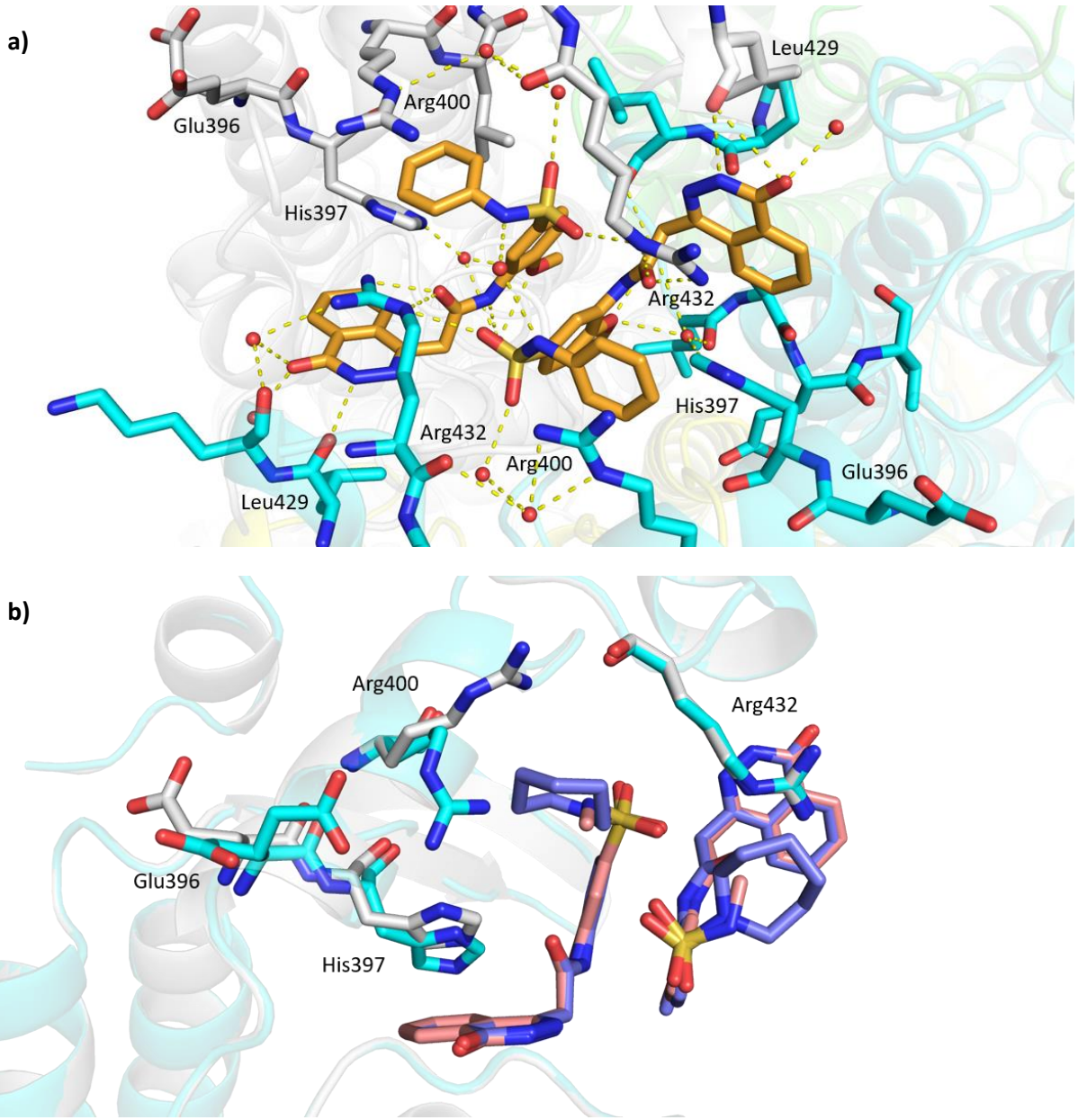


Figure S10: Overlays of the X-ray crystal structure of *Mtb* fumarase in complex with **1** (PDB code 5F91, subunit A = white, subunits B to D not shown, **1** = lilac) with the structures of *Mtb* fumarase (subunit A = cyan, subunits B to D not shown) in complex with a) **16b** (beige) (PDB code 6S43) and b) **16h** (orange) (PDB code 6S7S).

