Inhibitors of histone acetyltransferases KAT6A/B induce senescence and arrest tumor growth

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Methods

General Chemistry Methods

Experimental reagents

All non-aqueous reactions were performed under an atmosphere of dry nitrogen, unless otherwise specified. Solvents were of analytical grade: ethyl acetate (EtOAc); dichloromethane (DCM); dimethylformamide (DMF); acetonitrile (MeCN); tetrahydrofuran (THF); petroleum benzine (PB); and dioxane. TLC was performed on silica gel $60F_{254}$ pre-coated aluminum sheets (0.25 mm, Merck). Flash column chromatography was carried out using Merck silica gel 60, 0.63 – 0.20 mm (70-230 mesh). Automated flash column chromatography was performed with a Biotage Isolera One instrument using Biotage SNAP cartridges packed with silica (KP-SilTM).

Nuclear Magnetic Resonance

^{1}H and ^{13}C Nuclear Magnetic Resonance (NMR) spectra were recorded at 400.13 Hz on an Avance III Nanobay 400 MHz Bruker spectrometer (^{1}H at 400.13 MHz and ^{13}C at 101 MHz, respectively) coupled to the BACS 60 automatic sample changer. Results are recorded as follows: chemical shifts (δ) in ppm acquired in either $CDCl_3$ (7.26 ppm for ^{1}H) or DMSO-d_6 (2.50 ppm for ^{1}H) as a reference. Solvents used for NMR studies are from Cambridge Isotope Laboratories. Each proton resonance was assigned according to the following convention: chemical shift (δ), multiplicity, coupling constant (*J* Hz) and number of protons. In reporting of the spectral data, the following abbreviations are utilised: s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet.

Low resolution mass spectrometry analyses

Low resolution mass spectrometry (LCMS) analyses were performed on one of the two following instruments.

(1) LCMS-A: Agilent 6100 Series Single Quad LC/MS coupled with an Agilent 1200 Series HPLC, 1200 Series G1311A quaternary pump, 1200 series G1329A thermostatted autosampler and 1200 series G1314B variable wavelength detector. The conditions for liquid chromatography were: reverse phase HPLC analysis fitted with a Phenomenex Luna C8(2) 5 µm (50 x 4.6 mm) 100 Å column; column temperature: 30°C; injection volume: 5 µl; solvent: 99.9% acetonitrile, 0.1% formic acid; gradient: 5-100% of solvent over 10 min; detection: 254 nm. The conditions for mass spectrometry were: quadrupole ion source; ion mode: multimode-ES; drying gas temp: 300°C; vaporizer temperature: 200°C; capillary voltage:

2000 V (positive), 4000 V (negative); scan range: 100-1000 *m/z*; step size: 0.1 sec; acquisition time: 10 min.

(2) LCMS-B: Agilent 6224 TOF LC/MS Mass Spectrometer coupled to an Agilent 1290 Infinity (Agilent, Palo Alto, CA). All data were acquired and reference mass corrected via a dual-spray electrospray ionisation (ESI) source. Each scan or data point on the Total Ion Chromatogram (TIC) is an average of 13,700 transients, producing a spectrum every second. Mass spectra were created by averaging the scans across each peak and background subtracted against the first 10 seconds of the TIC. Acquisition was performed using the Agilent Mass Hunter Data Acquisition software version B.05.00 Build 5.0.5042.2 and analysis was performed using Mass Hunter Qualitative Analysis version B.05.00 Build 5.0.519.13. Chromatographic separation was performed using an Agilent Zorbax SB-C18 Rapid Resolution HT 2.1 x 50 mm, 1.8 μm column (Agilent Technologies, Palo Alto, CA) using an acetonitrile gradient (5% to 100%) over 3.5 min at 0.5 ml/min.

High resolution mass spectrometry

High resolution mass spectrometry was performed with an Agilent 6224 TOF LC/MS coupled to an Agilent 1290 Infinity LC. All data were acquired and reference mass corrected via a dual-spray electrospray ionisation (ESI) source. Each scan or data point on the total ion chromatogram (TIC) is an average of 13,700 transients, producing a spectrum every second. Mass spectra were created by averaging the scans across each peak and subtracting the background from the first 10 s of the TIC. Acquisition was performed using the Agilent Mass Hunter Data Acquisition software ver. B.05.00 Build 5.0.5042.2 and analysis was performed using Mass Hunter Qualitative Analysis ver. B.05.00 Build 5.0.519.13. Acquisition parameters: mode, ESI; drying gas flow, 11 l/min; nebuliser pressure, 45 psi; drying gas temperature, 325°C; voltages: capillary, 4000 V; fragmentor, 160 V; skimmer, 65 V; octapole RF, 750 V; scan range, 100–1500 m/z; positive ion mode internal reference ions, m/z 121.050873 and 922.009798. LC conditions: Agilent Zorbax SB-C18 Rapid Resolution HT (2.1 × 50 mm, 1.8 mm column), 30°C; sample 13 (5 μl) was eluted using a binary gradient (solvent A: 0.1% aq. HCO_2H; solvent B: 0.1% HCO_2H in CH_3CN; 5–100% B [3.5 min], 0.5 ml/min).

Analytical high-performance liquid chromatography

Analytical HPLC was acquired on an Agilent 1260 Infinity analytical HPLC coupled with a G1322A degasser, G1312B binary pump, G1367E high-performance autosampler, and G4212B diode array detector. Conditions were as follows: Zorbax Eclipse Plus C18 rapid

resolution column (4.6 × 100 mm) with UV detection at 254 and 214 nm, 30°C; the sample was eluted using a gradient of 5−100% solvent B in solvent A, where solvent A was 0.1% aq. TFA and solvent B was 0.1% TFA in CH_3CN (5−100% B [9 min], 100% B [1min]; 0.5 ml/min).

Experimental Procedures Chemistry

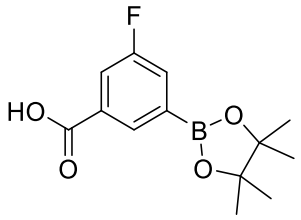
Copies of all spectral data, including intermediates can be found in the Supplementary Information.

General Method A: Amide Coupling

To a solution of carboxylic acid (1 eq.) in ACN (0.08 M) was added EDCI.HCl (1.2 eq.) and HOAt (1.2 eq.) at room temperature (r.t.). The mixture was heated to 50^{o}C and after 10 min, the arylsulfonohydrazide (1.2 eq.) was added. The mixture was allowed to stir at this temperature overnight. The reaction was then cooled to r.t. and concentrated *in vacuo*. The residue was partitioned between H_2O and EtOAc, and the layers separated. The aqueous layer was further washed with EtOAc (2x). The combined organic layers were washed with brine, dried over $MgSO_4$, and concentrated *in vacuo*. The crude material was purified by silica gel chromatography (Isolera Biotage, 0-50% EtOAc/petroleum benzine). Product-containing fractions were combined and concentrated *in vacuo* to give the title compound.

Preparation of WM-1119

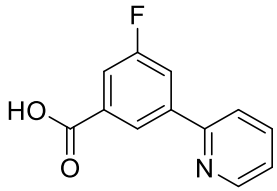
3-Fluoro-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)benzoic acid



To a degassed solution of 1,4-dioxane (220 ml), under an atmosphere of nitrogen, was added 3-bromo-5-fluorobenzoic acid (4.9 g, 22.4 mmol, 1 eq.), bis(pinacolato)diboron (8.52 g, 33.6 mmol, 1.5 eq.) and KOAc (9.66 g, 98.4 mmol, 4.4 eq.) sequentially. The mixture was degassed once again, then $Pd(dppf)Cl_2$ (0.82 g, 1.12 mmol, 0.05 eq.) was added. The resulting solution was heated to 110^{o}C overnight. The solvent was removed *in vacuo* to give a dark gummy residue which was taken up into EtOAc and H_2O. The organic layer was separated and the aqueous phase further extracted with EtOAc (2x). The combined organic

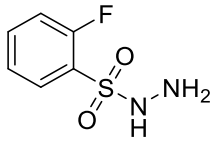
layers were washed with 2 M HCl, dried over $MgSO_4$ and concentrated *in vacuo* to give a dark brown solid. The resulting solid was triturated with petroleum benzine to give the title compound as a brown solid (5.34 g, 90% yield). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.34 (s, 1H), 7.86 (ddd, *J* = 9.0, 2.7, 1.5 Hz, 1H), 7.72 (dd, *J* = 8.4, 2.6, 1H), 1.37 (s, 12H), carboxylic acid proton not observed. LCMS B: Rt 3.86 min, *m/z* 267.2 $[M + H]^+$. Spectral data in agreement with reported literature values.[1]

3-Fluoro-5-(pyridin-2-yl)benzoic acid



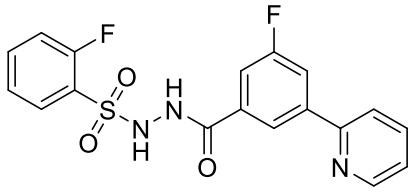
To a degassed solution of DMF/H_2O (10:1) (12.5 ml), under an atmosphere of nitrogen, was added 3-fluoro-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)benzoic acid (1.00 g, 3.76 mmol, 1 eq.), 2-bromopyridine (0.54 ml, 5.64 mmol, 1.5 eq.) and Cs_2CO_3 (5.39 g, 16.5 mmol, 4.4 eq.). The mixture was again degassed and $Pd(PPh_3)_4$ (0.22 g, 0.19 mmol, 0.05 eq.) was added. The resulting solution was heated to 110^{o}C overnight. The solvent was removed *in vacuo* to give a dark gummy residue which was dissolved with H_2O, then acidified with 10% w/v citric acid to ~pH 4. The resulting precipitate was collected by filtration, washed with H_2O (2x) and dried to give the title compound as a beige solid (0.44 g, 54% yield). ^{1}H-NMR (400 MHz, DMSO) δ 8.72 (ddd, *J* = 4.8, 1.7, 0.9 Hz, 1H), 8.55 (t, *J* = 1.5 Hz, 1H), 8.18 (ddd, *J* = 10.1, 2.5, 1.6 Hz, 1H), 8.11 (d, *J* = 8.0 Hz, 1H), 7.95 (td, *J* = 7.8, 1.8 Hz, 1H), 7.73 (ddd, *J* = 8.9, 2.5, 1.4 Hz, 1H), 7.45 (ddd, *J* = 7.5, 4.8, 0.9 Hz, 1H), carboxylic acid proton not observed. ^{13}C-NMR (101 MHz, DMSO) δ 166.6 (J_{CF} = 3.0 Hz), 162.4 (J_{CF} = 244.4 Hz), 154.0 (J_{CF} = 3.0 Hz), 149.7, 141.0 (J_{CF} = 8.1 Hz), 137.5, 136.6 (J_{CF} = 6.1 Hz), 123.4, 123.2 (J_{CF} = 2.0 Hz), 120.6, 116.2 (J_{CF} = 21.2 Hz), 116.0 (J_{CF} = 22.2 Hz). ^{19}F-NMR (376 MHz, DMSO) δ -112.8. LCMS-B: Rt 3.41 min, *m/z* 218.1 $[M + H]^+$. HRMS (ESI): *m/z* $[M + H]^+$ for $C_{12}H_9FNO_2^+$ calculated 218.0612, found 218.0615. HPLC purity >95%, Rt 3.60 min.

2-Fluorobenzenesulfonohydrazide



To a solution of 2-fluorobenzenesulfonyl chloride (4.0 g, 20.6 mmol, 1 eq.) in DCM (20 ml) at 0°C was added a solution of hydrazine monohydrate (19.9 ml, 411 mmol, 20 eq.) in DCM (20 ml). The reaction was allowed to stir for 3 h at r.t., upon which time H_2O was added. The layers were separated and the organics were washed further with H_2O (2x), brine (2x), dried with $MgSO_4$ and concentrated *in vacuo* to give the title compound as a pale yellow solid (2.8 g, 72%). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.65 (s, 1H), 7.79 (td, J = 7.6, 1.8 Hz, 1H), 7.75 – 7.67 (m, 1H), 7.40 (m, 2H), 4.33 (br s, 2H). ^{13}C NMR (101 MHz, DMSO) δ 158.6 (J_{CF} = 254.5 Hz), 135.4 (J_{CF} = 9.1 Hz), 131.2, 126.1 (J_{CF} = 14.1 Hz), 124.7 (J_{CF} = 4.0 Hz), 117.2 (J_{CF} = 21.2 Hz). ^{19}F NMR (376 MHz, DMSO) δ -109.63. LCMS-B: Rt 4.78 min, *m/z* 158.9 [M-$NHNH_2$]$^-$. HRMS (ESI): *m/z* [M – ($NHNH_2$)+ H]$^+$ for $C_6H_6FO_2S^+$ calculated 161.0067, found 161.0066.

***N*'-(3-Fluoro-5-(pyridin-2-yl)benzoyl)-2-fluorobenzenesulfonohydrazide, WM-1119**

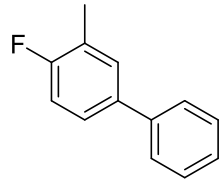


N'-(3-Fluoro-5-(pyridin-2-yl)benzoyl)-2-fluorobenzenesulfonohydrazide, **WM-1119**, was prepared as per general method **A**, from 3-fluoro-5-(pyridin-2-yl)benzoic acid (0.30 g, 1.38 mmol) and 2-fluorobenzenesulfonohydrazide (0.30 g, 1.66 mmol), to give a colourless solid (0.3 g, 56% yield). ^{1}H NMR (400 MHz, DMSO) δ 10.98 (br s, 1H), 10.43 (br s, 1H), 8.71 (d, J = 4.4 Hz, 1H), 8.31 (s, 1H), 8.13 – 8.02 (m, 2H), 7.96 (t, J = 7.7 Hz, 1H), 7.82 (t, J = 7.3 Hz, 1H), 7.70 (dd, J = 12.7, 7.3 Hz, 1H), 7.54 (d, J = 9.8 Hz, 1H), 7.43 (dd, J = 17.4, 7.9 Hz, 2H), 7.31 (t, J = 7.6 Hz, 1H). ^{13}C-NMR (101 MHz, DMSO) δ 164.73, 162.8 (J_{CF} = 245.4 Hz), 159.6 (J_{CF} = 256.5 Hz), 154.1, 150.2, 142.1 (J_{CF} = 7.1 Hz), 138.0, 136.3 (J_{CF} = 8.1 Hz), 134.9 (J_{CF} = 7.1 Hz), 130.8, 127.9 (J_{CF} = 14.1 Hz), 124.8 (J_{CF} = 3.0 Hz), 124.2, 122.0, 121.3, 117.7 (J_{CF} = 21.2 Hz), 117.1 (J_{CF} = 23.2 Hz), 115.1 (J_{CF} = 23.2 Hz). ^{19}F NMR (376 MHz, DMSO)

δ -107.10, -111.87. LCMS B Rt 3.49 min, *m/z* 390.1 $[M + H]^+$. HRMS (ESI): *m/z* $[M + H]^+$ for $C_{18}H_{14}F_2N_3O_3S^+$ calculated 390.0718, found 390.0721. HPLC purity >98%, Rt 4.69 min.

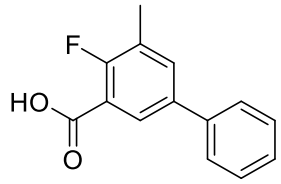
Preparation of WM-8014

4-Fluoro-3-methyl-1,1'-biphenyl



4-Bromo-1-fluoro-2-methylbenzene (6.00 g, 31.7 mmol, 1.0 eq), phenylboronic acid (4.26 g, 34.9 mmol, 1.1 eq), K_2CO_3 (6.58 g, 47.6 mmol, 1.5 eq) and $PdCl_2(PPh_3)_2$ (1.11 g, 1.59 mmol, 0.05 eq.), under a nitrogen atmosphere, were suspended in THF/H_2O (1:1) (130 ml). The reaction was heated to reflux and stirred overnight. The mixture was cooled to r.t. and separated between EtOAc and H_2O. The organic layer was washed with H_2O, brine, dried over Na_2SO_4, filtered, and concentrated *in vacuo*. The crude product was purified by flash column chromatography using 0-10% EtOAc/petroleum benzine to give the title compound as a colourless oil (5.91 g, quantitative yield). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.58 – 7.51 (m, 2H), 7.47 – 7.31 (m, 5H), 7.11 – 7.03 (m, 1H), 2.35 (d, J = 1.9 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 162.6 (J_{CF} = 245.4 Hz), 140.5, 137.1 (J_{CF} = 3.0 Hz), 130.2 (J_{CF} = 5.1 Hz), 128.8, 127.1, 127.0, 126.0 (J_{CF} = 8.1 Hz), 125.0 (J_{CF} = 17.2 Hz), 115.2 (J_{CF} = 23.2 Hz), 14.7 (J_{CF} = 4.0 Hz). ^{19}F NMR (376 MHz, $CDCl_3$) δ -120.15. This compound does not ionize and could not be detected by mass spectrometry.

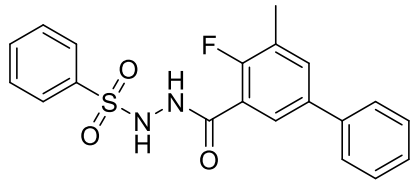
4-Fluoro-5-methyl-[1,1'-biphenyl]-3-carboxylic acid



To a solution of 4-fluoro-3-methyl-1,1'-biphenyl (5.16 g, 27.7 mmol, 1 eq.) in THF (28 ml) at -78°C was added a 2.5 M solution of *n*-butyllithium in hexanes (12.2 ml, 30.5 mmol, 1.1 eq). The reaction mixture was allowed to stir at -78°C for 40 min before being poured onto a vast excess of dry ice. After the majority of dry ice had evaporated, the mixture was

concentrated *in vacuo*. The resulting solid was suspended in a small amount of H_2O then acidified with 1 M HCl. The solid was filtered, washed with 1 M HCl, petroleum benzine and air dried to give the title compound as a pale yellow solid (2.09 g, 33% yield). ^{1}H-NMR (400 MHz, DMSO) δ 13.29 (br s, 1H), 7.88 (dd, *J* = 6.4, 2.4 Hz, 1H), 7.83 (dd, *J* = 6.4, 2.0 Hz, 1H), 7.72 – 7.61 (m, 2H), 7.49-7.45 (m, 2H), 7.40-7.37 (m, 1H), 2.34 (d, *J* = 2.0 Hz, 3H). ^{13}C NMR (101 MHz, DMSO) δ 165.2 (3.0 Hz), 159.0 (J_{CF} = 257.6 Hz), 138.8 (J_{CF} = 4.0 Hz), 135.7, 133.8 (J_{CF} = 6.1 Hz), 129.0, 127.7, 127.1, 126.6, 126.5, 119.6 (J_{CF} = 12.1 Hz), 14.3 (J_{CF} = 4.0 Hz). ^{19}F-NMR (376 MHz, DMSO) δ -117.28. LCMS A: Rt 6.41 min, *m/z* 229.3 [M - H]$^-$. HRMS (ESI): *m/z* [M + H]$^+$ for $C_{14}H_{12}FO_2^+$ calculated 231.0816, found 231.0818. HPLC purity >95%, Rt 6.24 min.

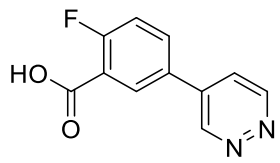
_N_'-(4-Fluoro-5-methyl-[1,1'-biphenyl]-3-carbonyl)benzenesulfonohydrazide, WM-8014



N'-(4-Fluoro-5-methyl-[1,1'-biphenyl]-3-carbonyl)benzenesulfonohydrazide, **WM-8014**, was prepared as per general method **A**, from 4-fluoro-5-methyl-[1,1'-biphenyl]-3-carboxylic acid (68 mg, 0.30 mmol) and commercially available benzenesulfonohydrazide (68.5 mg, 0.36 mmol), to give a colourless solid (39.1 mg, 35% yield). ^{1}H-NMR (400 MHz, DMSO) δ 10.64 (br s, 1H), 10.16 (br s, 1H), 7.89 (d, *J* = 7.3 Hz, 2H), 7.72 (dd, *J* = 6.6, 1.7 Hz, 1H), 7.69 – 7.60 (m, 3H), 7.57 (t, *J* = 7.5 Hz, 2H), 7.48 (t, *J* = 7.6 Hz, 2H), 7.44 – 7.35 (m, 2H), 2.30 (s, 3H). ^{13}C-NMR (101 MHz, DMSO) δ 162.9, 158.4 (J_{CF} = 252.5 Hz), 138.9, 138.5, 135.9 (J_{CF} = 4.0 Hz), 133.1, 132.3 (J_{CF} = 5.1 Hz), 129.0, 128.8, 127.7, 126.6, 126.1, 125.8, 125.1 (J_{CF} = 2.0 Hz), 121.9 (J_{CF} = 16.2 Hz), 14.3 (J_{CF} = 4.0 Hz). ^{19}F-NMR (376 MHz, DMSO) δ -120.52. LCMS A Rt 6.48 min, *m/z* 385.1 [M + H]$^+$. HRMS (ESI): *m/z* [M + H]$^+$ for $C_{20}H_{18}FN_2O_3S^+$ calculated 385.1022, found 385.1014. HPLC purity >98%, Rt 6.71 min.