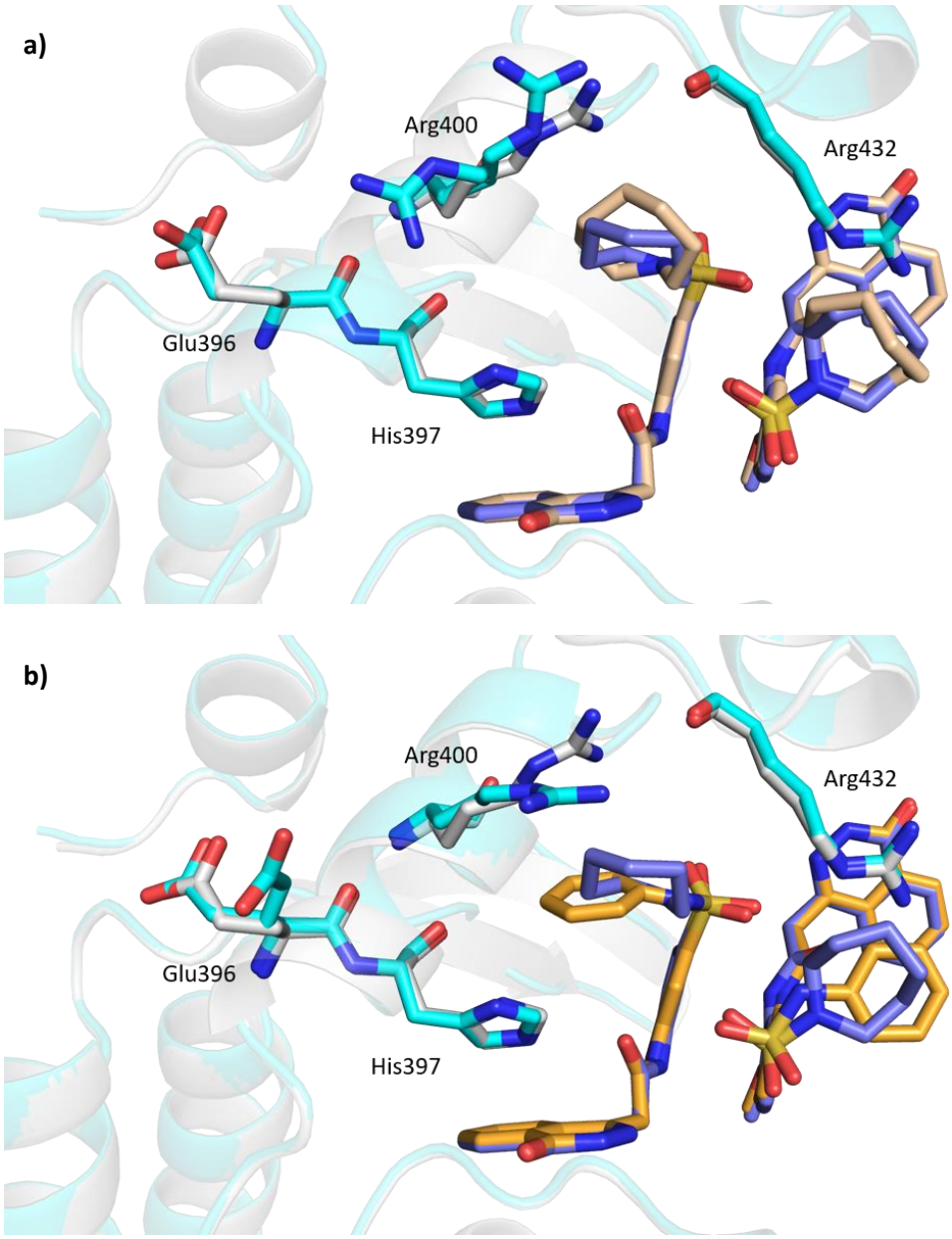
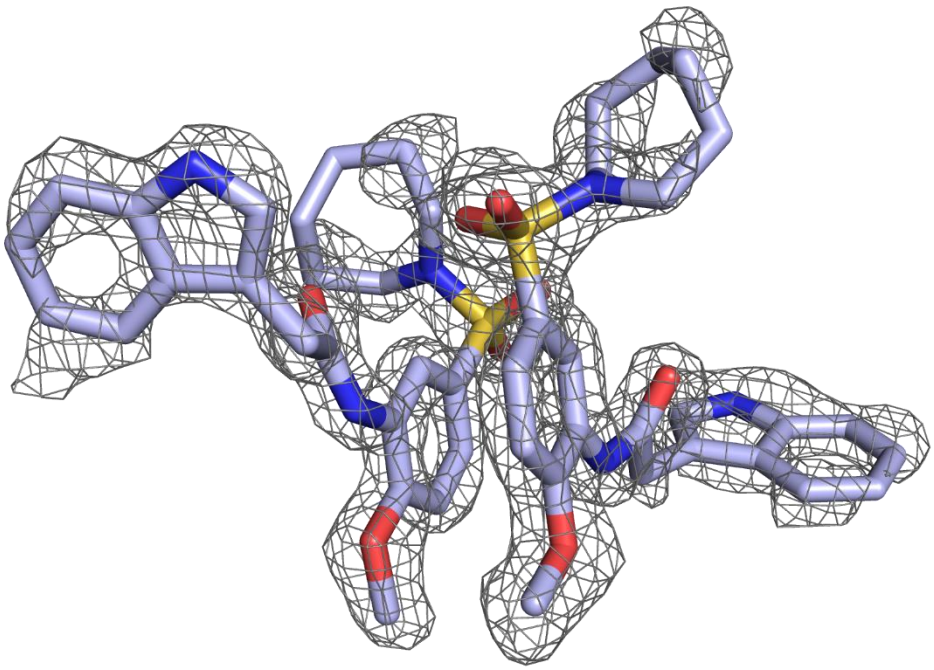