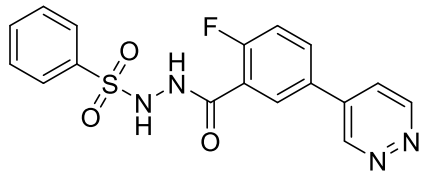
Preparation of WM-2474

2-Fluoro-5-(pyridazin-4-yl)benzoic acid



3-Carboxy-4-fluorobenzeneboronic acid (0.50 g, 2.72 mmol, 1 eq.), 4-bromo pyridazine hydrobromide salt (0.72 g, 2.99 mmol, 1.1 eq.) and $PdCl_2$(dppf) (0.05 eq.) were suspended in 1,4-dioxane (13.6 ml). A solution of K_2CO_3 (0.56 g, 4.08 mmol, 1.5 eq.) in H_2O (13.5 ml) was added and the mixture degassed. The reaction was irradiated in a CEM microwave at 120°C for 30 min. The mixture was cooled, and the volatile solvents removed *in vacuo*. The aqueous residue was diluted with H2O and shaken with DCM. The mixture was filtered through Celite, the aqueous layer separated and washed with a further portion of DCM. The organic extracts were discarded. The aqueous phase was diluted with H_2O and treated with 5% w/v citric acid. The resulting precipitate was collected by filtration, washed with H_2O and dried under vacuum to give the title compound as a tan solid (0.11 g, 21% yield). ^{1}H-NMR (400 MHz, DMSO) δ 13.55 (s, 1H), 9.66 (d, *J* = 1.4 Hz, 1H), 9.29 (d, *J* = 5.4 Hz, 1H), 8.33 (dd, *J* = 6.8, 2.5 Hz, 1H), 8.20 (ddd, *J* = 8.5, 4.4, 2.6 Hz, 1H), 8.06 (dd, *J* = 5.5, 2.5 Hz, 1H), 7.54 (dd, *J* = 10.5, 8.7 Hz, 1H). ^{13}C-NMR (101 MHz, DMSO) δ 164.6 (J_{CF} = 3.0 Hz), 162.0 (J_{CF} = 261.6 Hz), 151.6, 149.3, 135.5, 133.4 (J_{CF} = 10.1 Hz), 130.7 (J_{CF} = 2.0 Hz), 130.5 (J_{CF} = 4.0 Hz), 123.4, 120.6 (J_{CF} = 11.1 Hz), 118.2 (J_{CF} = 23.2). ^{19}F-NMR (376 MHz, DMSO) δ -109.1. LCMS B: Rt 3.12 min, *m/z* 219.1 $[M + H]^+$. HRMS (ESI): *m/z* $[M + H]^+$ for $C_{11}H_8FN_2O_2^+$ calculated 219.0564, found 219.0567. HPLC purity >95%, Rt 3.43 min.

***N*'-(2-Fluoro-5-(pyridazin-4-yl)benzoyl)benzenesulfonohydrazide, WM-2474**



N'-(2-Fluoro-5-(pyridazin-4-yl)benzoyl)benzenesulfonohydrazide, **WM-2474**, was prepared as per general method **A**, from 2-fluoro-5-(pyridazin-4-yl)benzoic acid (65 mg, 0.29 mmol) and commercially available benzenesulfonohydrazide (62 mg, 0.36 mmol), to give a pale

brown solid (47.7 mg, 43% yield). ^{1}H-NMR (400 MHz, DMSO) δ 10.72 (br s, 1H), 10.20 (br s, 1H), 9.69 (dd, *J* = 2.5, 1.2 Hz, 1H), 9.34 (dd, *J* = 5.5, 1.2 Hz, 1H), 8.08 (dd, *J* = 5.5, 2.5 Hz, 1H), 7.94 (dd, *J* = 11.2, 1.7 Hz, 1H), 7.91 – 7.87 (m, 2H), 7.84 (dd, *J* = 8.1, 1.7 Hz, 1H), 7.68 – 7.63 (m, 1H), 7.61 – 7.54 (m, 3H). ^{13}C-NMR (101 MHz, DMSO) δ 162.1, 160.2 (J_{CF} = 256.4 Hz), 151.6, 149.3, 138.8, 135.6, 133.1, 132.0 (J_{CF} = 9.1 Hz), 130.4 (J_{CF} = 3.0 Hz), 128.9, 128.7 (J_{CF} = 3.0 Hz), 122.8, 123.4, 122.7 (J_{CF} = 15.1 Hz), 117.5 (J_{CF} = 22.2 Hz). ^{19}F-NMR (376 MHz, DMSO) δ -111.81. LCMS B Rt 3.31 min, *m/z* 373.2 $[M + H]^+$. HRMS (ESI): *m/z* $[M + H]^+$ for $C_{17}H_{14}FN_4O_3S^+$ calculated 373.0765, found 373.0766. HPLC purity >95%, Rt 4.59 min.

Protein Biochemistry methods

Histone acetyltransferase assay and protein purification

Compounds were tested for inhibitory activity against histone acetyltransferases using our previously reported assay format.[2] Briefly, purified enzymes were mixed with biotinylated histone substrate peptide, acetyl coenzyme A (AcCoA) at the K_M determined for each enzyme and an acetyl-lysine-specific antibody. After 60 min at r.t., Alphascreen General IgG Protein A beads (PerkinElmer, Waltham, MA) were added and, after a further incubation of at least 2 h, Alphascreen signal read on an Envision 2013 plate reader in HTS mode. The MYST domains containing the enzyme activity were expressed and purified in-house (KAT5, KAT6A, KAT8). KAT6A was expressed as a NusA fusion protein as described previously,[2] KAT5 and KAT8 were His-tagged and expressed in *E. coli*. For crystallization studies a modified MYST domain ($MYST^{Cryst}$) was used, in which the amino acids in the AcCoA binding pocket are derived from the KAT6A sequence while surface amino acids (based on the KAT8 sequence) were modified to increase solubility (Extended Data Fig. 1). $MYST^{Cryst}$ was His-tagged and expressed in *E. coli*. Additional lysine acetyltransferases were purchased from SignalChem (Richmond, Canada; KAT2A (K311-381G), KAT3A (K312-381G), KAT3B (P07-31G), KAT6B (K315-381BG), KAT7 (K316-380G)) and Millipore (Billerica, MA) (KAT2B). Signals from samples in the presence of compound dilutions were normalized to negative and positive control samples (DMSO only and no AcCoA, respectively) and the data fitted using a four-parameter logistic model to determine an IC_{50}.

Surface plasmon resonance (SPR) binding assays

Direct SPR assay for the verification of binding of small molecule inhibitors to immobilized KAT6A protein was previously described.[2] This method utilizes biotinylated KAT protein stably captured on streptavidin-coated SPR chips. Briefly, minimal biotinylation of a KAT

protein was typically achieved by mixing 100 nmol of protein (1 eq.) with 75 nmol of freshly water-dissolved EZ-link sulfoNHS LC-LC-biotin (0.75 eq.; Thermofisher Scientific). Reaction mixture was incubated on ice for 2 h and unreacted biotin reagent removed from the mixture using size exclusion chromatography on a Superdex200 HR (10/300) column equilibrated in storage buffer (20 mM HEPES, pH 7.5, 500 mM NaCl, 5 mM DTT and 5% [v/v] glycerol). A protein peak containing biotinylated KAT protein was collected and stored in 50 µl aliquots at -80°C.

All SPR immobilization and binding experiments were performed at 25°C using Biacore T200 instrument (GE Healthcare). Immobilization of high levels of streptavidin (>8 kRU; 1 kRU = 1 ng of protein per mm^2) onto CM5 sensor chips (GE Healthcare) was performed as described.[3] Minimally biotinylated KAT5, KAT6A and KAT7 were each captured in a separate channel on a streptavidin chip with instrumental fluidics primed in SPR binding buffer (50 mM HEPES, pH 7.4, 500 mM NaCl, 0.05% [v/v] Tween 20, 5 mM DTT, 10% [w/v] glycerol, 2% [v/v] DMSO). KAT proteins (~5 µg/ml) were injected at a constant flow-rate of 2 µl/min for several minutes until SPR response increases were no longer observed - typically resulting in protein immobilization levels of >12 kRU. To determine binding interactions, compounds were analyzed using concentration response experiments, with tighter binding compounds analyzed using single-cycle kinetic experiments. Thus, compound solutions (10 mM in DMSO) were serially diluted (typically 3-fold) in SPR binding buffer and resulting concentrations injected over KAT proteins at a flow rate of 60 µl/min for 30 sec (multi-cycle kinetics) or 90 sec (single-cycle kinetics). The KAT surfaces were regenerated between each binding cycle by washing the instrument running buffer over protein surfaces until the compound fully dissociated (up to 1800 sec). Scrubber software (version 2c) was utilized for data processing and analysis (www.biologic.com.au) as described previously.[4] To determine binding kinetics of each interaction, experimental data were globally fitted to a 1:1 binding model that incorporated a mass transport component.[5] Where dissociation rates were insufficiently slow to allow fitting to a kinetic binding model, responses at equilibrium for each analyte were fitted to a 1:1 steady-state affinity model to determine K_D.

Crystallization of MYST domain in complex with WM-8014, AcCoA, or WM-1119

WM-8014 was dissolved in DMSO and incubated with MYST domain ($MYST^{Cryst}$) at 20°C for 30 min prior to crystallization. $MYST^{Cryst}$ complexed with WM-8014 was screened against the MCSG suite (Microlytic) in 200 + 200 nl sitting drops dispensed by an ARI Gryphon LCP crystallization robot (Art Robbins Instruments). Following optimization, the

highest quality crystals grew in hanging drops of 1 µl protein solution (6 mg/ml $MYST^{Cryst}$, 350 µM WM-8014, 50 mM sodium citrate pH 6.5, 50 mM NaCl, 5 mM DTT) and 1 µl precipitant solution (2 M NaCl, 0.1 M Tris pH 7, 0.22 M $MgCl_2$). Crystals of MYST domain complexed with AcCoA were obtained by isolating and soaking crystals of MYST domain with WM-8014 in fresh precipitant solution supplemented with 50 mM AcCoA (Sigma). Crystals of WM-1119 in complex with $MYST^{Cryst}$ were obtained by crystal soaking. $MYST^{Cryst}$ was crystallized at the CSIRO Collaborative Crystallisation Centre (C3) (Parkville, Victoria, Australia). 200 + 200 nl sitting drops of the protein solution (9.2 mg/ml $MYST^{Cryst}$, 50 mM sodium citrate pH 6.5, 200 mM NaCl, 5 mM DTT) and precipitant solution (0.196 M $MgCl_2$, 19.5 % PEG 3350, 0.1 M bis-tris chloride pH 5.5) were dispensed using a Crystal Phoenix robot (Art Robbins Instruments). Crystal soaking lasted 6 days and was achieved by adding 1 ul of 200 µM WM-1119 diluted in precipitant solution atop the crystallization drop. Crystals were mounted in cryoloops, transferred to a cryoprotectant solution supplemented with 20% glycerol for $MYST^{Cryst}$ in complex with WM-8014 and AcCoA, 25% glycerol for $MYST^{Cryst}$ in complex with WM-1119, and flash-cooled by rapid immersion in liquid nitrogen.

X-ray diffraction data for all crystals were collected at the Australian Synchrotron (Clayton, Victoria, Australia) using Blue-ice software.[6] Data for $MYST^{Cryst}$ in complex with WM-8014 and AcCoA were collected on the MX2 beamline and data for $MYST^{Cryst}$ in complex with WM-1119 were collected on the MX1 beamline. Data sets were acquired at 100 K using a single wavelength of 0.9537 Å. Diffraction images were integrated using XDS[7] and scaled with Aimless of the CCP4 program suite[8,9] labeling 5% of the reflections for the Rfree set.

The structure of $MYST^{cryst}$ complexed with WM-8014 was determined by molecular replacement with PHASER[10] using the structure of the human MOF catalytic domain (PDB ID: 3QAH) as the search model. The structure was refined through cycles of manual building using COOT[11] and refinement using PHENIX Refine.[12] Well-defined density was immediately apparent for the complete WM-8014 molecule, however it was not modeled until the protein structure was nearing completion. A restraint library for WM-8014 was developed through PHENIX eLBOW[13] Structure validation was monitored with MolProbity[14] and refinement statistics are shown in Supplementary Table 4. The Ramachandran plot showed that 97.3% of the residues were in allowed regions with no outliers.

The structure of MYST^{cryst} complexed with AcCoA was determined by molecular replacement with PHASER[10] using the MYST^{cryst}:WM-8014 complex structure as the search model. This structure was refined as for the MYST^{cryst}:WM-8014 complex structure. Well-defined density was immediately apparent for the complete AcCoA molecule with additional density supporting isomerism of the 3-phosphate group of the ribose ring. A restraint library for this species was developed as for WM-8014 and structure validation was monitored with MolProbity[14] and refinement statistics are shown in Supplementary Table 4. The Ramachandran plot showed that 97.4% of residues were in allowed regions with a single outlier, P764. This residue was located in an exposed loop where the electron density was not well defined.

The structure of MYST^{cryst} complexed with WM-1119 was determined by molecular replacement with PHASER[10] using a MYST^{cryst} complex structure as the search model. This structure was refined as for the MYST^{cryst}:WM-8014 complex structure. Well-defined density was immediately apparent for the complete WM-1119 molecule for which a restraint library was built as for WM-8014. Structure validation was monitored MolProbity[14] and refinement statistics are shown in Supplementary Table 5. The Ramachandran plot showed that 96.6% of residues were in allowed regions with no outliers, The coordinates for the MYST^{cryst}:WM-8014, MYST^{cryst}:AcCoA, and MYST^{cryst}:WM-1119 complex structures have been deposited in the protein data bank (PDB) with the accession codes 6BA2, 6BA4, and 6CT2, respectively.

Determination of Compound Physicochemical Parameters

Percentage binding in cell culture media.

A 1 mg/ml compound stock solution was prepared in dimethylsulfoxide (DMSO) and diluted in 50% acetonitrile/water to 100 µg/ml. Assay media composed of 10% fetal calf serum in DMEM at 37°C were spiked with each compound to a final concentration of 1000 ng/ml (final DMSO and acetonitrile concentrations of 0.2% v/v and 0.4% v/v, respectively). Spiked media samples were centrifuged (Beckman Ultracentrifuge rotor type 42.2 Ti; 223,000 x g, 37°C) for 4.2 h to separate proteins and an aliquot of protein-free supernatant was analyzed by UPLC-MS to obtain the unbound concentration ($C_{unbound}$). Total concentrations (C_{total}) were determined in non-centrifuged samples incubated similarly in parallel. The percentage bound was calculated as 100 * (C_{total} - $C_{unbound}$)/ C_{total}. The UPLC-MS method employed either a Micromass Xevo TQ or Micromass Quattro Ultima Premier coupled to an Acquity UPLC, and compound concentrations were quantified using calibration standards prepared in

blank media diluted 1:1 with Dulbecco phosphate buffered saline. Diazepam was added as an internal standard to all samples before precipitating proteins with acetonitrile and centrifuging to separate the supernatant for analysis. The analytical limit of quantitation for each compound was 0.5-10 ng/ml.

Stability in cell culture media

DMSO/acetonitrile/water solutions of each compound were spiked into cell culture media at 37°C to a concentration of 1000 ng/ml. Aliquots of each spiked sample were immediately snap-frozen in dry ice (i.e. 0 h) while the remaining samples were maintained at 37°C in a humidified incubator (under an atmosphere of 5% CO_2) over 48 h. At various time points, additional sample aliquots were removed from the incubator and immediately snap-frozen in dry ice. All samples were stored frozen at -80°C until analysis by UPLC-MS using methods analogous to those described above. The percentage remaining at 48 h was calculated relative to the samples quenched at t = 0 h.

Cell permeability

Caco-2 cells were seeded onto 0.3 cm^2 polycarbonate filter Transwell® inserts at a density of 60,000 cells per well. The transport experiment was conducted using confluent cell monolayers on days 23 or 24 post-seeding. The integrity of the cell monolayers was determined on the day of the permeability experiment by measuring the transepithelial electrical resistance (TEER) and only monolayers with TEER values of >200 $\Omega.cm^2$ were utilized. The permeability for control compounds including propranolol or ^{3}H-propranolol (high permeability marker) and ^{14}C-mannitol or lucifer yellow (low permeability markers) were assessed in the same experiment to ensure consistency with literature and in-house values. Permeability experiments were performed using Hanks balanced salt solution containing 20 mM HEPES buffer (pH 7.4) in both the apical and basolateral chambers. Donor solutions were prepared by spiking a DMSO solution of each compound into transport buffer (final DMSO concentration of 0.1% v/v) at a nominal concentration of 20 μM and equilibrating at 37°C for 4 h before centrifuging to remove any undissolved material. Samples from the donor chamber were taken at the start and end of the experiment to confirm mass balance. Compound flux was assessed over 90 min, with samples taken from the acceptor chamber at 4-5 time points. At each sample time, the volume of acceptor solution removed was replaced with blank transport buffer. Acceptor chamber concentrations were assayed for each test compound by UPLC-MS as described above, and concentrations were corrected for dilution that occurred with buffer replacement. Apparent A to B permeability

(A-B P_{app}) values were calculated using the linear portion of the flux profiles. Flux profiles were well defined and the mass balance was high (≥89%) for each compound. The P_{app} values indicate that permeability is high for CTx-0124143, WM-0550, WM-8014 and WM-1119, while the lower permeability of WM-2474 is consistent with it being the least lipophilic and having the highest polar surface area of the four compounds, although the potential role of efflux transporters has not been ruled out.

Cell Culture, Molecular and In Vivo Assays

Mice

All mice were maintained on a C57BL/6 or C57BL/6 albino background. All experiments were performed with approval from the Walter and Eliza Hall Institute's Animal Ethics Committee and according to the Australian code for the care and use of animals for scientific purposes. Mice transplanted with lymphoma cells were monitored by animal technicians daily for the duration of the experiment and lymphoma progression was monitored using bioluminescence imaging. The animals were euthanised approximately five days before the ethical endpoint would have typically been reached in this tumour model. The ethical endpoint was never reached, but is defined in the following manner: when the experimental mice display major distress and symptoms including apathy, anorexia, breathing difficulties and weight loss. Posture, movement (or absence thereof), coat condition, feed consumption and bodyweight development are used to monitor the wellbeing of the animals. Animals are euthanised, if they lose more than 10% of their initial weight and exhibit apathy, hunched posture and rough coat or exhibited loss of 15% of their initial weight on more than three consecutive days irrespective of other signs of wellbeing.

Cell culture and expression plasmids

Primary mouse embryonic fibroblasts (MEFs) were derived from E12-14 embryos of the following genotypes C57BL/6 wild-type, Fucci[15], *Trp53*$^{-/-}$ [16], *Moz*$^{lox/lox}$ [17], *Rosa26Cre*^{ERT2} [18], or *Ink4a-Arf*$^{-/-}$ (*Cdkn2a*$^{-/-}$)[19] and cultured under near physiological oxygen conditions (3% O2, 5% CO2, 92% N2).

MEFs were maintained in Dulbecco's modified Eagle's medium (DMEM) supplemented with 2 mM L-glutamine, 100 U/ml penicillin, 100 μg/ml streptomycin and 10% fetal calf serum (FBS; Thermo-Fisher, Waltham, MA, USA).

Eμ-Myc transgenic mouse lymphoma cells were cultured in high-glucose DMEM supplemented with 10% FBS (Gibco), 50 mM ß-mercaptoethanol (Sigma-Aldrich), 100 mM asparagine (Sigma-Aldrich), 100 U/mL penicillin, and 100 mg/mL streptomycin (Gibco) at 37°C and 10% CO2, as previously described.[20]

To induce recombination of the *Kat6a*$^{lox/lox}$ locus, mice were crossed to the *Rosa26Cre*^{ERT2} strain. Cre-recombinase-ERT2 protein was induced to translocate to the nucleus by treating *Kat6a*$^{lox/lox}$;*Rosa26Cre*^{ERT2} MEFs and control *Rosa26Cre*^{ERT2} MEFs wild type at the *Kat6a* locus with 1 μM 4-OH tamoxifen. Cells were treated for 96 h (not split) then re-plated in medium only. At the time points indicated throughout the text, cells were counted using trypan blue exclusion (Gibco, 15250) and an automated cell counter (Countess, Thermo Fisher) and re-plated at 1 x 10^5 live cells per well of a 6-well plate in triplicate. All compounds were prepared in DMSO (Sigma, D-2650) and added to the cell media at a 1:1000 dilution. DMSO was used as vehicle control for all experiments.

The following plasmids were used for generating retroviruses: *pBABE-puro* retroviral vector (Addgene #1764); *pBABE-puro Ras V12* retroviral vector (Addgene #1768); *pMD1-Gag-Pol* structural vector and *pCAG-Eco* (Addgene #35617) envelope vector. For packaging, HEK293T cells were plated at 3 x 10^6 in a 10 cm dish, incubated for 24 h, and then transfected using FuGENE® 6 transfection reagent (Promega) with 2 μg of either *pBABE-puro* or *pBABE-puro Ras V12*, 2 μg of *pMD1-gag-pol* and 1 μg of *pCAG-Eco*. The viral supernatant was collected after 48 h (1st collection) and 72 h (2nd collection). The viral supernatant was filtered through a 0.45 μm pore-size filter and supplemented with 4 μg/ml polybrene (Sigma). MEF cultures were maintained as above, then 24 h before infection, MEFs were plated at 1 x 10^5 cells per well of a 6-well dish. On the day of infection, culture medium was replaced with 1 ml of viral supernatant per well and incubated for 16 h. Infected MEFs were selected using 2 μg/ml puromycin. While under puromycin selection, MEFs were plated at 1 x 10^5 cells per well of a 6-well dish. At the indicated times, cell counts were performed for each well, and MEFs were re-plated at 1 x 10^5 cells per well of a 6-well dish. Within each experiment, each time point was determined in triplicate. Each experiment was performed at least three times.

Cell cycle analysis

The proportion of cells in different phases of the cell cycle was determined by flow cytometry. Cells were incubated with BrdU (10 μM) for 1 h. Incorporated BrdU was detected

using the BD BrdU Flow Kit (BD Biosciences, 552598), using an allophycocyanin conjugated anti-BrdU antibody (1:50) and DNA content was stained using 7AAD followed by flow cytometry as per the manufacturer's instructions. Fucci cells[15] were analyzed by flow cytometry. In the Fucci cells, Azami green indicates mid S, G_2, M, Kusabira orange mid-late G_1, double positive yellow cells are in early S and double negative (DN) cells in early G_1.

Detection of DNA damage.

Flow cytometry was used to detect γH2A.X, a marker of DNA double strand breaks. The BD Cytofix/Cytoperm kit (BD Biosciences, 554714) was used according to the manufacturers' instructions together with an anti γH2A.X (serine 139; clone: JBW301) biotin-conjugated antibody (1:400, Merck, Kenilworth, NJ, USA, cat: 16-193), which was detected using allophycocyanin-streptavidin essentially as described by Muslimovic and colleagues.[21]

Apoptosis/viability assays

For viability assays, cells were analyzed by flow cytometry after staining with propidium iodide and fluorescein isothiocyanate-conjugated annexin V (Thermo-Fisher A13199, 1:200) as described previously.[22]

β-Galactosidase staining

β-galactosidase activity was detected using 5-dodecanoylaminofluorescein di-β-galactopyranoside (C_{12}FDG, Life Technologies D-2893, USA) and analyzed by flow cytometry as described.[23]

Western blot analysis

Western blot analyses were performed on histone lysates as previously described[24] and analyzed for histone acetylation marks using the following antibodies: rabbit monoclonal anti-H3K9ac (Cell signaling Technology, Danvers Massachusetts: C5B11), rabbit monoclonal anti-H3K14ac (Cell signaling Technology, Danvers Massachusetts: D4B9) and rabbit polyclonal anti- H4 (Millipore, Temecula California: 05-858).

Quantitative reverse transcriptase-PCR

RT-qPCR was carried out using the LightCycler 480 (Roche, Basel, Switzerland) and SYBR green chemistry using the oligonucleotide primers listed in Supplementary Table 7.

Statistical analysis

Statistical analyses were carried out using the Stata v12 software (Stata Corporation, College Station, TX, USA). Data were analyzed as described in the figure legends, or for RNA-sequencing in the next section "RNA-sequencing".

RNA sequencing of MEFs treated with WM-8014 and WM-2474

Total RNA was purified from MEFs treated with WM-8014 and control compound WM-2474. Additionally, total RNA was purified from MEFs treated with control compound WM-2474 or vehicle (DMSO). Specifically, three independent primary cell cultures were treated for 4 days and 10 days with each compound, resulting in a total number of 24 samples. Uniquely indexed libraries were generated per sample with the TruSeq Stranded mRNA LT Sample Prep Kit (Illumina), according to manufacturer's instructions. Indexed libraries were sequenced using the Illumina NextSeq platform, generating 80 bp long paired-end reads, yielding a minimum of ~12 million total reads per sample. After assessing sequence quality, Rsubread[25] was used to map reads to the mouse genome (mm10). Mapping efficiency was >94% in all experiments. The featureCount function of Rsubread was used to assign mapped reads to Entrez RefSeq annotated genes.[26] . Differential expression analyses were undertaken using the edgeR[27] and limma[28] software packages. The first set of MEFs treated with WM-8014 or WM-2474 were analyzed independently of those MEFs treated with WM-2474 or DMSO. In each case, obsolete Entrez Ids were removed. Furthermore, in the first data set, genes that failed to achieve 0.3 counts per million (CPM) in at least three samples were filtered from the analysis leaving 15,082 genes for the downstream analysis of the WM-8014 vs. WM-2474. For the second data set, genes were filtered using the filterByExpr function in edgeR, resulting in 13,494 genes for the analysis of the WM-2474 vs. DMSO samples. The trimmed mean of log expression ratios (TMM) method[29] was used to normalize the library sizes. Multiple dimensional scaling (MDS) plots were used to determine if substantial variation existed within treatment groups. The distance between each pair of samples is computed as the leading fold change, defined as the root-mean-square of the largest 500 $\log_2$ fold changes between that pair of samples. Counts were transformed to $\log_2$ counts per million with associated precision weights using voom.[30] Differential expression was assessed using linear models and robust empirical Bayes' approach was used to obtain an estimate of the posterior gene-wise variances and to compute and test the moderated-t statistic.[31] The false discovery rate (FDR) was controlled at 0.05 using the Benjamini and Hochberg method. To increase precision for the WM-2474 vs. DMSO analysis, the linear model incorporated a correction for a replicate batch effect.

Enrichment of certain groups of genes was assessed using the curated C2 collection of the molecular signature database (MSigDB),[32] analyzed using two-sided rotation gene set tests (ROAST)[33] with 10,000 rotations and visualized using limma's barcodeplot function. The barcode plot compares differentially expressed genes in two data sets ordering genes of the first data set from left to right as most upregulated to most downregulated. Vertical red and blue bars indicate genes up or down regulated in the second dataset. Weights show $\log_2$-fold changes and worms show relative enrichment. Differentially expressed genes with larger fold changes were prioritized using the TREAT method[34] with a 2.5-fold-change threshold.

RNA sequencing of MEFs from *Kat6a*$^{-/-}$ and wild type embryos

MEFs were isolated from 3 *Kat6a*$^{-/-}$ and 2 age- and sex-matched wild type E12.5 embryos. Sequencing and bioinformatics analysis was similar to that for compound treated MEFs. Between 43 and 68 million 80 bp paired-end reads were generated per sample. Obsolete Entrez Ids and genes that failed to achieve 0.224 counts per million (CPM) in at least two samples were removed from the analysis, leaving 15,450 genes for downstream analysis. Differential expression analysis found 3220 up-regulated and 3513 down-regulated genes in the *Kat6a*$^{-/-}$ MEFs at FDR < 0.05. To prioritize DE genes with larger fold-changes, a TREAT analysis was conducted relative to a fold change threshold of 1.5. This resulted in 263 up-regulated and 709 down-regulated genes at TREAT FDR < 0.05, and these genes were used for expression signature analyses.

RNA sequencing of lymphoma cell line EMRK1184 with vehicle and WM-1119

Total RNA was purified from lymphoma cells treated, in triplicate, with WM-1119 or vehicle for 3 days and 6 days. Uniquely indexed libraries were generated per sample with the TruSeq Stranded mRNA LT Sample Prep Kit (Illumina), and indexed libraries were sequenced using the Illumina NextSeq platform and then analyzed using the bioinformatics tools as described above. Between 17 and 51 million mapped, 80 bp paired-end reads were generated per sample. Obsolete Entrez Ids were removed and genes expressed at low levels were filtered using edgeR's filterByExpr function, leaving 11,979 genes for downstream analysis. Differential expression analyses, performed as described above, found 1352 and 1862 upregulated, as well as 1556 and 1841 down-regulated genes, at day 3 and day 6 of treatment WM-1119 vs. DMSO, respectively, at FDR < 0.05 in the EMRK1184 cells.

Affymetrix microarrays

To obtain an expression signature for cell senescence vs proliferation, Affymetrix microarray data was download from GEO series GSE39469.[35] RMA normalization was applied and differential expression was assessed using the limma package.

Chromatin immunoprecipitation

Wild type mouse embryonic fibroblasts, treated with MYST inhibitor WM-8014, negative control compound WM-2474 or a DMSO vehicle control were processed for ChIP as described previously[36] with the following modifications. Cross-linked material was sonicated using the Covaris S2 (Covaris). 20 to 40 μg sonicated chromatin samples were made up to 200 μl in 1% SDS lysis buffer (1% SDS, 10 nM EDTA, 50 mM Tris-Cl (pH 8.1), 1x cOmplete™ protease inhibitor tablet (Sigma), EDTA-free (Sigma), 1x 10 mM sodium butyrate, in MQ-H_2O) and added to 1.8 ml ChIP dilution buffer (0.01% SDS, 1.1% Triton X-100, 1.2 mM EDTA, 16.7 mM Tris-Cl, pH 8.1, 167 mM NaCl, 1x cOmplete™ protease inhibitor tablet, EDTA-free (Sigma, 1x 10 mM sodium butyrate, MQ-H_2O). 100 μl were reserved as input and frozen at -20°C. Chromatin was incubated with anti-acetyl histone H3 (Lys9) antibody (Cell signaling, 9649) overnight at 4°C, followed by protein A+G magnetic beads (Millipore, 16-663) for 1 h at 4°C. Immunoprecipitated and input samples were processed from this point as reported previously[36] with DNA purification by phenol:chloroform extraction and ethanol precipitation. Genes of interest were analyzed by qPCR using primer sets described in Supplementary Table 8.

Zebrafish methods

Zebrafish experimentation was approved by the Animal Ethics Committee of the Walter and Eliza Hall Institute of Medical Research. Zebrafish were maintained at 28°C on a 12 h light/dark cycle using standard zebrafish husbandry procedures. For multiphoton microscopy of transgenic zebrafish *Tg(fabp10:RFP; elaA:EGFP)* and TO-*kras*^{G12V}, intact larvae were anaesthetized with benzocaine (50 mg/L) and mounted in 1% agarose. Images were acquired using an Olympus FVMPE-RS (C4.48) multiphoton microscope with a 25x objective and Olympus FV30-SW software. Excitation wavelengths for GFP and RFP were 840 nm and 1100 nm, respectively. Emission was detected at 550 nm and 580 nm, respectively. For volumetric analysis of whole livers, Z-stacks, step size 2 μm, were imported into ImageJ (1.49v) or Imaris. qPCR oligonucleotides for zebrafish *Cdkn2a* and *Cdkn1a* are listed in Supplementary Table 7.

To assess cell proliferation using incorporation of EdU, 7 dpf zebrafish larvae were incubated in 2 mM EdU for 1 h. After fixation, livers were dissected and EdU labelling carried out utilising the Click-iT EdU Alexa Fluor 647 imaging kit (Invitrogen) according to manufacturer's instructions. Image acquisition was performed using an Olympus FVMPE-RS multiphoton microscope with excitation wavelengths of 950 nm for Hoechst and 1160 nm for AF647. The number of Hoechst and EdU positive cells was quantified using Arivis Vision4D software.

Treatment and analysis of lymphoma progression

A lymphoma cell line EMRK1184 derived from an *Eμ-Myc* transgenic mouse,[37] was infected with a luciferase-expressing lentiviral imaging construct to allow quantitation of tumor burden throughout treatment. The lentivirus construct was generated by inserting Luciferase2 linked via a T2A peptide to eGFP downstream from the human ubiquitin promoter of the FUGW vector.[38] Lentiviruses were packaged using standard methods and supernatants containing infectious viral particles were harvested 2–3 d post-transfection. EMRK1184 was infected with imaging virus by spin infection. 1 x 10^5 lymphoma cells were resuspended in 5 ml of virus-containing supernatant containing 10 ng/ml polybrene, incubated for 30 min at 37°C/10% CO2, and then centrifuged at 2200 rpm for 2.5 h at 32°C. The supernatant was subsequently discarded, and the cells were resuspended in fresh medium for culture.

Cells were sorted for eGFP expression using an AriaW cell sorter (Becton Dickinson). 1 x 10^5 live, eGFP expressing Eμ-*Myc* lymphoma cells were resuspended in PBS and injected into the tail vein of male C57BL/6-albino recipient mice (6 to 8 weeks of age). Mice were randomly assigned to treatment groups. WM-1119 was prepared as a slurry at 12.5 mg/ml in 50%polyethylene glycol in water and sonicated. Mice used in this experiment weighed 23 to 27 g. Treatment started at 12 pm on day 3 after lymphoma cell transplant after baseline bioluminescence imaging. For the 4x/day cohort mice were given intraperitoneal injections with either vehicle or WM-1119 slurry at 6 am, noon, 6 pm and midnight; the 3x/day cohort was injected at 8 am, 4 pm and midnight. All experiments were performed under approval from the Walter and Eliza Hall Institute's Animal Ethics Committee.

The tumor burden was assessed by automated methods, namely by bioluminescence using the IVIS live-imaging system (Perkin Elmer) as described previously.[20] Briefly, 200 μl of 15 mg/ml D-luciferin potassium salt (Caliper Life Sciences) in PBS was administered to the mice by intraperitoneal (i.p.) injection. Mice were then anaesthetized with isoflurane inhalant

and imaged using the IVIS Spectrum In Vivo Imaging System (Perkin Elmer) to detect luciferase bioluminescence exactly 15 min after administration of the luciferin substrate. The tumor burden was quantified by measuring the total photon flux per second emitted from a region of interest (ROI) drawn around the whole mouse.

Flow cytometry methods:

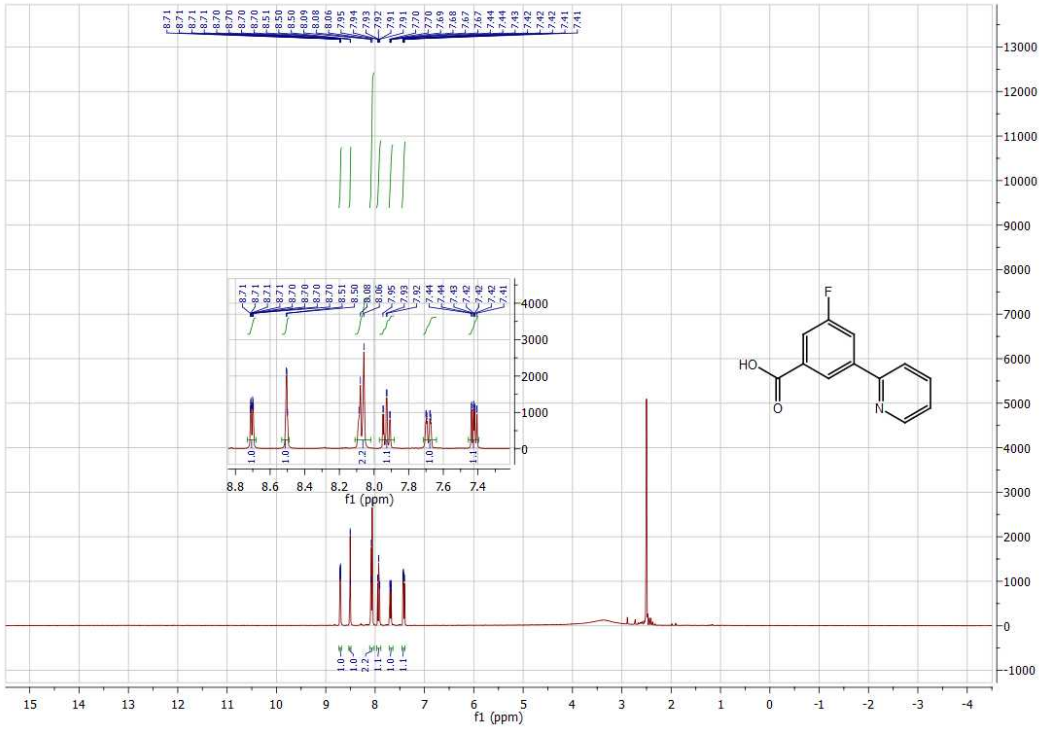
Tissues were processed and erythrocyte-lysed as described.[39] WBC counts were obtained using a Countess® Automated Cell Counter (ThermoFisher) for spleen and bone marrow and the Advia 3120 analyzer (Bayer) for blood. Cells were stained with CD19-PeCy7 (clone 1D3, BD Biosciences # 561739), B220-A700 (clone 16A, WEHI), IgM-A647 (clone II/41, WEHI), and IgD-PE (clone 11-26c, WEHI) for 30 min, washed, resuspended in Fluoro-Gold viability marker (Sigma: 39286), and analyzed on a LSRII (BD Biosciences). Gating strategy is shown in the Supplementary Data section.

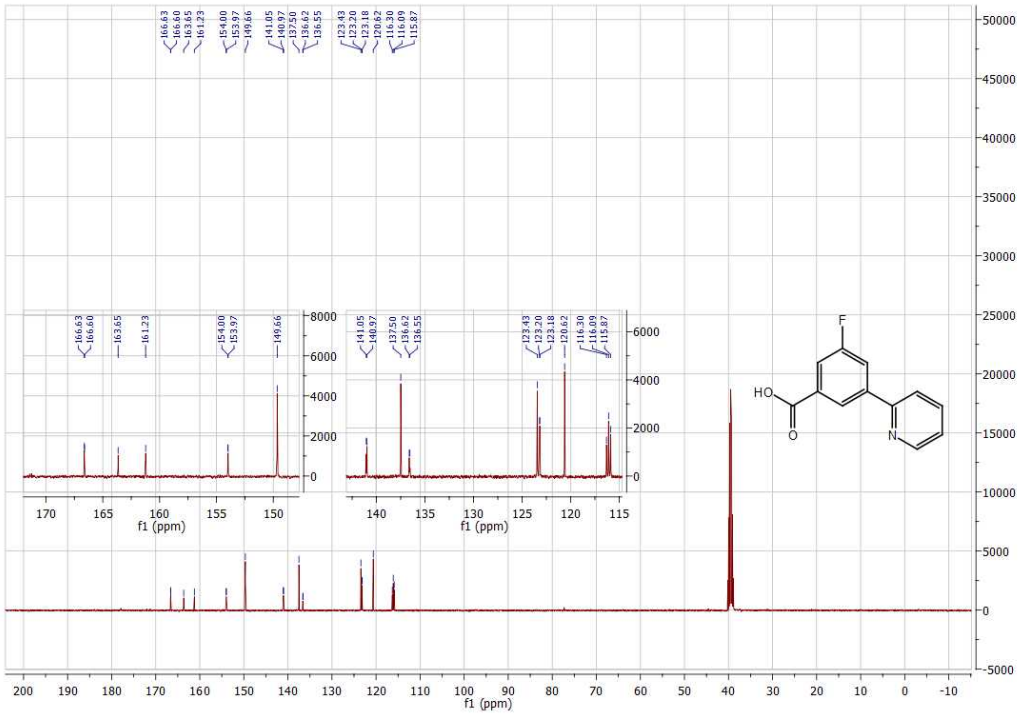
Determination of histone acetylation levels was performed by flow cytometry after intracellular staining. Cells were incubated with the LIVE/DEAD™ Fixable Green Dead Cell Stain (Invitrogen: L34969) as per the manufacturer's instructions prior to staining with cell surface markers as above. Cells were fixed and permeabalized using the Foxp3 Transcription Factor Staining Buffer Kit (Invitrogen, A24866A) as we have described previously.[40] Cells were stained with antibodies for acetyl-H3K9 (clone C5B11, Rabbit mAb, #9649, Cell Signaling), acetyl-H3K14 antibody (clone D4B9, Rabbit mAb,#7627, Cell Signaling) or an isotype control IgG antibody (clone DA1E, Rabbit mAb IgG XP® Isotype Control,#3900, Cell Signaling) overnight in Foxp3 Transcription Factor Wash Buffer (Invitrogen, A24261) with 2% normal goat serum, then for 1 h with a goat anti-rabbit secondary antibody (DyLight™ 405 AffiniPure Jackson ImmunoResearch Laboratories Inc., 111-475-003). Fixed cells were analysed on a Fortessa (BD Biosciences). Data were analysed using FlowJo (Treestar Inc.).

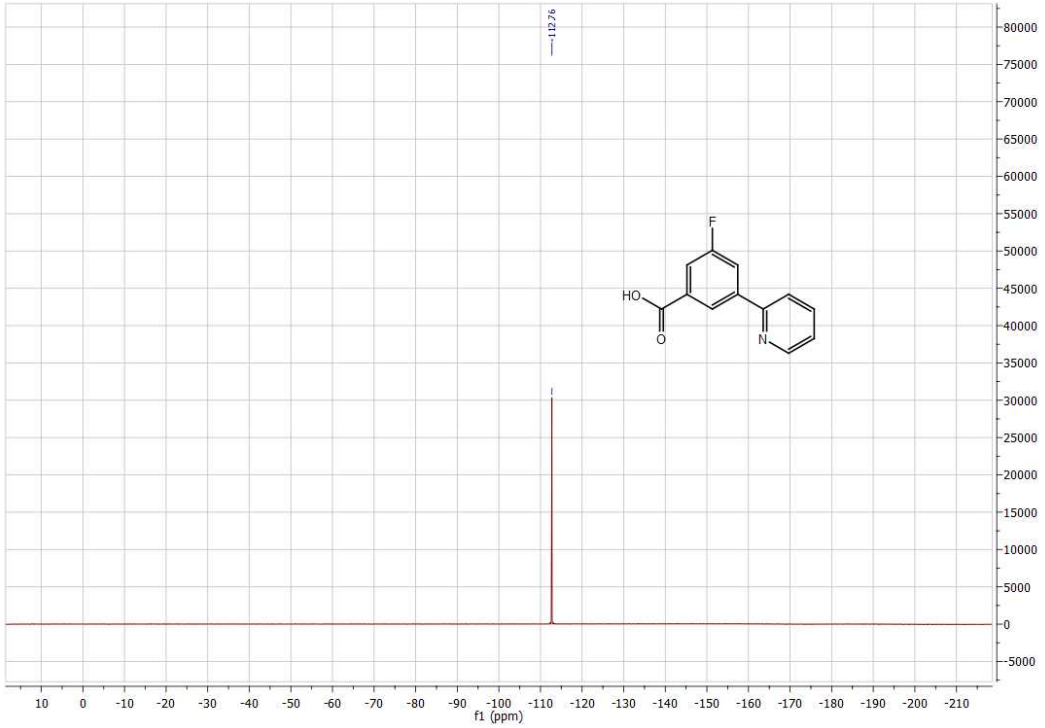
Supplementary data: Spectra

^{1}H NMR, ^{13}C NMR, ^{19}F NMR (where appropriate), HRMS and HPLC spectra for experimental compounds: WM-8014, WM-2474, WM-1119 and their intermediates

WM-1119- Intermediates







Qualitative Compound Report

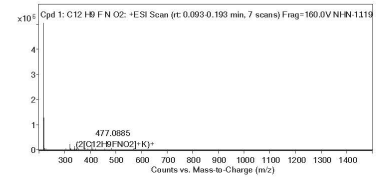
Data File	NHN-1119 Pre.d	Sample Name	NHN-1119 Pre
Sample Type	Sample	Position	P1-B8
Instrument Name	Instrument 1	User Name	Dr Jason Dang
Acq Method	Monash_Direct.m	Acquired Time	30-Nov-17 9:02:36 PM
IRM Calibration Status	Success	DA Method	Monash_Accuracy.m
Comment			
Sample Group		Info.	
Formula	C12H9FNO2	Stream Name	LC 1
Acquisition SW Version	6200 series TOF/6500 series Q-TOF B.06.01 (B6172.SP1)		