Figure S11: Omit F_o-F_c electron density maps (gray mesh) contoured to 2σ for ligands a) **15a** and b) **15g** from the X-ray crystal structures in complex with *Mtb* fumarase.

a)



b)

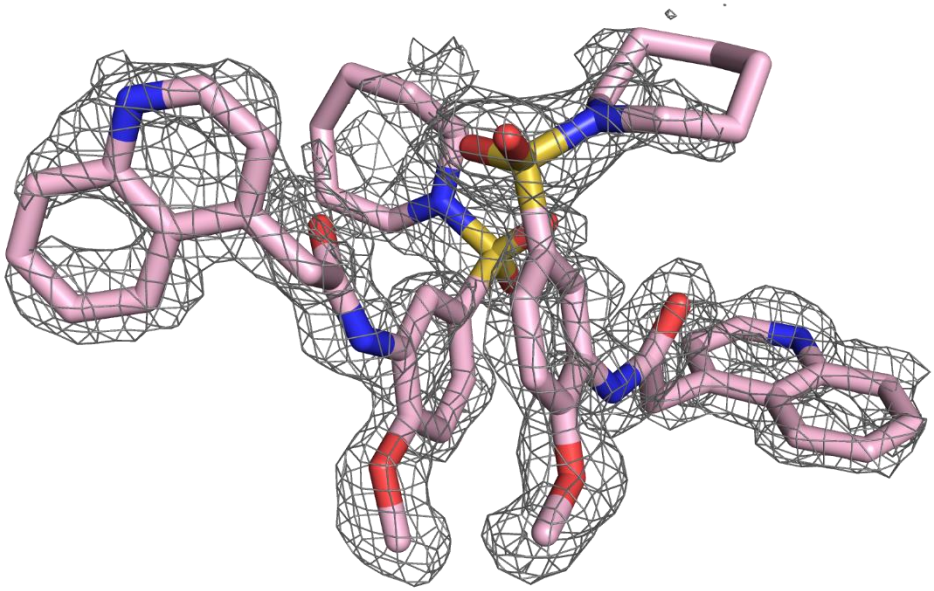
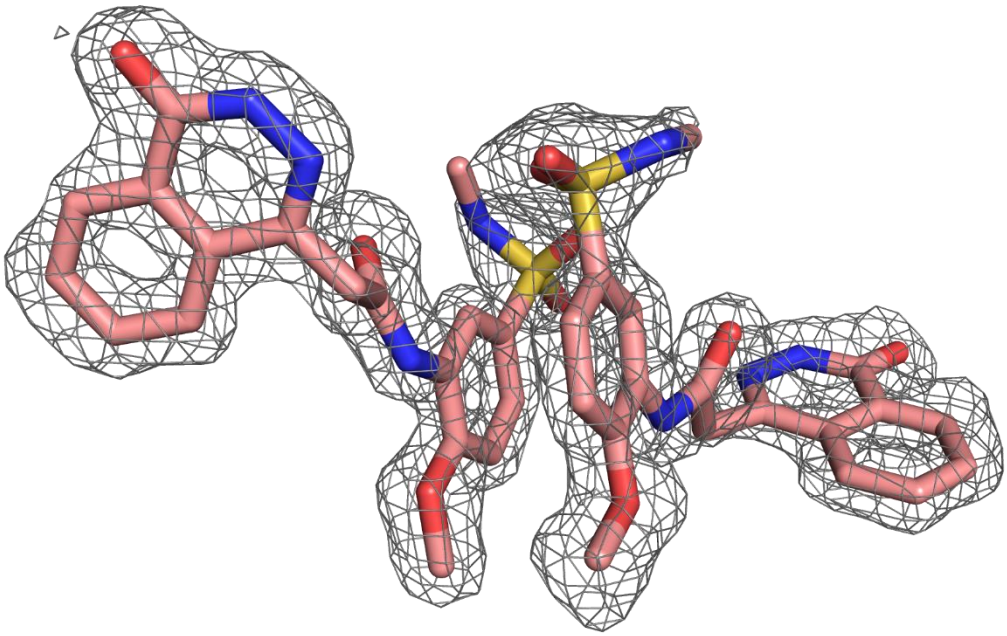