Compound Table

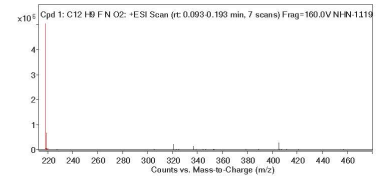
Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)
Cpd 1: C12 H9 F N O2	0.109	218.0621	5066.304	C12 H9 F N O2	218.0617	1.59

Compound Label	m/z	RT	Algorithm	Mass
Cpd 1: C12 H9 F N O2	218.0615	0.109	Find By Formula	218.0621

MS Spectrum



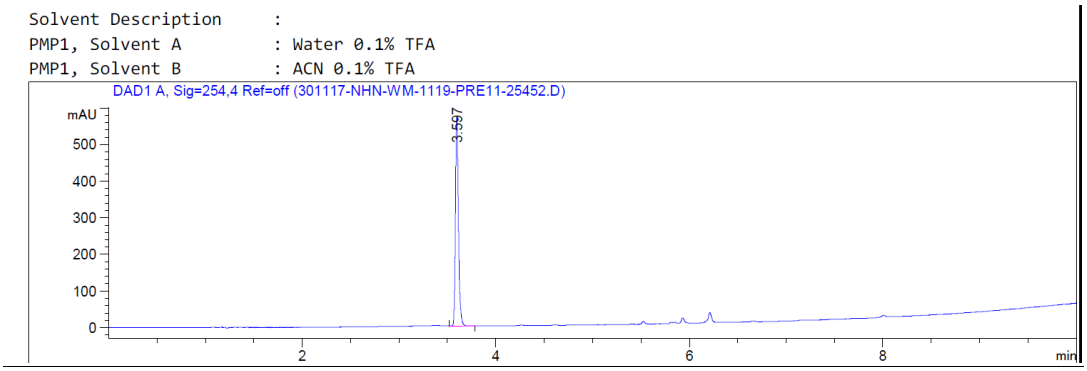
MS Zoomed Spectrum



MS Spectrum Peak List

m/z	Calc m/z	Diff(ppm)	z	Abund	Formula	Ion
218.0615	218.0612	-1.39	1	5066.303	C12H9FNO2	M+
241.0502	241.051	3.22	1	130	C12H9FNO2	(M+Na)+
475.0888	475.0866	-4.61	1	3120.51	C12H9FNO2	(2M+K)+

— End Of Report —


Solvent Description :
PMP1, Solvent A : Water 0.1% TFA
PMP1, Solvent B : ACN 0.1% TFA
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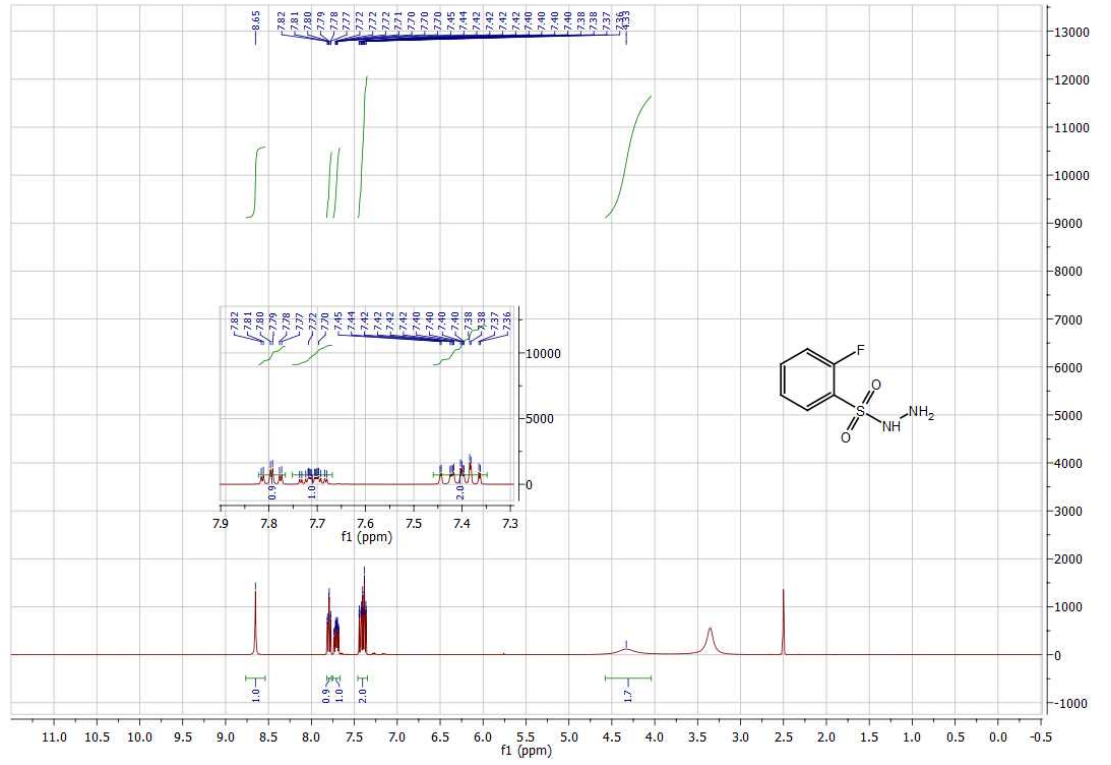
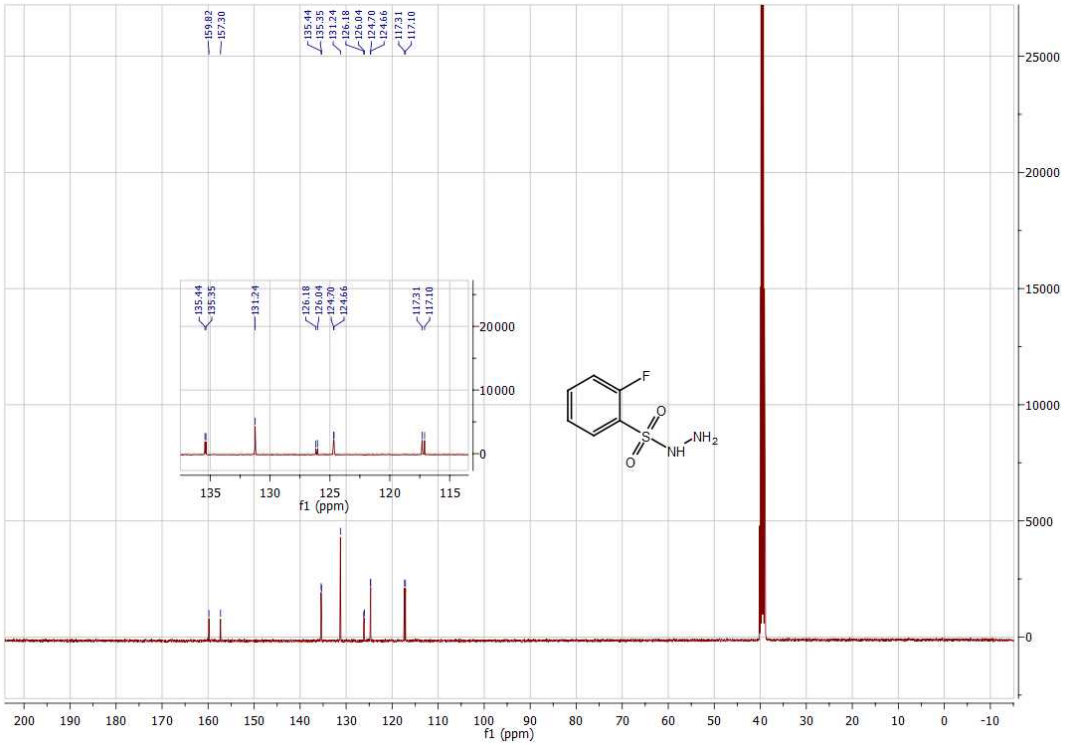
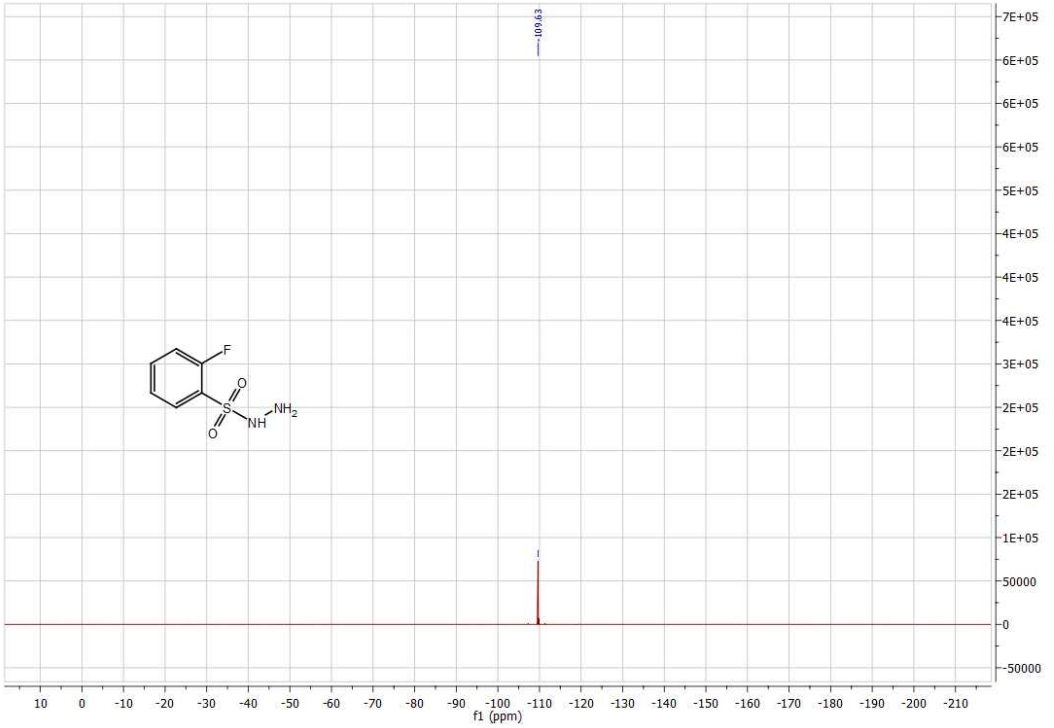
3.597
min


f1 (ppm)
F
O
S
NH
NH2


159.82
157.30
135.44
135.35
131.24
126.18
126.04
124.70
124.66
117.31
117.10
F
O
S
O
NH
NH2
f1 (ppm)
f1 (ppm)


-109.63
F
O
S
O
NH
NH2
f1 (ppm)

Qualitative Compound Report

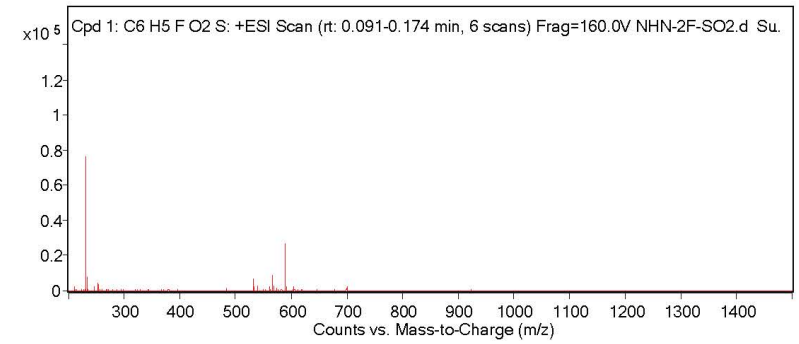
Data File	NHN-2F-SO2.d	Sample Name	NHN-2F-SO2
Sample Type	Unknown	Position	P1-A1
Instrument Name	Instrument 1	User Name	Dr Jason Dang
Acq Method	Monash_Direct.m	Acquired Time	14-Feb-18 10:48:46 PM
IRM Calibration Status	Success	DA Method	Monash_Accuracy.m
Comment			
Sample Group		Info.	
Formula	C6H7FN2O2S	Stream Name	LC 1
Acquisition SW Version	6200 series TOF/6500 series Q-TOF B.06.01 (B6172 SP1)		

Compound Table

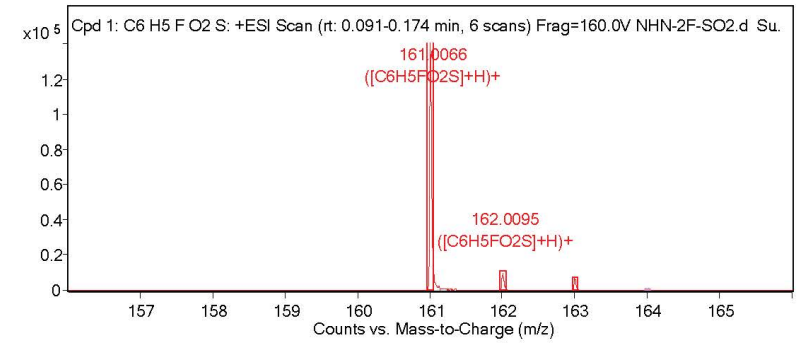
Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)
Cpd 1: C6 H5 F O2 S	0.107	159.9993	146873	C6 H5 F O2 S	159.9994	-0.81

Compound Label	*m/z*	RT	Algorithm	Mass
Cpd 1: C6 H5 F O2 S	161.0066	0.107	Find By Formula	159.9993

MS Spectrum



MS Zoomed Spectrum

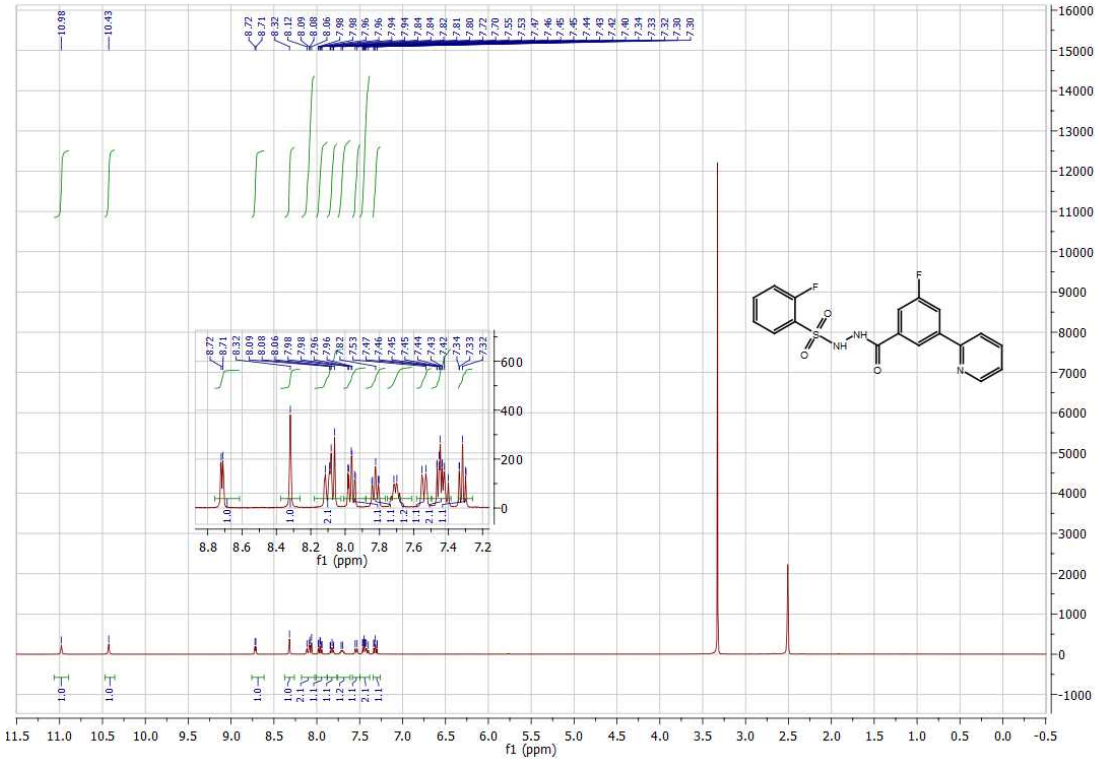


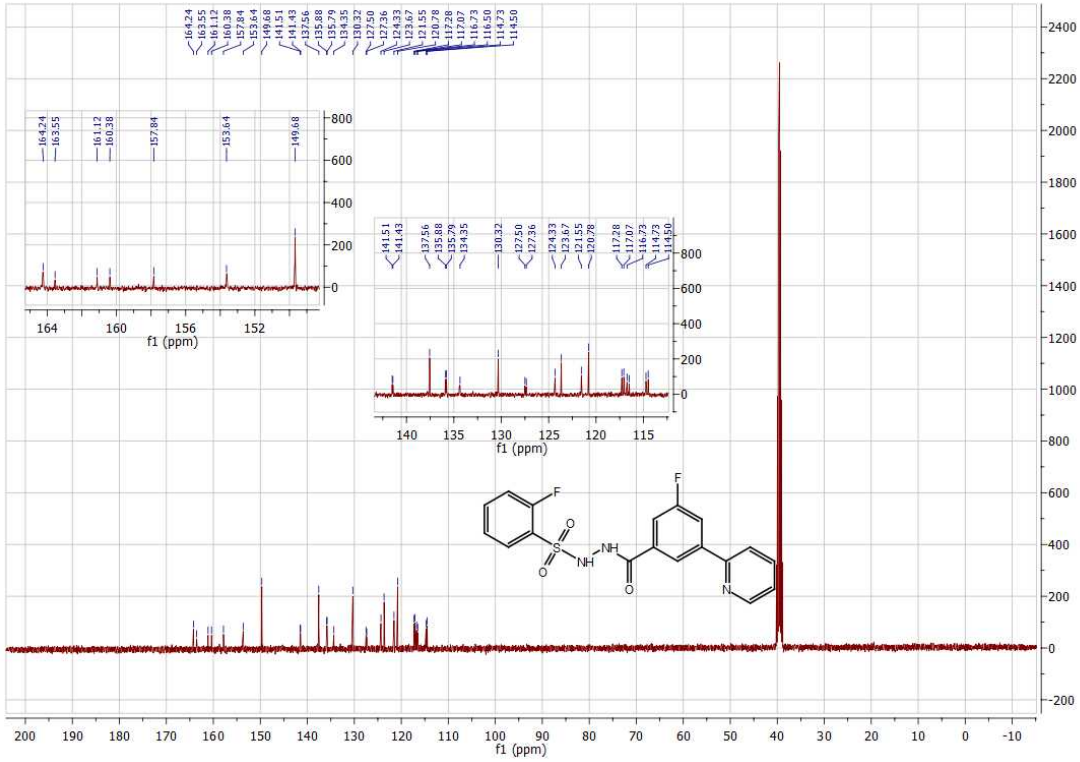
MS Spectrum Peak List

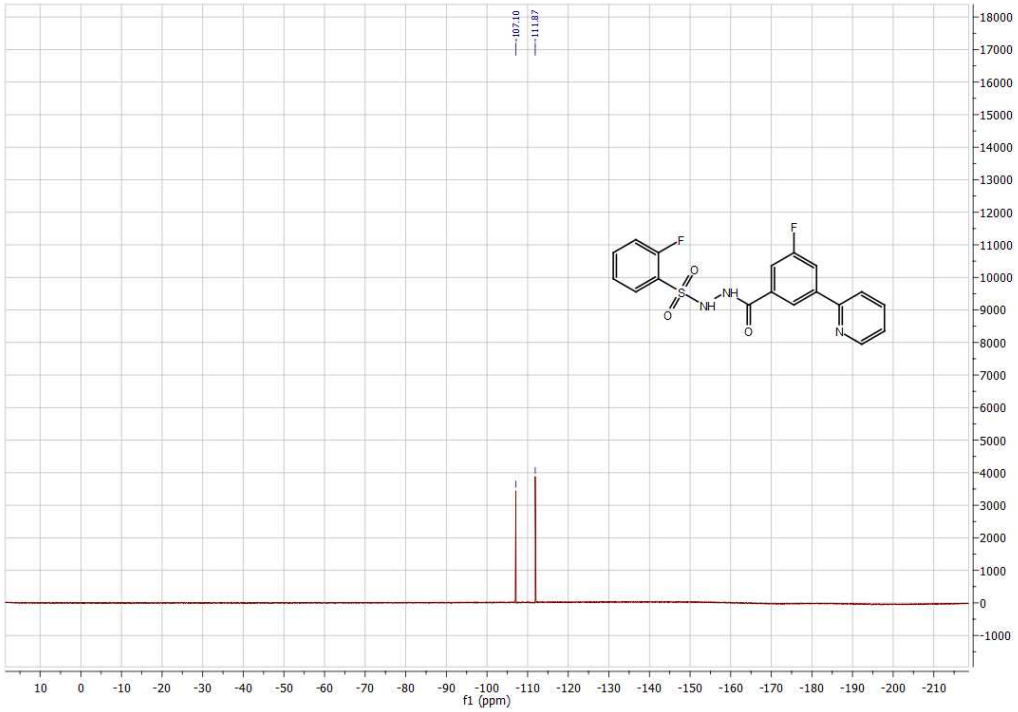
m/z	Calc *m/z*	Diff(ppm)	z	Abund	Formula	Ion
161.0066	161.0067	0.66	1	146872.55	C6H5FO2S	(M+H)+
162.0095	162.0097	1.04	1	9746.65	C6H5FO2S	(M+H)+
163.003	163.0036	3.68	1	6572.59	C6H5FO2S	(M+H)+

--- End Of Report ---

WM-1119







Qualitative Compound Report

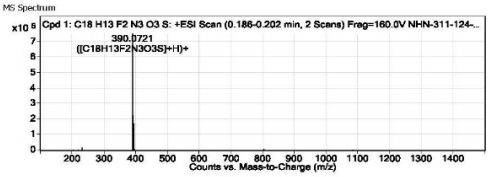
Data File	NHN-311-124-01.d	**Sample Name**	NHN-311-124-01
Sample Type	Sample	**Position**	P1-B1
Instrument Name	Instrument 1	**User Name**	Dr Jason Dang
Acq Method	Monash_Direct.m	**Acquired Time**	28-Jul-15 10:57:14 AM
IRM Calibration Status	Success	**DA Method**	Monash_Accuracy.m
Comment			
Sample Group		**Info.**	
Formula	C18H13F2N3O3S	**Acquisition SW Version**	6200 series TOF/6500 series Q-TOF B.05.01 (B5125.1)