Figure S12: Omit F_o-F_c electron density maps (gray mesh) contoured to 2σ for ligands a) **16a** and b) **16b** from the X-ray crystal structures in complex with *Mtb* fumarase.

a)



b)

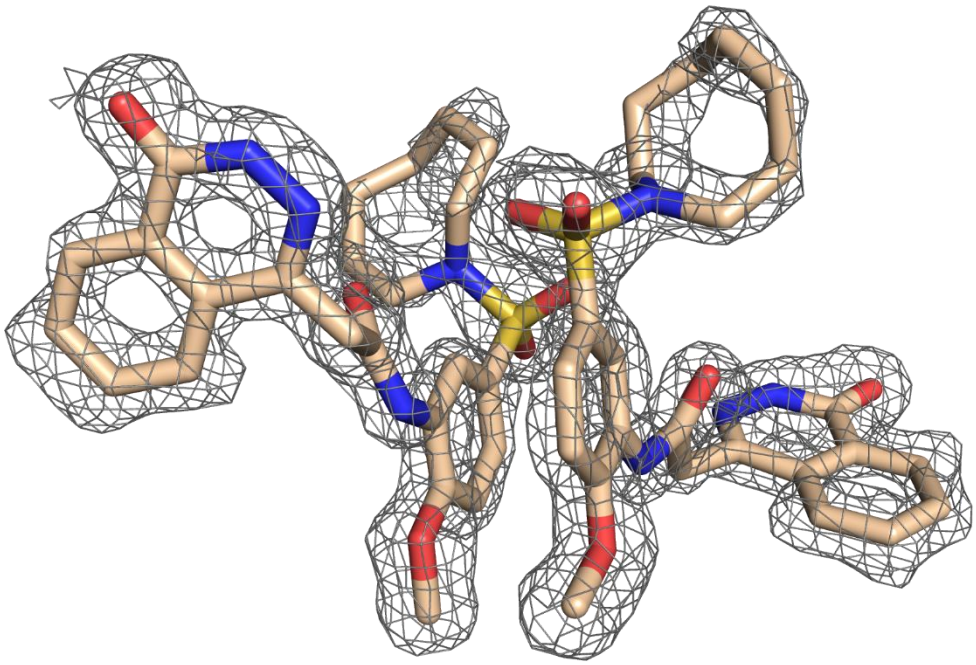
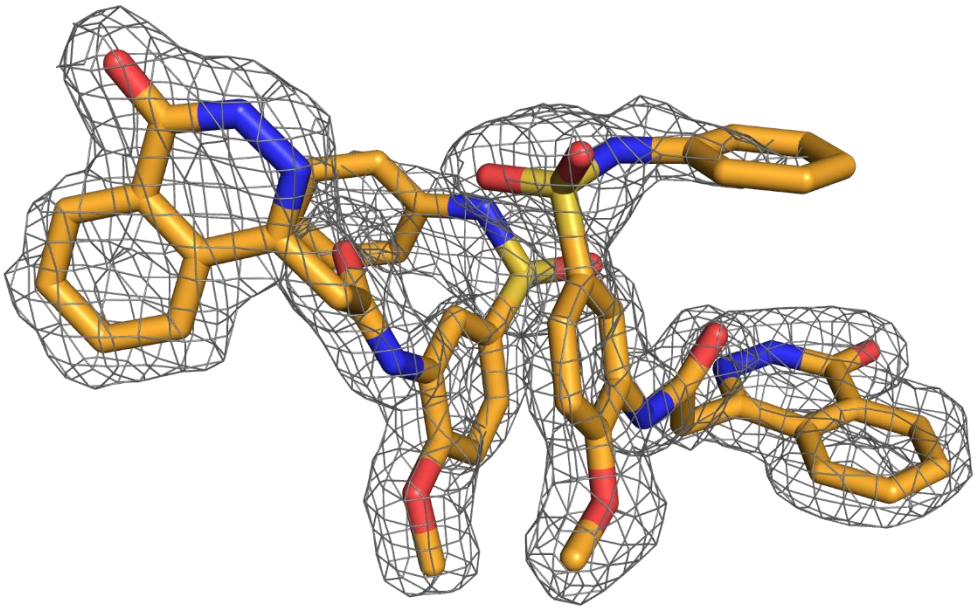


Figure S13: Omit F_o-F_c electron density maps (gray mesh) contoured to 2σ for ligands a) **16h** and b) **16j** from the X-ray crystal structures in complex with *Mtb* fumarase.

a)



b)

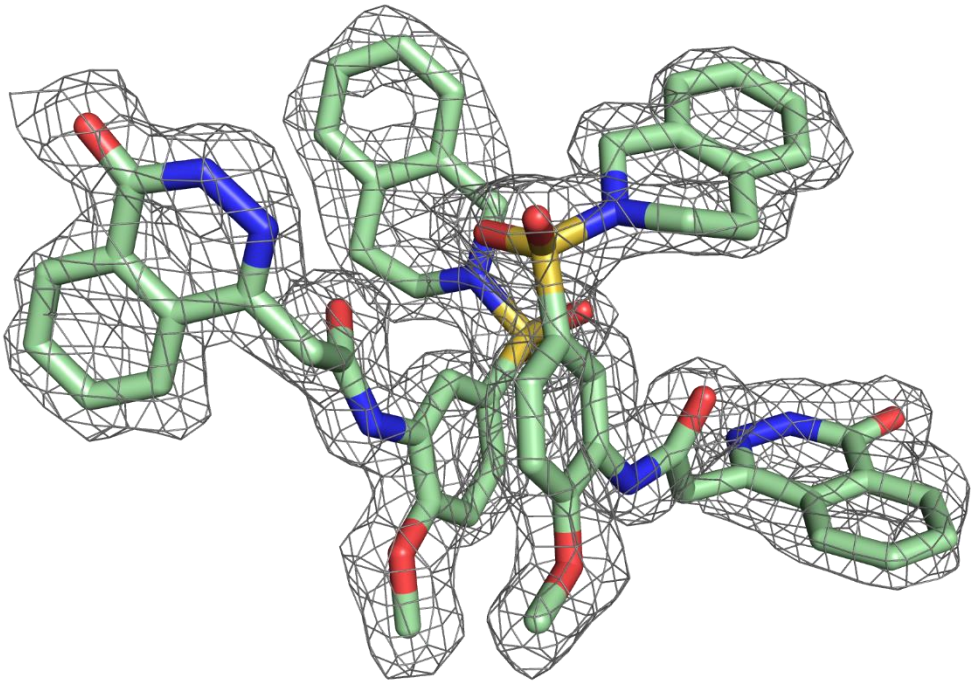


Figure S14: Omit F_o-F_c electron density map (gray mesh) contoured to 2σ for ligand **16l** from the X-ray crystal structure in complex with *Mtb* fumarase.