Compound Table

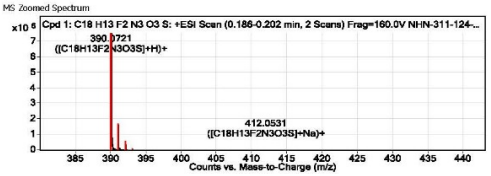
Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)	MFG Formula	DB Formula
Cpd 1: C18 H13 F2 N3 O3 S	0.119	389.0649	7610386	C18 H13 F2 N3 O3 S	389.0646	0.87	C18 H13 F2 N3 O3 S	C18 H13 F2 N3 O3 S

Compound Label	m/z	RT	Algorithm	Mass
Cpd 1: C18 H13 F2 N3 O3 S	390.0721	0.119	Find By Formula	389.0649

MS Spectrum



MS Zoomed Spectrum



MS Spectrum Peak List

m/z	Calc m/z	Diff(ppm)	z	Abund	Formula	Ion
390.0721	390.0718	-0.66	1	7610386.25	C18H13F2N3O3S	(M+H)+
412.0531	412.0538	1.72	1	16776.26	C18H13F2N3O3S	(M+Na)+
428.0273	428.0277	0.97	1	3119.51	C18H13F2N3O3S	(M+K)+

--- End Of Report ---

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Solvent Description      :
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PMP1, Solvent B          : ACN 0.1% TFA
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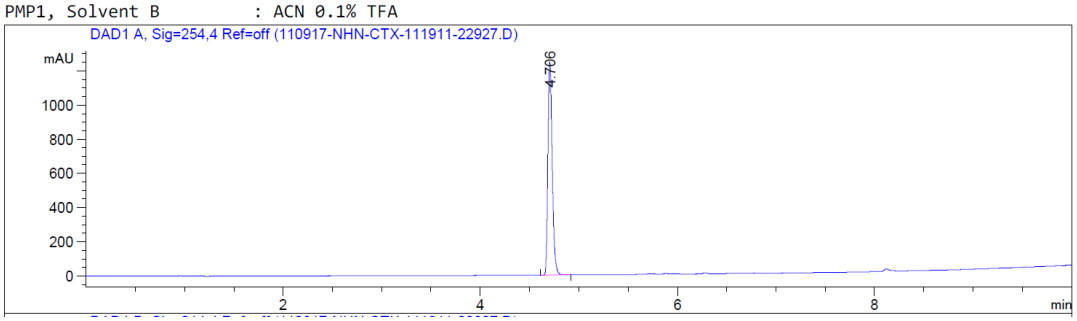
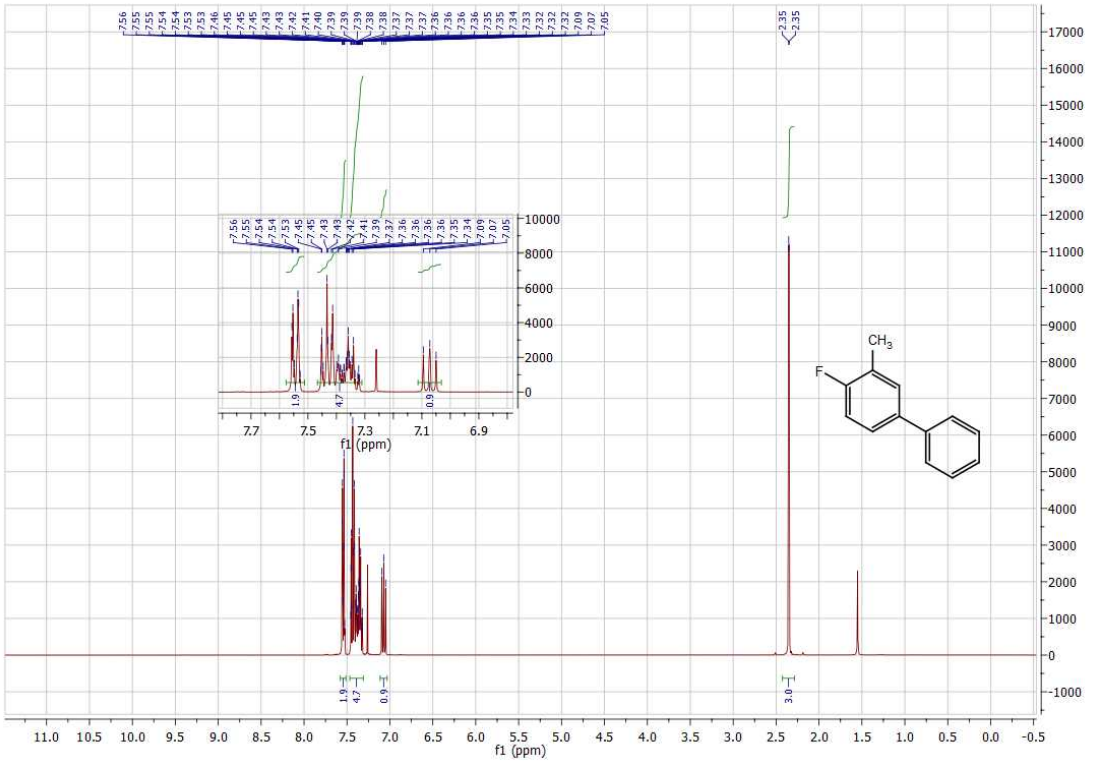
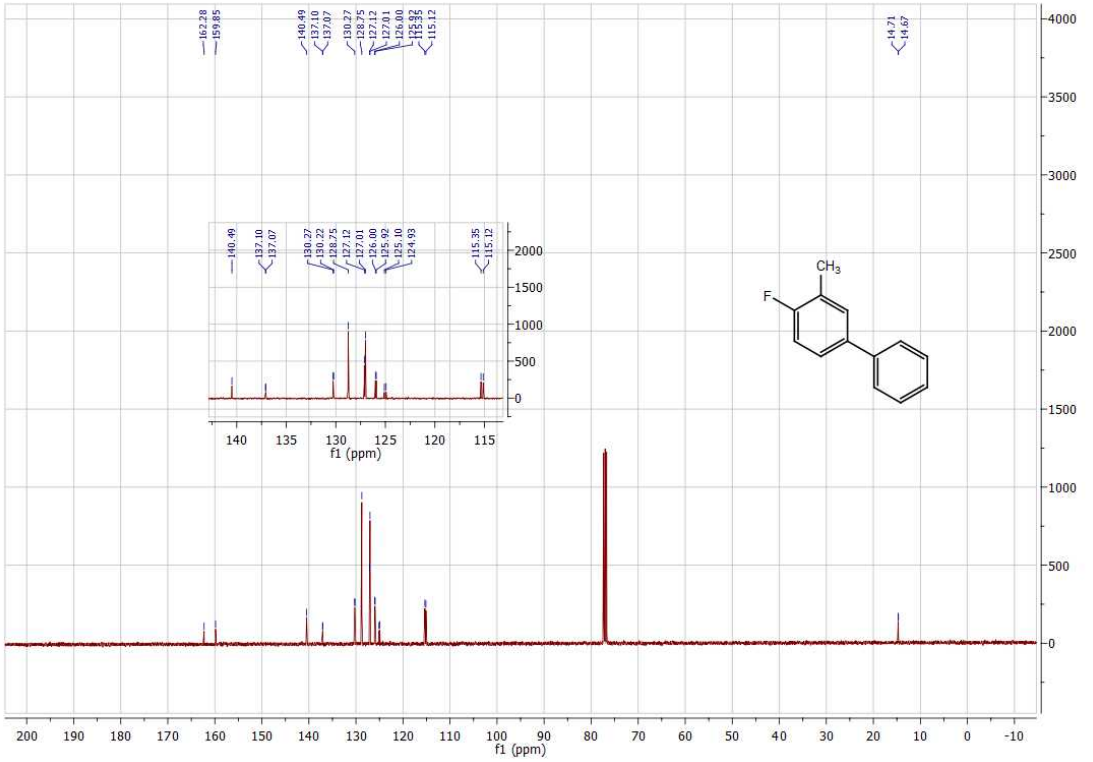


WM-8014- Intermediates




162.28
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137.07
130.27
128.75
127.12
127.01
126.00
125.92
125.10
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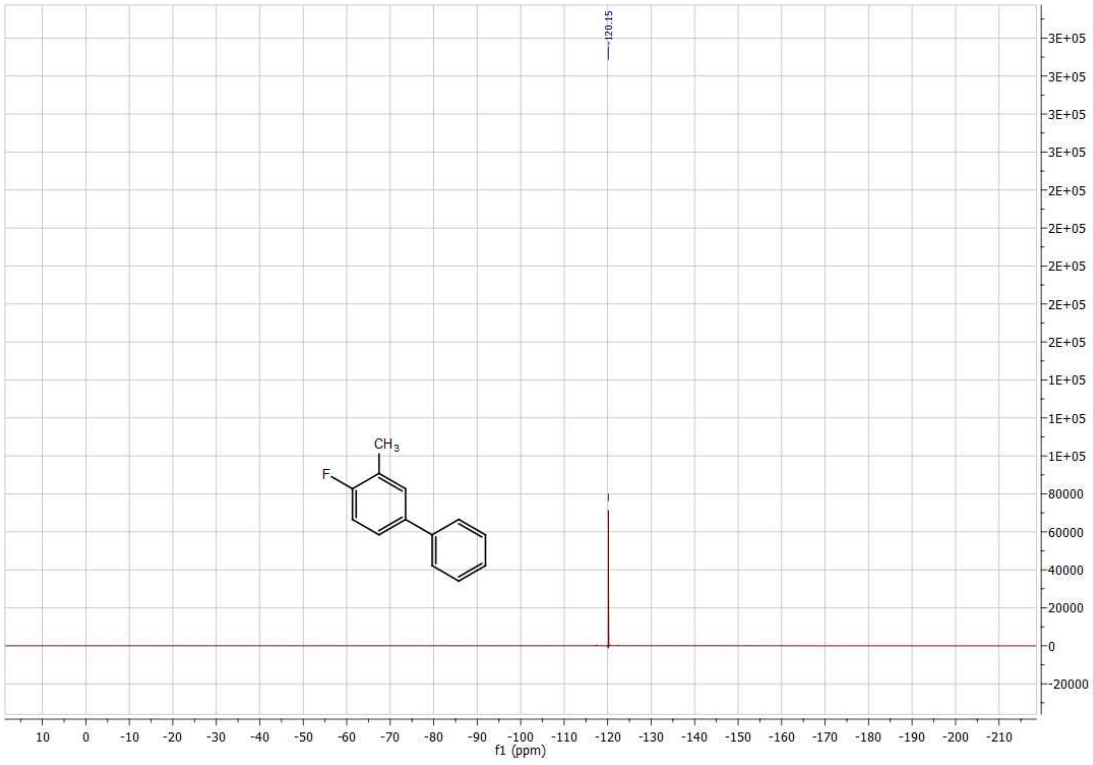
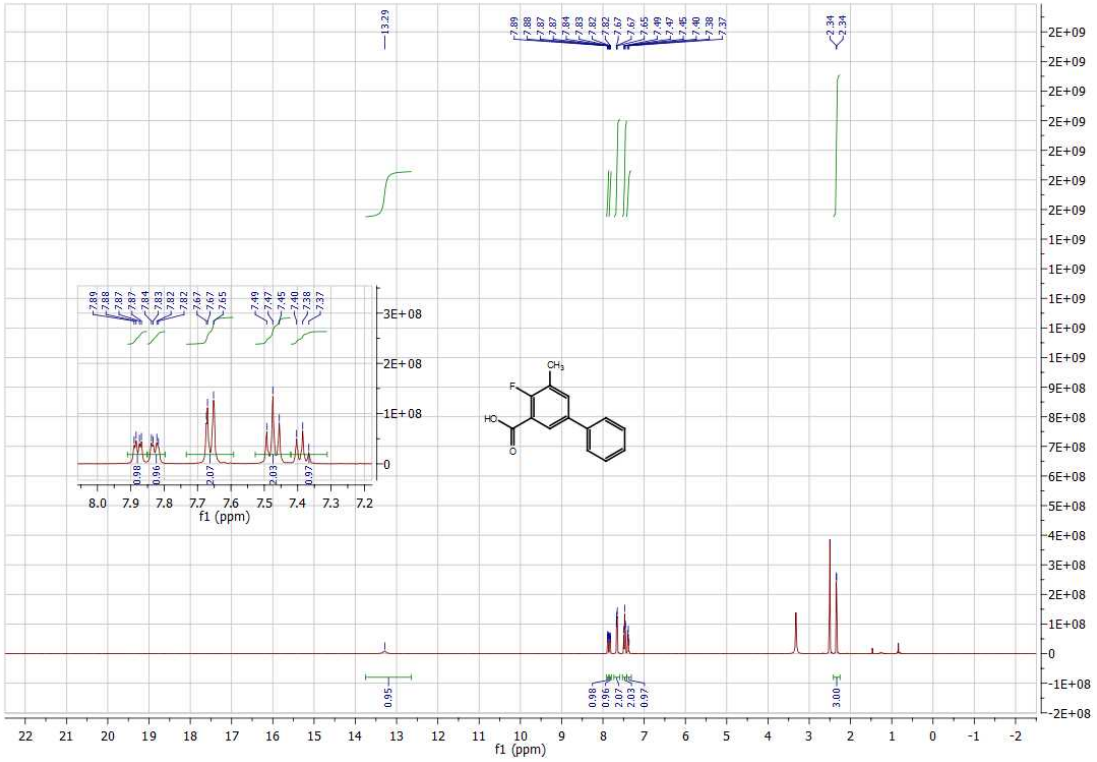
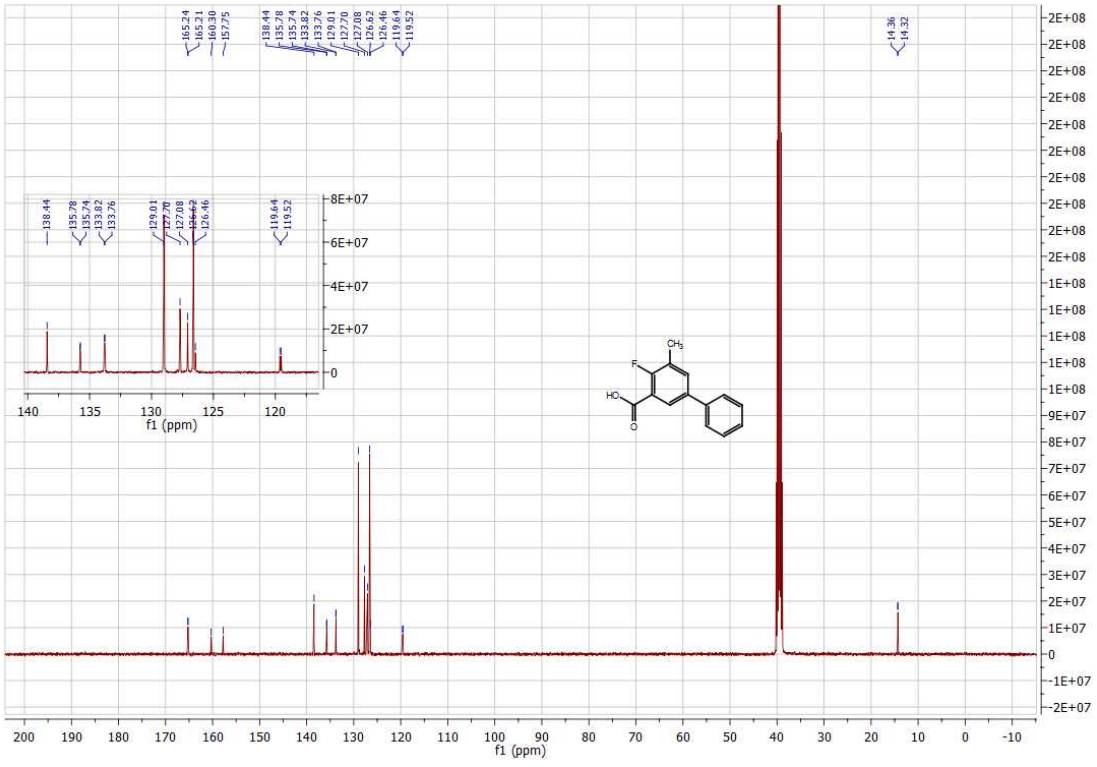
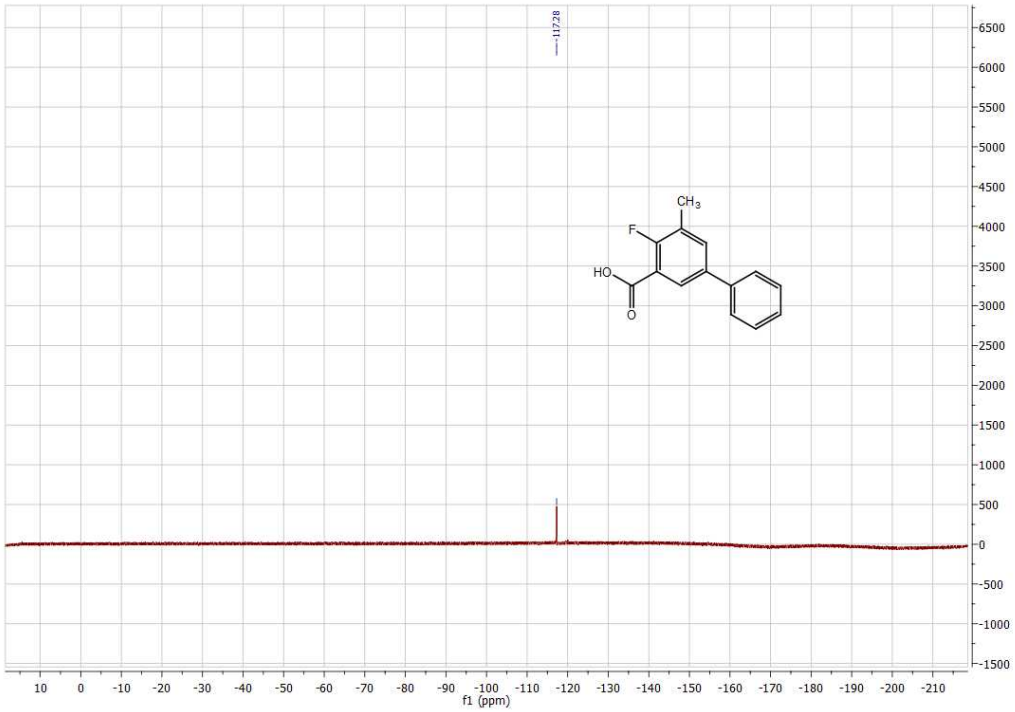
115.12
14.71
14.67
CH3
F
f1 (ppm)


120.15
CH3
F
f1 (ppm)


f1 (ppm)


f1 (ppm)



Qualitative Compound Report

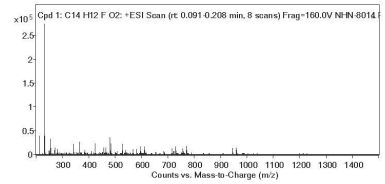
Data File	NHN-8014 Pre.d	Sample Name	NHN-8014 Pre
Sample Type	Sample	Position	P1-B6
Instrument Name	Instrument 1	User Name	Dr Jason Dang
Acq Method	Monash_Direct.m	Acquired Time	30-Nov-17 8:56:02 PM
IRM Calibration Status	Success	DA Method	Monash_Accuracy.m
Comment			
Sample Group		Info.	
Formula	C14H12FO2	Stream Name	LC 1
Acquisition SW Version	6200 series TOF/6500 series Q-TOF B.06.01 (B6172.SP1)		

Compound Table

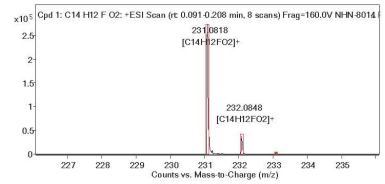
Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)
Cpd 1: C14 H12 F O2	0.108	231.0823	275275	C14 H12 F O2	231.0821	0.77

Compound Label	m/z	RT	Algorithm	Mass
Cpd 1: C14 H12 F O2	231.0818	0.108	Find By Formula	231.0823

MS Spectrum



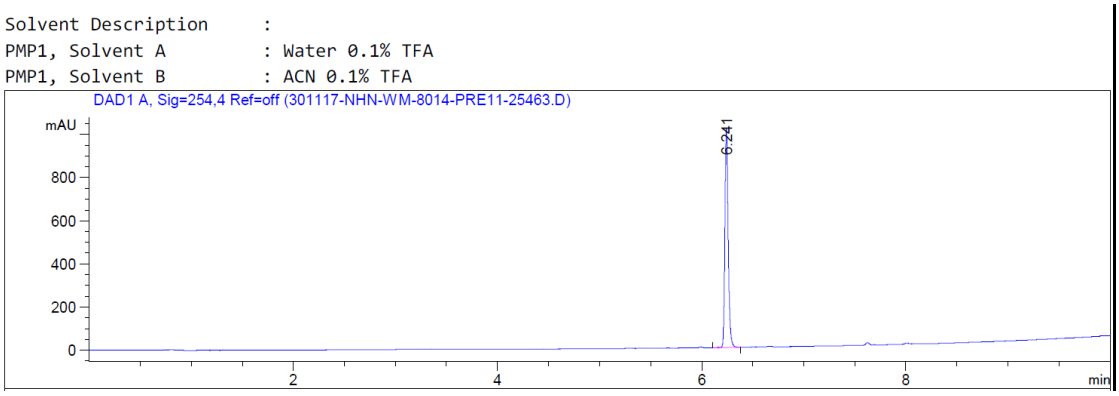
MS Zoomed Spectrum



MS Spectrum Peak List

m/z	Calc. m/z	Diff(ppm)	z	Abund	Formula	Ion
231.0818	231.0836	-0.97	1	275274.56	C14H12FO2	M+
232.0848	232.085	-0.57	1	38958.17	C14H12FO2	M+
233.0876	233.0877	0.29	1	3729.52	C14H12FO2	M+