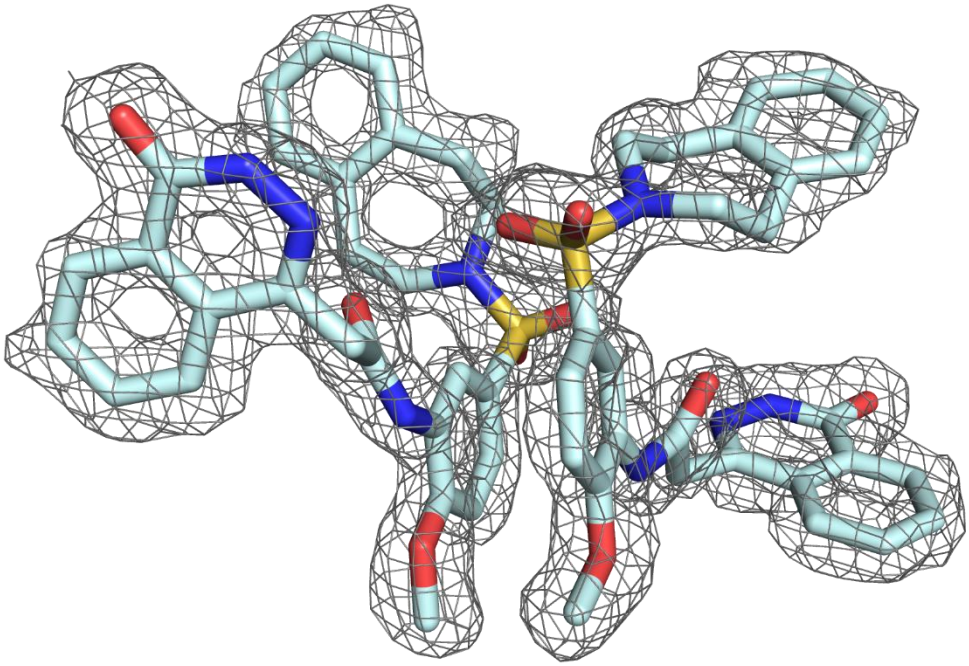


Table S1: Protein Data Bank accession numbers and X-ray crystallographic data collection and refinement statistics for crystal structures described in this study.

Ligand	15a	15g	16a	16b
PDB Code	6S7U	6S7W	6S7K	6S43
Synchrotron	Diamond Light Source	Diamond Light Source	Diamond Light Source	Diamond Light Source
Beamline	I03	I03	I03	I03
Collection Date	24/09/2018	21/10/2018	24/09/2018	24/09/2018
Wavelength (Å)	0.9763	0.9762	0.9763	0.9763
Resolution range	85.47 - 1.48 (1.533 - 1.48)	48.97 - 1.44 (1.491 - 1.44)	48.67 - 1.552 (1.607 - 1.552)	63.61 - 1.424 (1.475 - 1.424)
Space group	C 1 2 1	C 1 2 1	C 1 2 1	C 1 2 1
Unit cell	175.258 96.662 124.259 90 102.746 90	175.682 96.963 124.736 90 102.706 90	174.669 96.388 123.707 90 102.655 90	175.313 96.567 124.349 90 102.701 90
Total reflections	2395184 (366843)	1836514 (276801)	1955581 (307235)	2627362 (403102)
Unique reflections	322623 (33323)	349365 (35951)	269929 (28745)	359386 (37498)
Multiplicity	7.4 (7.5)	5.2 (5.2)	7.2 (7.3)	7.3 (7.4)
Completeness (%)	96.17 (99.80)	91.55 (86.20)	93.67 (99.90)	95.48 (99.97)
Mean I/sigma(I)	8.4 (1.1)	6.8 (0.9)	9.5 (1.1)	9.6 (1.1)
Wilson B-factor	16.95	12.88	19	16.99
R-merge	0.141 (1.856)	0.131 (2.001)	0.118 (1.626)	0.106 (1.682)
R-meas	0.151 (1.993)	0.146 (2.248)	0.127 (1.753)	0.114 (1.811)
R-pim	0.055 (0.719)	0.064 (1.004)	0.047 (0.647)	0.042 (0.663)
CC1/2	0.998 (0.442)	0.995 (0.806)	0.998 (0.449)	0.998 (0.465)
Reflections used in refinement	322588 (33319)	336514 (31625)	269859 (28716)	359347 (37494)
Reflections used for R-free	16113 (1588)	16825 (1562)	13407 (1433)	17870 (1773)
R-work	0.1700 (0.2912)	0.2069 (0.5116)	0.1695 (0.3211)	0.1669 (0.3075)
R-free	0.1941 (0.2855)	0.2256 (0.5191)	0.1934 (0.3377)	0.1878 (0.3061)
Number of non-hydrogen atoms	15777	15336	15611	15622
macromolecules	14014	13882	14060	13888
ligands	67	68	61	73
solvent	1696	1386	1490	1661
Protein residues	1805	1807	1808	1805
RMS(bonds)	0.016	0.016	0.015	0.016
RMS(angles)	1.88	1.9	1.85	1.88
Ramachandran favored (%)	97.54	97.87	97.65	97.76
Ramachandran allowed (%)	2.35	2.13	2.23	2.24
Ramachandran outliers (%)	0.11	0	0.11	0
Rotamer outliers (%)	1.03	1.18	1.3	0.97
Clashscore	2.57	2.42	3.3	3.06
Average B-factor	24.36	23.58	25.41	24.16
macromolecules	23.28	22.85	24.63	23.15
ligands	28.42	23.87	21.93	24.72
solvent	33.11	30.82	32.97	32.57

Ligand	16h	16j	16l
PDB Code	6S7S	6S7Z	6S88
Synchrotron	Diamond Light Source	Diamond Light Source	Diamond Light Source
Beamline	I03	I04	I03
Collection Date	24/09/2018	17/09/2018	21/10/2018
Wavelength (Å)	0.9763	0.9795	0.9762
Resolution range	48.96 - 1.7 (1.761 - 1.7)	52.14 - 1.85 (1.916 - 1.85)	46.98 - 1.59 (1.647 - 1.59)
Space group	C 1 2 1	C 1 2 1	C 1 2 1
Unit cell	175.566 96.924 124.753 90 102.858 90	175.465 97.132 124.78 90 102.744 90	175.71 96.775 124.796 90 102.745 90
Total reflections	1365156 (210332)	915578 (105262)	1357807 (225699)
Unique reflections	190941 (22175)	172935 (16430)	244289 (26216)
Multiplicity	7.1 (6.4)	5.3 (4.3)	5.5 (5.8)
Completeness (%)	84.65 (99.30)	99.20 (93.56)	89.04 (96.36)
Mean I/sigma(I)	10.3 (0.9)	6.8 (1.2)	8.0 (1.2)
Wilson B-factor	22.7	21.18	19.45
R-merge	0.119 (1.874)	0.185 (1.187)	0.123 (1.431)
R-meas	0.129 (2.039)	0.206 (1.349)	0.137 (1.572)
R-pim	0.048 (0.791)	0.088 (0.622)	0.058 (0.645)
CC1/2	0.999 (0.412)	0.991 (0.477)	0.997 (0.434)
Reflections used in refinement	189157 (22140)	172623 (16212)	243036 (26216)
Reflections used for R-free	9479 (1118)	8588 (794)	12256 (1309)
R-work	0.1835 (0.3330)	0.1960 (0.3629)	0.1782 (0.2936)
R-free	0.2136 (0.3348)	0.2288 (0.3801)	0.1999 (0.3073)
Number of non-hydrogen atoms	15135	15112	15219
macromolecules	13859	13811	13829
ligands	69	76	78
solvent	1207	1225	1312
Protein residues	1807	1804	1807
RMS(bonds)	0.015	0.016	0.015
RMS(angles)	1.82	1.87	1.84
Ramachandran favored (%)	97.48	97.81	97.32
Ramachandran allowed (%)	2.35	2.19	2.63
Ramachandran outliers (%)	0.17	0	0.06
Rotamer outliers (%)	1.39	0.91	0.84
Clashscore	2.71	3.19	2.72
Average B-factor	31.45	27.05	26.51
macromolecules	31.08	26.74	26.01
ligands	29.47	22.54	21.9
solvent	35.75	30.81	32.14