--- End Of Report ---


Solvent Description :
PMP1, Solvent A : Water 0.1% TFA
PMP1, Solvent B : ACN 0.1% TFA
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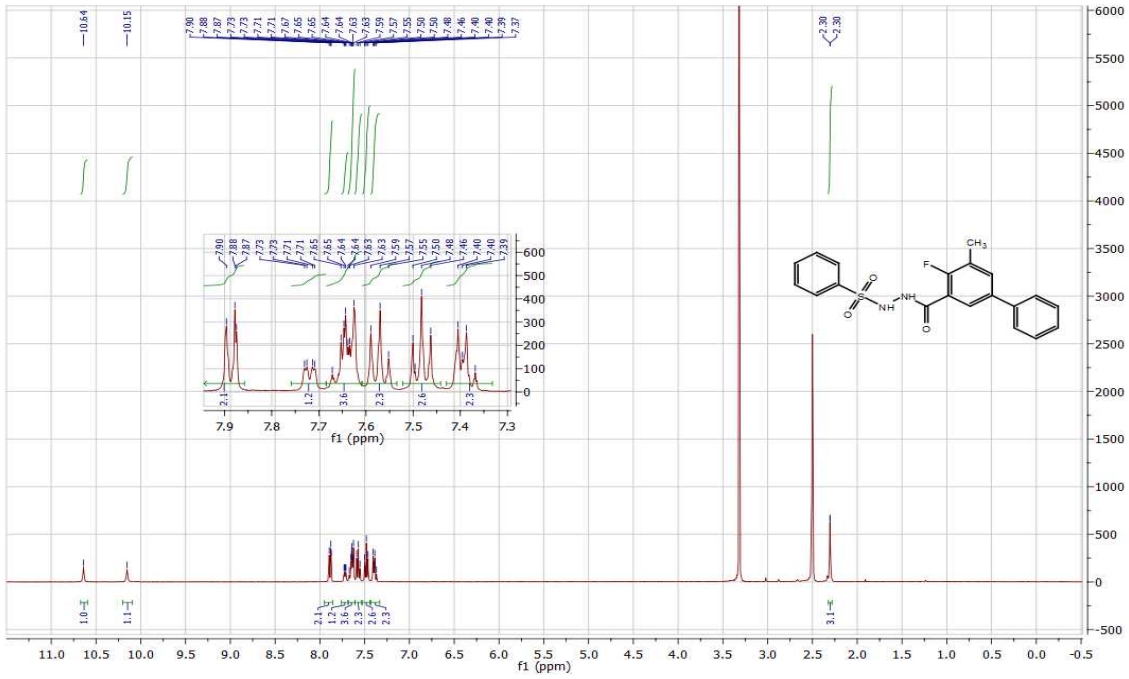
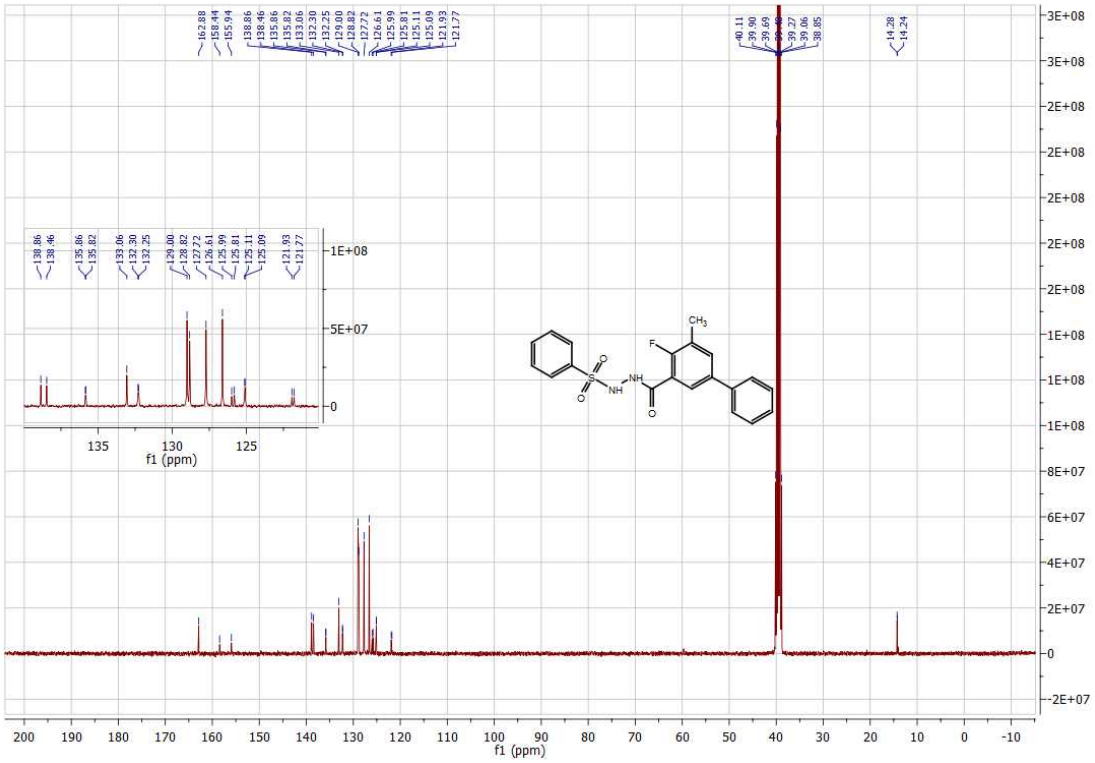
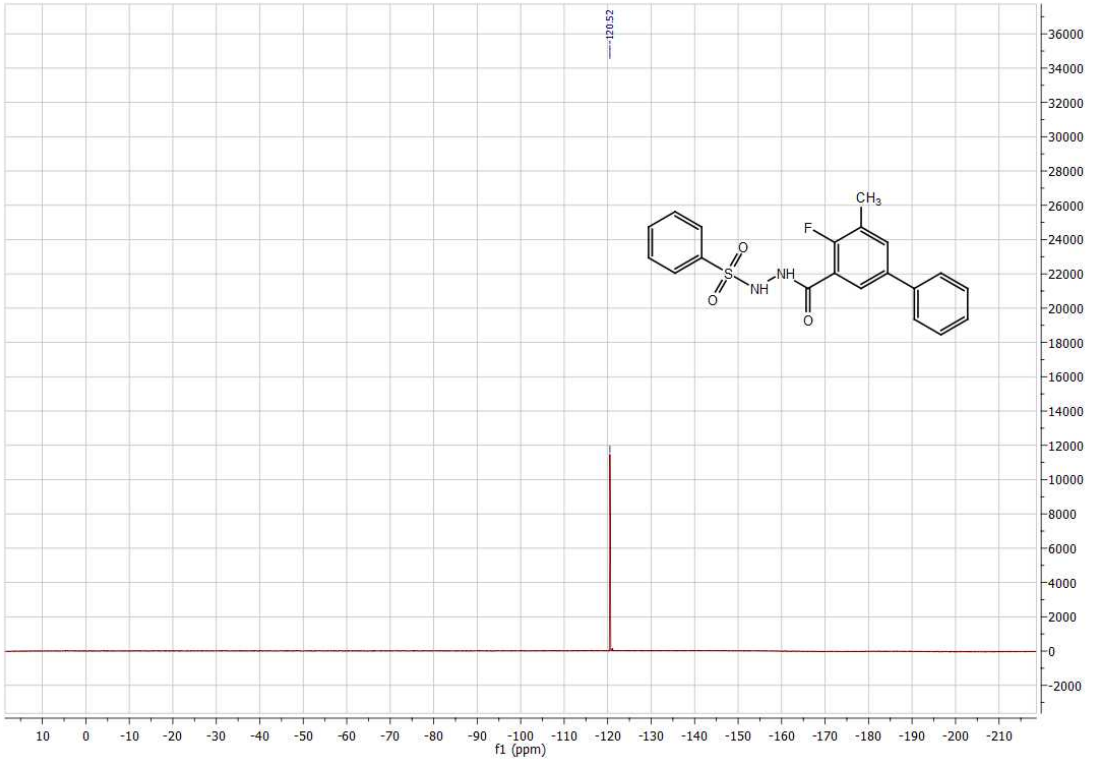
mAU
6.241
min

WM-8014


f1 (ppm)


f1 (ppm)


f1 (ppm)

Qualitative Compound Report

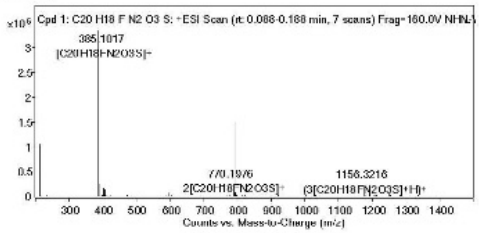
Data File	NHN-NHN-301-1	Sample Name	NHN-NHN-301-1
Sample Type	Sample	Position	P1-E9
Instrument Name	Instrument 1	User Name	Dr Jason Dang
Acq Method	Monash_Direct.m	Acquired Time	30 Nov 17 9:06:57 PM
IRM Calibration Status	Success	DA Method	Monash_Accuracy.m
Comment			
Sample Group		Info.	
Formula	C20H18FN2O3S	Stream Name	LC 1
Acquisition SW Version	6200 series TOF/6500 series Q-TOF B.05.01 (B5172.5P1)		

Compound Table

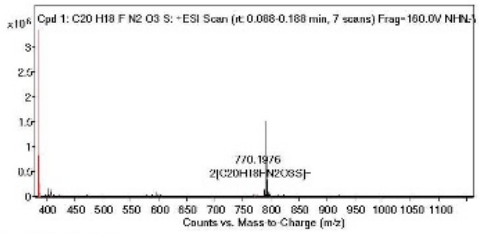
Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff [ppm]
Cpd 1: C20 H18 F N2 O3 S	0.107	385.1023	34364.93	C20 H18 F N2 O3 S	385.1023	0.10

Compound Label	m/z	RT	Algorithm	Mass
Cpd 1: C20 H18 F N2 O3 S	385.1017	0.104	Find By Formula	385.1023

MS Spectrum



MS Zoomed Spectrum



MS Spectrum Peak List

m/z	Calc m/z	Diff [ppm]	z	Abund	Formula	Ion
385.1017	385.1023	-15.15	1	34364.93	C20H18FN2O3S	M+
408.0861	408.0914	-12.98	1	31662.07	C20H18FN2O3S	[M+Na]+
770.1976	770.2036	-8.22	1	14901.98	C20H18FN2O3S	2M+

End Of Report

Printed at 9:08 PM on 30-Nov-2017

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Solvent Description     :
PMP1, Solvent A         : Water 0.1% TFA
PMP1, Solvent B         : ACN 0.1% TFA
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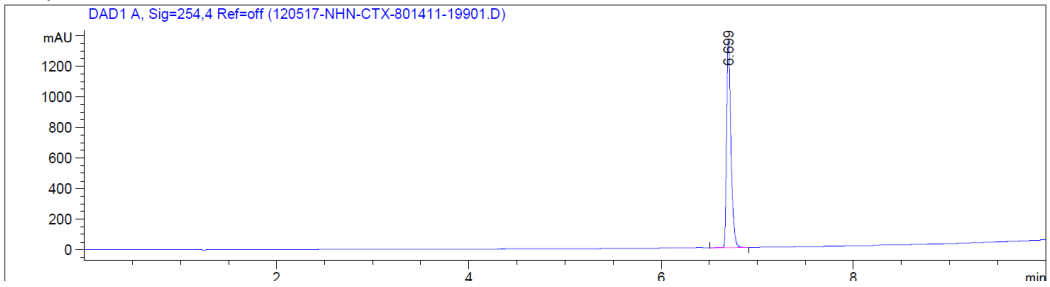
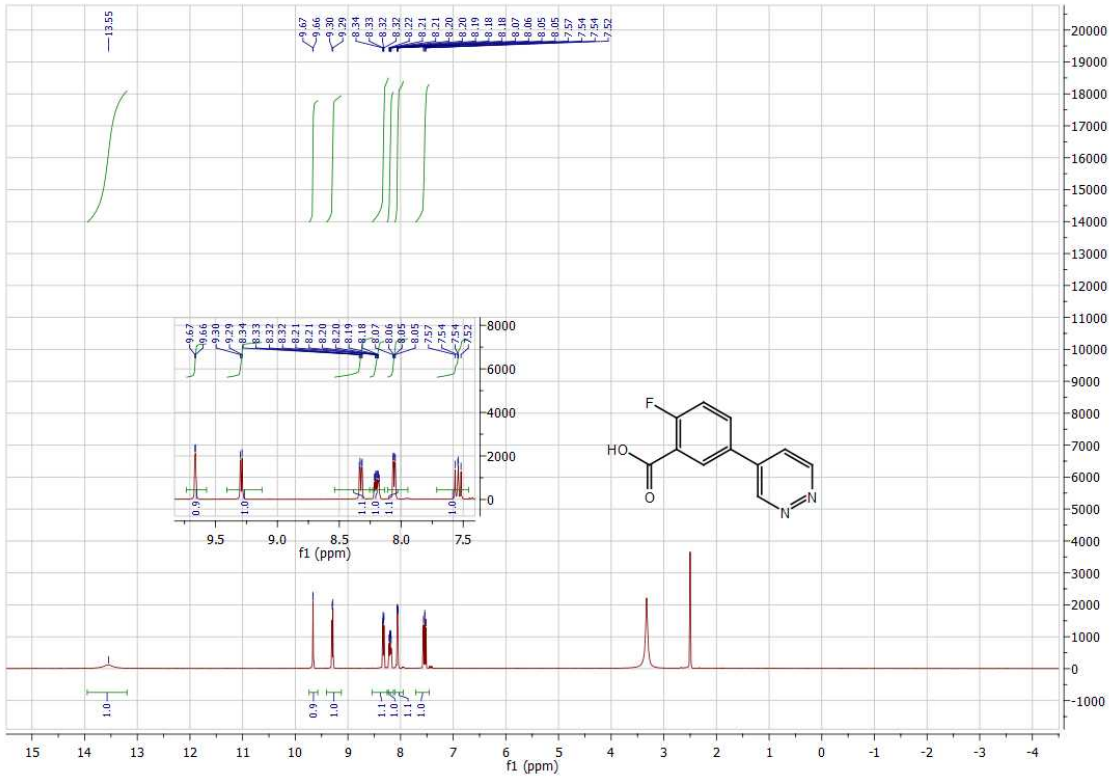
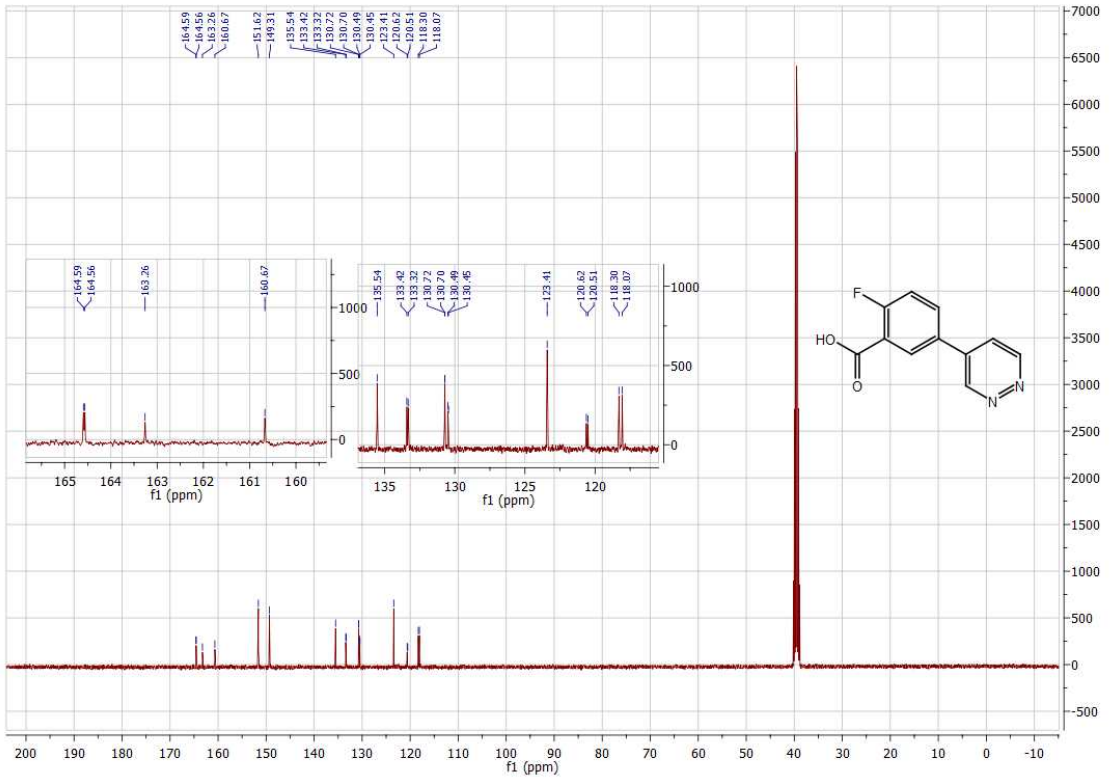
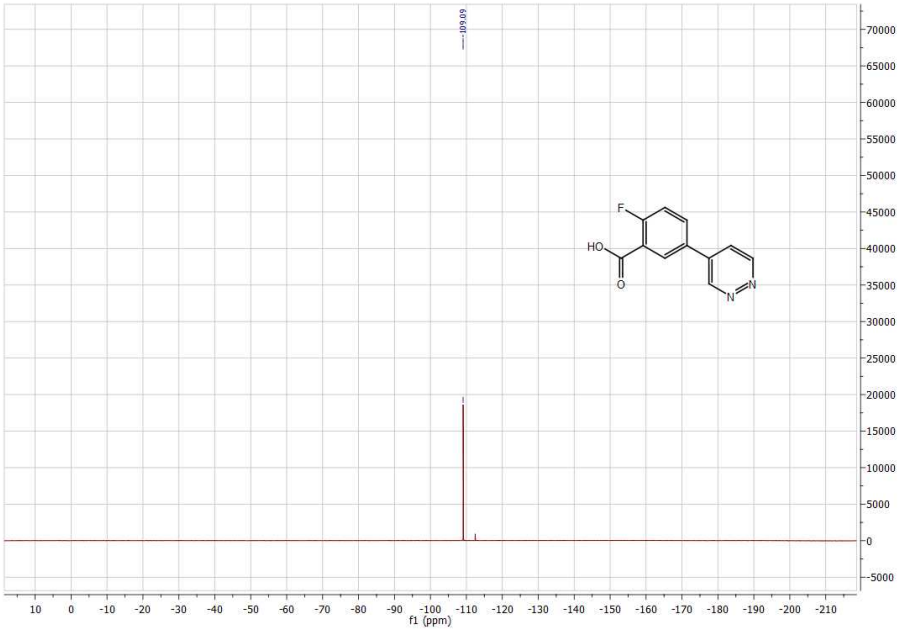


WM-2474-Intermediates







Qualitative Compound Report

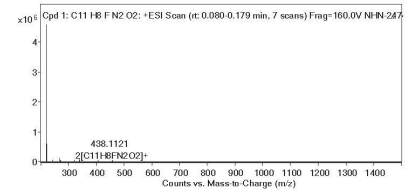
Data File	NHN-2474 Pre.d	Sample Name	NHN-2474 Pre
Sample Type	Sample	Position	P1-B7
Instrument Name	Instrument 1	User Name	Dr Jason Dang
Acq Method	Monash_Direct.m	Acquired Time	30-Nov-17 8:59:20 PM
IRM Calibration Status	Success	DA Method	Monash_Accuracy.m
Comment			
Sample Group		Info.	
Formula	C11H8FN2O2	Stream Name	LC 1
Acquisition SW Version	6200 series TOF/6500 series Q-TOF B.06.01 (B6172.SP1)		

Compound Table

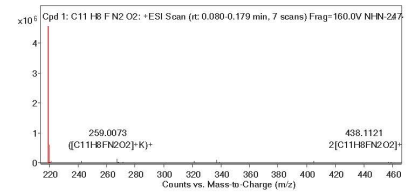
Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)
Cpd 1: C11 H8 F N2 O2	0.096	219.0572	4601769	C11 H8 F N2 O2	219.057	1.09

Compound Label	m/z	RT	Algorithm	Mass
Cpd 1: C11 H8 F N2 O2	219.0567	0.096	Find By Formula	219.0572

MS Spectrum



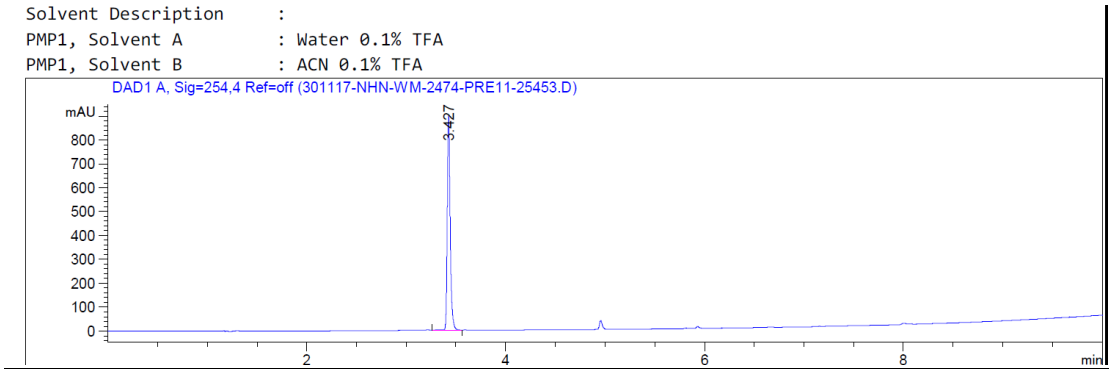
MS Zoomed Spectrum



MS Spectrum Peak List

m/z	Calc m/z	Diff(ppm)	z	Abund	Formula	Ion
219.0567	219.0564	-1.04	1	4601769.15	C11H8FN2O2	M+
259.0217	258.0201	-5.94	1	916.16	C11H8FN2O2	(M+K)+
438.1121	438.1134	3.11	1	1035.79	C11H8FN2O2	2M+

--- End Of Report ---


Solvent Description :
PMP1, Solvent A : Water 0.1% TFA
PMP1, Solvent B : ACN 0.1% TFA
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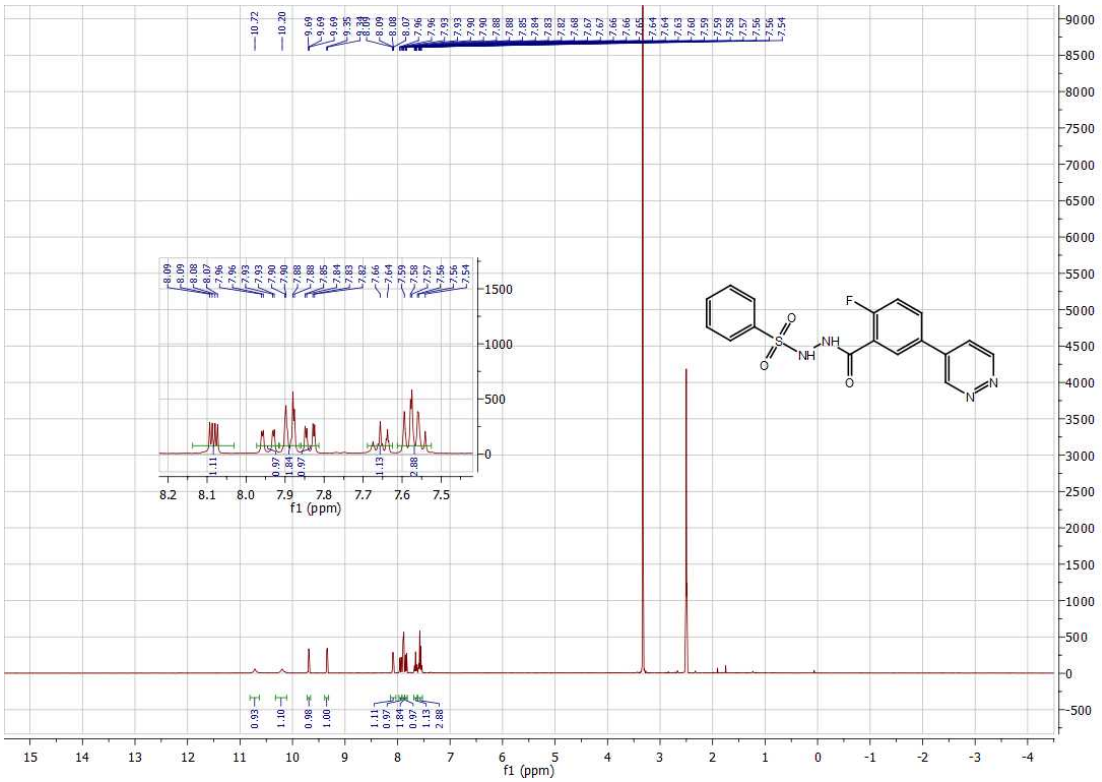
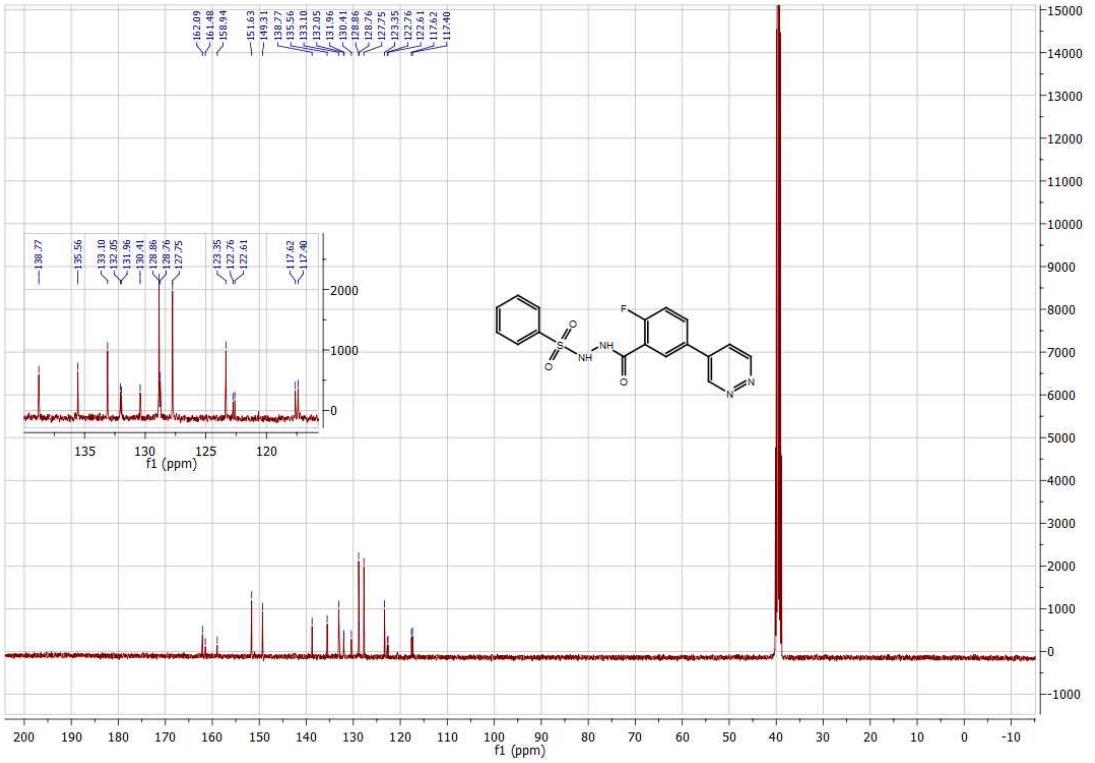
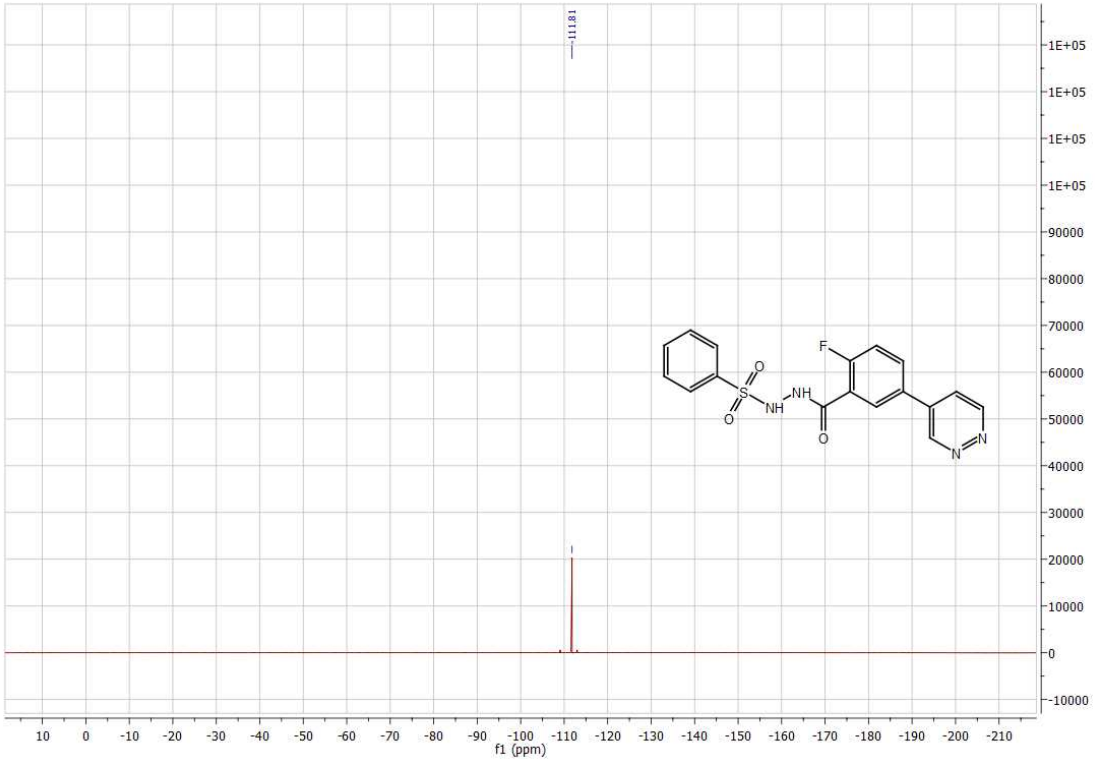
mAU
3.427
min

WM-2474


f1 (ppm)
F
O
S
NH
NH
O
N
N


162.09
161.48
158.94
151.63
149.31
138.77
135.56
133.10
132.05
131.96
130.41
128.86
128.76
127.75
123.35
122.76
122.61
117.62
117.40
f1 (ppm)
f1 (ppm)


-111.81
f1 (ppm)

Qualitative Compound Report

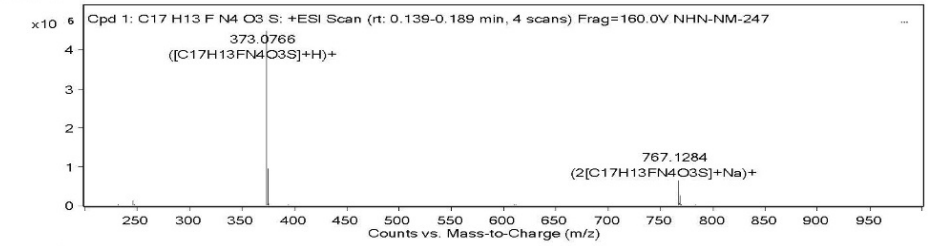
Data File	NHN-NM-2474.d	Sample Name	NHN-NM-2474
Sample Type	Sample	Position	P2-C9
Instrument Name	Instrument 1	User Name	Dr Jason Dang
Acq Method	Monash_Direct.m	Acquired Time	26-Sep-17 1:18:36 PM
IRM Calibration Status	Success	DA Method	Monash_Accuracy.m
Comment			
Sample Group		Info.	
Formula	C13H13FN4O3S	Stream Name	LC 1
Acquisition SW Version	6200 series TOF/6500 series Q-TOF B.06.01 (B6172.SP1)		

Compound Table

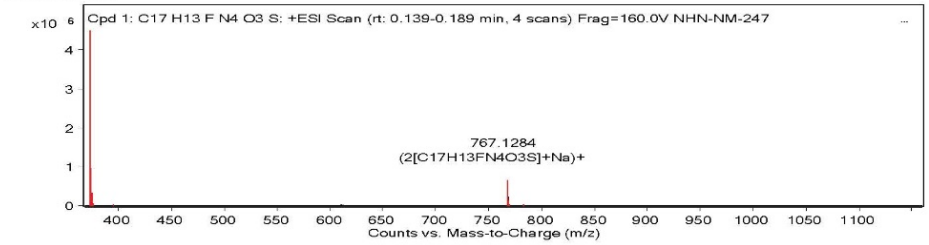
Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)
Cpd 1: C17 H13 F N4 O3 S	0.169	372.0694	4456966	C17H13FN4O3S	372.0692	0.49

Compound Label	m/z	RT	Algorithm	Mass
Cpd 1: C17 H13 F N4 O3 S	373.0766	0.169	Find By Formula	372.0694

MS Spectrum



MS Zoomed Spectrum



MS Spectrum Peak List

m/z	Calc. m/z	Diff (ppm)	z	Abund	Formula	Ion
373.0766	373.0765	0.66	1	4456966.67	C17H13FN4O3S	(M+H)+
395.0585	395.0585	1.56	1	25868.79	C17H13FN4O3S	(M+Na)+
767.1284	767.1277	0.87	1	640582.57	C17H13FN4O3S	(2M+Na)+

End Of Report

Agilent Technologies

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Solvent Description     :
PMP1, Solvent A         : Water 0.1% TFA
PMP1, Solvent B         : ACN 0.1% TFA
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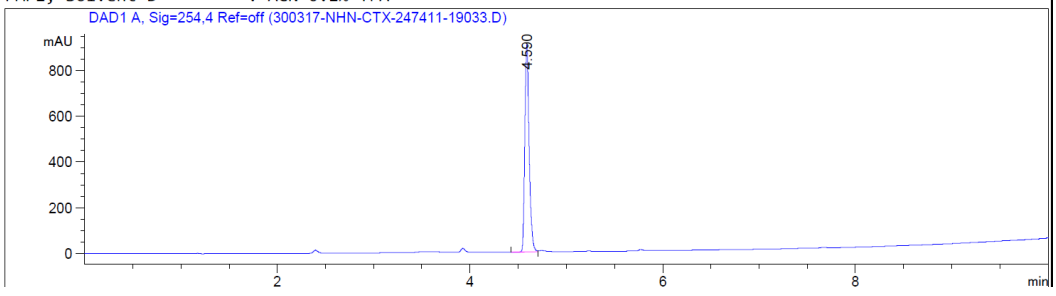

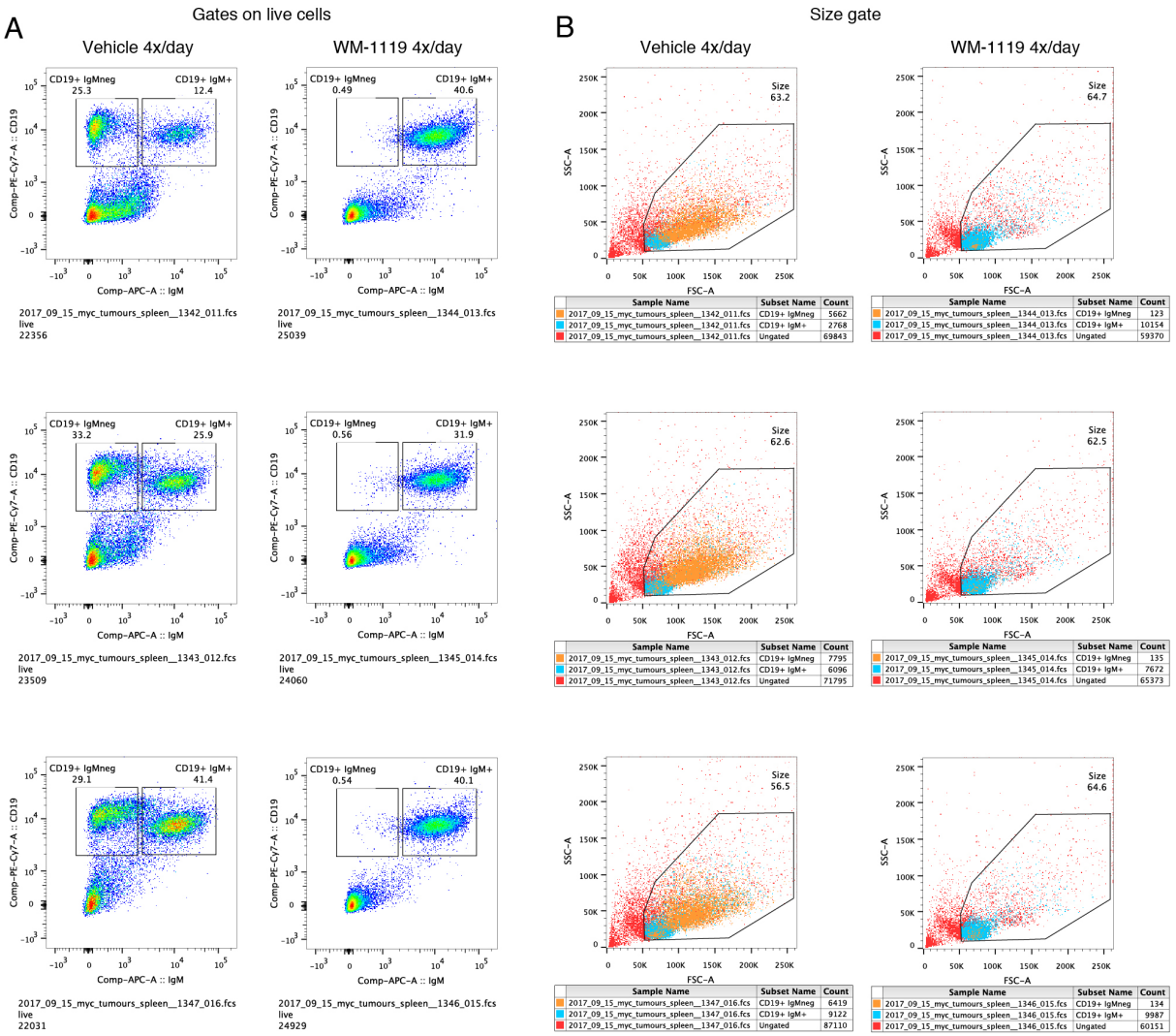

Gating strategy used in flow cytometry analysis of spleen and peripheral blood.

Supplementary Table 1: Half-inhibitory concentration [IC_{50} in µM] of WM-8014 determined in the biochemical assay using Alpha Screen technology, compared to the HTS hit CTx-0124143 and the intermediate of medicinal chemistry optimization WM-0550, as well as the inactive control compound WM-2474. Histone acetyltransferases were assayed using AcCoA at the K_M experimentally determined for each enzyme.

Compound	KAT6A	KAT6B	KAT5	KAT7	KAT8	KAT2A	KAT2B	KAT3A	KAT3B
CTx-0124143	0.49	1. 7	32.7	39.6	125	125	125	125	125
WM-0550	0.016	0.147	1.4	5.5	75	125	125	125	78
WM-8014	0.0082	0.028	0.224	0.342	11	125	125	125	111
WM-2474	125	125	125	125	125	125	125	125	125
Assay conditions									
AcCoA [µM]	0.4	0.4	5	1	0.4	1	4	4	0.4
Substrate	H4 peptide	H4 peptide	H4 peptide	H4 FL	H4 peptide	H4 peptide	H4 peptide	H4 peptide	H4 peptide
Antibody	Abcam ab15823	Abcam ab15823	Abcam ab15823	Abcam ab15823	Abcam ab15823	Abcam ab15823	Abcam ab15823	Abcam ab15823	Abcam ab15823

Supplementary Table 2: KAT6A inhibition summary, physicochemical parameters / drug-likeness of key compounds.

	IC_{50} [a] (μM)	K_D [b] (μM)	MW [c]	cLog P [c]	cLogD $_{pH\ 7.4}$ [c]	tPSA [c] (Å)	pKa [d]	$Sol_{pH6.5}$ [e] (μM)	Medium Binding [f] (% bound)	Medium Stability [g] (% remaining)	A-B P_{app} [h] (10^{-6} cm/s)
CTx-0124143	0.49	n.d.	344	3.0	2.7	72.5	5.4	10-20	88.5%	87.5%	51 ± 9.9
WM-2474	>125	n.d.	372	1.4	1.1	98.3	5.6	34-67	16.4%	90.5%	4.9 ± 0.3
WM-550	0.016	0.014	370	3.6	3.3	72.5	5.4	8-17	92.1%	87.7%	52 ± 6.6
WM-8014	0.0082	0.0051	384	4.1	3.8	72.5	6.8	8-16	97.5%	100%	78 ± 12.5
WM-1119	0.0063	0.0021	389	2.4	2.7	88.2	6.8	64-128	75.0%	100%	60.0 ± 6.1

[a] Enzymatic activity assay data of KAT6A measured using Alpha Screen technology.
[b] K_D determined from a kinetic model, n.d., not determined.
[c] Calculated using the ChemAxon JChem for Excel software (ver 6.2).
[d] Experimentally determined by potentiometric titration as described previously[41]
[e] Kinetic solubility determined by nephelometry, as described previously.[42]
[f] Percentage binding in cell culture medium (10% FCS).
[g] Stability in cell culture medium incubated at 37°C for 48 h.
[h] Permeability across Caco-2 cells in apical to basolateral (A to B) direction

Supplementary Table 3: Pharmacological profiling of WM-8014. Profiling of WM-8014 against a panel of (A) 70 CEREP binding assay targets[a], (B) 27 CEREP enzyme and/or cell-assay targets[a], (C) 4 supplementary custom-selected safety CEREP targets[a], (D) Reaction Biology 11 histone deacetylases[b], (E) Reaction Biology 7 phosphatases[b], (F) Reaction Biology 23 diverse kinases[b], (G), Reaction Biology 19 Caspases/Cathepsins[b], (H) Reaction Biology 4 SIRTs[b]. (I) Of these 158 different targets, summary of the 8 targets affected by more than 50% at 10 μM.

(A) Binding Assay (Eurofin Cerep 71 Receptor Diversity Panel)	Catalog Ref	Test Concentration. (M)	% Inhibition of Control Specific Binding	% of Control Specific Binding	Reference Compound	IC_{50} Ref (M)	Ki Ref (M)
A1 (h) (antagonist radioligand)	0002	1.0E-06	11	89.5	DPCPX	1.2E-09	7.6E-10
A1 (h) (antagonist radioligand)	0002	1.0E-05	56	43.8	DPCPX	1.2E-09	7.6E-10
A2A (h) (agonist radioligand)	0004	1.0E-06	12	88.2	NECA	2.4E-08	1.9E-08
A2A (h) (agonist radioligand)	0004	1.0E-05	43	56.6	NECA	2.4E-08	1.9E-08
A3 (h) (agonist radioligand)	0006	1.0E-06	44	56.1	IB-MECA	2.6E-10	1.6E-10
A3 (h) (agonist radioligand)	0006	1.0E-05	87	12.6	IB-MECA	2.6E-10	1.6E-10
alpha 1 (non-selective) (antagonist radioligand)	0008	1.0E-06	2	98.4	prazosin	4.2E-10	1.1E-10
alpha 1 (non-selective) (antagonist radioligand)	0008	1.0E-05	2	98.0	prazosin	4.2E-10	1.1E-10
alpha 2 (non-selective) (antagonist radioligand)	0011	1.0E-06	9	91.0	yohimbine	6.4E-08	2.7E-08
alpha 2 (non-selective) (antagonist radioligand)	0011	1.0E-05	0	99.9	yohimbine	6.4E-08	2.7E-08
beta 1 (h) (agonist radioligand)	0018	1.0E-06	-5	105.3	atenolol	3.6E-07	2.0E-07
beta 1 (h) (agonist radioligand)	0018	1.0E-05	-2	102.1	atenolol	3.6E-07	2.0E-07
beta 2 (h) (agonist radioligand)	0020	1.0E-06	-12	112.0	ICI 118551	1.1E-09	3.8E-10
beta 2 (h) (agonist radioligand)	0020	1.0E-05	-1	101.3	ICI 118551	1.1E-09	3.8E-10
AT1 (h) (antagonist radioligand)	0024	1.0E-06	0	99.6	saralasin	7.1E-10	3.6E-10
AT1 (h) (antagonist radioligand)	0024	1.0E-05	-12	111.8	saralasin	7.1E-10	3.6E-10
AT2 (h) (agonist radioligand)	0026	1.0E-06	10	90.1	angiotensin-II	2.8E-10	1.4E-10
AT2 (h) (agonist radioligand)	0026	1.0E-05	2	98.3	angiotensin-II	2.8E-10	1.4E-10
BZD (central) (agonist radioligand)	0028	1.0E-06	-17	116.8	diazepam	9.1E-09	7.7E-09
BZD (central) (agonist radioligand)	0028	1.0E-05	-12	112.5	diazepam	9.1E-09	7.7E-09
B1 (h) (agonist radioligand)	1189	1.0E-06	-8	107.8	desArg10-KD	4.9E-10	9.6E-11
B1 (h) (agonist radioligand)	1189	1.0E-05	-5	105.4	desArg10-KD	4.9E-10	9.6E-11
B2 (h) (agonist radioligand)	0033	1.0E-06	-13	113.1	NPC 567	2.3E-08	1.2E-08
B2 (h) (agonist radioligand)	0033	1.0E-05	-7	107.0	NPC 567	2.3E-08	1.2E-08
CB1 (h) (agonist radioligand)	0036	1.0E-06	6	94.1	CP 55940	7.8E-10	6.9E-10
CB1 (h) (agonist radioligand)	0036	1.0E-05	10	89.7	CP 55940	7.8E-10	6.9E-10

CB2 (h) (agonist radioligand)	0037	1.0E-06	3	97.4	WIN 55212-2	1.7E-09	1.1E-09
CB2 (h) (agonist radioligand)	0037	1.0E-05	36	64.4	WIN 55212-2	1.7E-09	1.1E-09
CCK1 (CCKA) (h) (agonist radioligand)	0039	1.0E-06	-7	106.9	CCK-8s	6.9E-11	5.1E-11
CCK1 (CCKA) (h) (agonist radioligand)	0039	1.0E-05	13	87.3	CCK-8s	6.9E-11	5.1E-11
CCK2 (CCKB) (h) (agonist radioligand)	0041	1.0E-06	-5	104.8	CCK-8s	2.3E-10	9.1E-11
CCK2 (CCKB) (h) (agonist radioligand)	0041	1.0E-05	15	85.0	CCK-8s	2.3E-10	9.1E-11
CRF1 (h) (agonist radioligand)	1467	1.0E-06	-9	109.1	sauvagine	1.7E-10	1.0E-10
CRF1 (h) (agonist radioligand)	1467	1.0E-05	-3	102.7	sauvagine	1.7E-10	1.0E-10
D1 (h) (antagonist radioligand)	0044	1.0E-06	-10	110.2	SCH 23390	6.0E-10	2.4E-10
D1 (h) (antagonist radioligand)	0044	1.0E-05	5	94.8	SCH 23390	6.0E-10	2.4E-10
D2S (h) (antagonist radioligand)	0046	1.0E-06	-20	120.0	(+)butaclamol	2.0E-09	6.6E-10
D2S (h) (antagonist radioligand)	0046	1.0E-05	-5	105.3	(+)butaclamol	2.0E-09	6.6E-10
D3 (h) (antagonist radioligand)	0048	1.0E-06	2	97.9	(+)butaclamol	1.7E-09	3.7E-10
D3 (h) (antagonist radioligand)	0048	1.0E-05	2	98.1	(+)butaclamol	1.7E-09	3.7E-10
D4.4 (h) (antagonist radioligand)	0049	1.0E-06	-4	103.8	clozapine	5.3E-08	2.1E-08
D4.4 (h) (antagonist radioligand)	0049	1.0E-05	3	97.1	clozapine	5.3E-08	2.1E-08
ETA (h) (agonist radioligand)	0054	1.0E-06	4	95.9	endothelin-1	6.5E-11	3.2E-11
ETA (h) (agonist radioligand)	0054	1.0E-05	-6	106.1	endothelin-1	6.5E-11	3.2E-11
ETB (h) (agonist radioligand)	0056	1.0E-06	13	86.6	endothelin-3	4.9E-11	2.8E-11
ETB (h) (agonist radioligand)	0056	1.0E-05	5	94.6	endothelin-3	4.9E-11	2.8E-11
GABA (non-selective) (agonist radioligand)	0057	1.0E-06	2	97.7	GABA	2.1E-08	1.2E-08
GABA (non-selective) (agonist radioligand)	0057	1.0E-05	-11	111.1	GABA	2.1E-08	1.2E-08
AMPA (agonist radioligand)	0064	1.0E-06	-11	111.5	L-glutamate	3.6E-07	3.3E-07
AMPA (agonist radioligand)	0064	1.0E-05	-11	111.0	L-glutamate	3.6E-07	3.3E-07
kainate (agonist radioligand)	0065	1.0E-06	18	81.9	kainic acid	2.5E-08	2.0E-08
kainate (agonist radioligand)	0065	1.0E-05	11	88.7	kainic acid	2.5E-08	2.0E-08
NMDA (antagonist radioligand)	0066	1.0E-06	-6	105.8	CGS 19755	1.8E-07	1.5E-07
NMDA (antagonist radioligand)	0066	1.0E-05	-2	102.4	CGS 19755	1.8E-07	1.5E-07
H1 (h) (antagonist radioligand)	0870	1.0E-06	-10	109.7	pyrilamine	2.8E-09	1.8E-09
H1 (h) (antagonist radioligand)	0870	1.0E-05	11	89.4	pyrilamine	2.8E-09	1.8E-09
H2 (h) (antagonist radioligand)	1208	1.0E-06	-20	120.0	cimetidine	4.9E-07	4.8E-07
H2 (h) (antagonist radioligand)	1208	1.0E-05	-12	111.6	cimetidine	4.9E-07	4.8E-07
H3 (h) (agonist radioligand)	1332	1.0E-06	-7	107.4	(R)alpha -Me-histamine	2.3E-09	5.5E-10
H3 (h) (agonist radioligand)	1332	1.0E-05	19	81.3	(R)alpha -Me-histamine	2.3E-09	5.5E-10

I2 (antagonist radioligand)	0081	1.0E-06	-3	102.8	idazoxan	7.4E-09	4.9E-09
I2 (antagonist radioligand)	0081	1.0E-05	8	92.3	idazoxan	7.4E-09	4.9E-09
CysLT1 (LTD4) (h) (agonist radioligand)	0086	1.0E-06	9	91.4	LTD4	3.7E-10	1.6E-10
CysLT1 (LTD4) (h) (agonist radioligand)	0086	1.0E-05	-2	102.0	LTD4	3.7E-10	1.6E-10
MC4 (h) (agonist radioligand)	0420	1.0E-06	0	100.2	NDP-alpha -MSH	3.6E-10	3.3E-10
MC4 (h) (agonist radioligand)	0420	1.0E-05	3	97.0	NDP-alpha -MSH	3.6E-10	3.3E-10
MT1 (ML1A) (h) (agonist radioligand)	1538	1.0E-06	20	79.8	melatonin	3.0E-10	2.4E-10
MT1 (ML1A) (h) (agonist radioligand)	1538	1.0E-05	65	34.6	melatonin	3.0E-10	2.4E-10
M (non-selective) (antagonist radioligand)	0089	1.0E-06	-5	105.2	atropine	4.7E-10	7.8E-11
M (non-selective) (antagonist radioligand)	0089	1.0E-05	-13	113.0	atropine	4.7E-10	7.8E-11
NK1 (h) (agonist radioligand)	0100	1.0E-06	5	94.8	[Sar9,Met(O2)11] -SP	2.5E-10	1.1E-10
NK1 (h) (agonist radioligand)	0100	1.0E-05	44	56.1	[Sar9,Met(O2)11] -SP	2.5E-10	1.1E-10
NK2 (h) (agonist radioligand)	0102	1.0E-06	0	99.7	[Nleu10]-NKA (4-10)	5.0E-09	2.7E-09
NK2 (h) (agonist radioligand)	0102	1.0E-05	-6	106.2	[Nleu10]-NKA (4-10)	5.0E-09	2.7E-09
NK3 (h) (antagonist radioligand)	0104	1.0E-06	-25	124.9	SB 222200	6.1E-09	3.3E-09
NK3 (h) (antagonist radioligand)	0104	1.0E-05	19	81.1	SB 222200	6.1E-09	3.3E-09
Y (non-selective) (agonist radioligand)	0105	1.0E-06	13	87.2	NPY	2.0E-10	1.3E-10
Y (non-selective) (agonist radioligand)	0105	1.0E-05	-16	115.7	NPY	2.0E-10	1.3E-10
N neuronal alpha 4beta 2 (h) (agonist radioligand)	3029	1.0E-06	0	100.5	nicotine	3.1E-09	1.0E-09
N neuronal alpha 4beta 2 (h) (agonist radioligand)	3029	1.0E-05	-5	104.9	nicotine	3.1E-09	1.0E-09
opioid (non-selective) (antagonist radioligand)	0112	1.0E-06	-5	105.3	naloxone	1.2E-09	8.3E-10
opioid (non-selective) (antagonist radioligand)	0112	1.0E-05	2	97.5	naloxone	1.2E-09	8.3E-10
NOP (ORL1) (h) (agonist radioligand)	0358	1.0E-06	9	90.8	nociceptin	1.1E-09	1.4E-10
NOP (ORL1) (h) (agonist radioligand)	0358	1.0E-05	4	95.6	nociceptin	1.1E-09	1.4E-10
PPARgamma (h) (agonist radioligand)	0641	1.0E-06	22	78.0	rosiglitazone	1.6E-08	8.4E-09
PPARgamma (h) (agonist radioligand)	0641	1.0E-05	76	24.3	rosiglitazone	1.6E-08	8.4E-09
PCP (antagonist radioligand)	0124	1.0E-06	-2	102.3	MK 801	6.1E-09	3.5E-09
PCP (antagonist radioligand)	0124	1.0E-05	1	98.6	MK 801	6.1E-09	3.5E-09
EP2 (h) (agonist radioligand)	1955	1.0E-06	-4	103.9	PGE2	2.0E-09	9.9E-10
EP2 (h) (agonist radioligand)	1955	1.0E-05	9	90.6	PGE2	2.0E-09	9.9E-10
IP (PGI2) (h) (agonist radioligand)	2230	1.0E-06	-12	112.3	iloprost	8.7E-09	5.0E-09
IP (PGI2) (h) (agonist radioligand)	2230	1.0E-05	14	85.8	iloprost	8.7E-09	5.0E-09
P2X (agonist radioligand)	0127	1.0E-06	-11	111.4	alpha ,beta -MeATP	5.4E-09	2.5E-09

P2X (agonist radioligand)	0127	1.0E-05	7	93.5	alpha ,beta - MeATP	5.4E-09	2.5E-09
P2Y (agonist radioligand)	0128	1.0E-06	11	88.7	dATPalpha S	4.4E-08	2.2E-08
P2Y (agonist radioligand)	0128	1.0E-05	6	93.8	dATPalpha S	4.4E-08	2.2E-08
Serotonin (5-Hydroxytryptamine) 5-HT1, Non-Selective	3970	1.0E-06	6	94.0	Serotonin (5-HT)	3.3E-09	7.6E-10
Serotonin (5-Hydroxytryptamine) 5-HT1, Non-Selective	3970	1.0E-05	1	99.5	Serotonin (5-HT)	3.3E-09	7.6E-10
sigma (non-selective) (h) (agonist radioligand)	3500	1.0E-06	-1	101.0	haloperidol	3.1E-08	2.5E-08
sigma (non-selective) (h) (agonist radioligand)	3500	1.0E-05	10	89.5	haloperidol	3.1E-08	2.5E-08
GR (h) (agonist radioligand)	0469	1.0E-06	2	97.5	dexamethasone	2.3E-09	1.1E-09
GR (h) (agonist radioligand)	0469	1.0E-05	17	83.3	dexamethasone	2.3E-09	1.1E-09
ER (non-selective) (h) (agonist radioligand)	0152	1.0E-06	-2	102.0	17-beta -estradiol	2.3E-10	7.7E-11
ER (non-selective) (h) (agonist radioligand)	0152	1.0E-05	1	99.5	17-beta -estradiol	2.3E-10	7.7E-11
PR (h) (agonist radioligand)	2341	1.0E-06	4	96.1	promegestone	4.3E-10	3.4E-10
PR (h) (agonist radioligand)	2341	1.0E-05	28	72.2	promegestone	4.3E-10	3.4E-10
AR (h) (agonist radioligand)	0933	1.0E-06	16	83.7	mibolerone	4.2E-09	1.9E-09
AR (h) (agonist radioligand)	0933	1.0E-05	20	80.0	mibolerone	4.2E-09	1.9E-09
TRH1 (h) (agonist radioligand)	1616	1.0E-06	-12	111.5	TRH	3.0E-08	2.0E-08
TRH1 (h) (agonist radioligand)	1616	1.0E-05	16	84.4	TRH	3.0E-08	2.0E-08
V1a (h) (agonist radioligand)	0159	1.0E-06	3	96.8	[d(CH2)51, Tyr(Me)2]-AVP	1.9E-09	1.2E-09
V1a (h) (agonist radioligand)	0159	1.0E-05	-2	101.5	[d(CH2)51, Tyr(Me)2]-AVP	1.9E-09	1.2E-09
V2 (h) (agonist radioligand)	0497	1.0E-06	-17	116.8	AVP	7.1E-10	5.1E-10
V2 (h) (agonist radioligand)	0497	1.0E-05	-22	121.8	AVP	7.1E-10	5.1E-10
Ca2+ channel (L, dihydropyridine site) (antagonist radioligand)	0161	1.0E-06	7	93.2	nitrendipine	1.2E-10	7.4E-11
Ca2+ channel (L, dihydropyridine site) (antagonist radioligand)	0161	1.0E-05	-30	130.0	nitrendipine	1.2E-10	7.4E-11
Ca2+ channel (L, diltiazem site) (benzothiazepines) (antagonist radioligand)	0162	1.0E-06	-24	124.0	diltiazem	3.3E-08	2.6E-08
Ca2+ channel (L, diltiazem site) (benzothiazepines) (antagonist radioligand)	0162	1.0E-05	-21	120.9	diltiazem	3.3E-08	2.6E-08
Ca2+ channel (L, verapamil site) (phenylalkylamine) (antagonist radioligand)	0163	1.0E-06	-8	107.7	D 600	5.0E-08	2.5E-08
Ca2+ channel (L, verapamil site) (phenylalkylamine) (antagonist radioligand)	0163	1.0E-05	-14	114.2	D 600	5.0E-08	2.5E-08
KATP channel (antagonist radioligand)	0165	1.0E-06	-10	109.7	glibenclamide	1.4E-10	4.6E-11

KATP channel (antagonist radioligand)	0165	1.0E-05	-16	116.1	glibenclamide	1.4E-10	4.6E-11
KV channel (antagonist radioligand)	0166	1.0E-06	6	94.1	alpha - dendrotoxin	8.2E-11	6.6E-11
KV channel (antagonist radioligand)	0166	1.0E-05	-4	104.1	alpha - dendrotoxin	8.2E-11	6.6E-11
SKCa channel (antagonist radioligand)	0167	1.0E-06	8	92.2	apamin	1.1E-11	5.3E-12
SKCa channel (antagonist radioligand)	0167	1.0E-05	0	99.6	apamin	1.1E-11	5.3E-12
Na+ channel (site 2) (antagonist radioligand)	0169	1.0E-06	7	92.9	veratridine	1.0E-05	9.3E-06
Na+ channel (site 2) (antagonist radioligand)	0169	1.0E-05	14	85.6	veratridine	1.0E-05	9.3E-06
Cl- channel (GABA-gated) (antagonist radioligand)	0170	1.0E-06	5	94.8	picrotoxinin	1.6E-07	1.3E-07
Cl- channel (GABA-gated) (antagonist radioligand)	0170	1.0E-05	27	73.0	picrotoxinin	1.6E-07	1.3E-07
norepinephrine transporter (h) (antagonist radioligand)	0355	1.0E-06	-2	101.7	protriptyline	2.9E-09	2.1E-09
norepinephrine transporter (h) (antagonist radioligand)	0355	1.0E-05	17	83.4	protriptyline	2.9E-09	2.1E-09
dopamine transporter (h) (antagonist radioligand)	0052	1.0E-06	7	93.0	BTCP	8.2E-09	4.3E-09
dopamine transporter (h) (antagonist radioligand)	0052	1.0E-05	56	44.0	BTCP	8.2E-09	4.3E-09
GABA transporter (antagonist radioligand)	0060	1.0E-06	2	97.9	nipecotic acid	6.6E-07	6.6E-07
GABA transporter (antagonist radioligand)	0060	1.0E-05	15	84.7	nipecotic acid	6.6E-07	6.6E-07
choline transporter (CHT1) (h) (antagonist radioligand)	1552	1.0E-06	-8	107.9	hemicholinium-3	9.4E-09	5.3E-09
choline transporter (CHT1) (h) (antagonist radioligand)	1552	1.0E-05	-13	113.1	hemicholinium-3	9.4E-09	5.3E-09
5-HT transporter (h) (antagonist radioligand)	0439	1.0E-06	-11	110.9	imipramine	3.0E-09	1.4E-09
5-HT transporter (h) (antagonist radioligand)	0439	1.0E-05	-15	115.0	imipramine	3.0E-09	1.4E-09

(Bi). 27 Enzyme & Cell-Based Assay (Eurofin Cerep Diversity Panel)	Catalog Ref	Test Concentration (M)	% Inhibition of Control Values	% of Control Values	Reference Compound	IC_{50} Ref (M)	nH Ref
COX1 (h)	0726	1.0E-06	21	78.8	diclofenac	3.7E-09	0.7
COX1 (h)	0726	1.0E-05	9	91.3	diclofenac	3.7E-09	0.7
5-lipoxygenase (h)	0772	1.0E-06	-10	109.6	NDGA	1.7E-07	2.0
5-lipoxygenase (h)	0772	1.0E-05	0	99.6	NDGA	1.7E-07	2.0
PDE1B (h)	2431	1.0E-06	-2	101.9	nitrendipine	2.1E-06	1.7
PDE1B (h)	2431	1.0E-05	11	89.2	nitrendipine	2.1E-06	1.7
PDE2A1 (h)	2426	1.0E-06	-5	105.3	EHNA	2.3E-06	1.5
PDE2A1 (h)	2426	1.0E-05	-15	114.8	EHNA	2.3E-06	1.5
PDE3A (h)	2432	1.0E-06	-5	104.7	milrinone	4.4E-07	0.7
PDE3A (h)	2432	1.0E-05	3	97.1	milrinone	4.4E-07	0.7
PDE4D2 (h)	2434	1.0E-06	-4	104.3	rolipram	6.5E-08	0.8
PDE4D2 (h)	2434	1.0E-05	-8	107.6	rolipram	6.5E-08	0.8
PDE5 (h) (non-selective)	0204	1.0E-06	5	94.5	dipyridamole	1.4E-06	0.8

PDE5 (h) (non-selective)	0204	1.0E-05	12	88.0	dipyridamole	1.4E-06	0.8
phosphatase 1B (h) (PTP1B)	2593	1.0E-06	7	92.7	Ammonium molybdate	1.6E-07	4.0
phosphatase 1B (h) (PTP1B)	2593	1.0E-05	8	92.0	Ammonium molybdate	1.6E-07	4.0
phosphatase CDC25A (h)	2561	1.0E-06	4	96.1	NSC 663284	4.1E-07	2.9
phosphatase CDC25A (h)	2561	1.0E-05	9	91.2	NSC 663284	4.1E-07	2.9
PKCalpha (h)	0348	1.0E-06	-1	100.7	Bis 10	2.7E-09	2.7
PKCalpha (h)	0348	1.0E-05	-1	101.2	Bis 10	2.7E-09	2.7
acetylcholinesterase (h)	0363	1.0E-06	-8	107.9	galanthamine	5.3E-07	0.6
acetylcholinesterase (h)	0363	1.0E-05	-4	104.1	galanthamine	5.3E-07	0.6
COMT (catechol- O-methyl transferase)	0457	1.0E-06	-8	107.9	Ro 41-0960	5.4E-08	2.2
COMT (catechol- O-methyl transferase)	0457	1.0E-05	-6	106.5	Ro 41-0960	5.4E-08	2.2
GABA transaminase	0461	1.0E-06	5	94.8	O-(Carboxy-méthyl) hydroxyl-amine 0.5HCl	1.1E-07	1.0
GABA transaminase	0461	1.0E-05	14	85.9	O-(Carboxy-méthyl) hydroxyl-lamine 0.5HCl	1.1E-07	1.0
MAO-A (h)	0191	1.0E-06	2	98.2	clorgyline	2.5E-08	1.5
MAO-A (h)	0191	1.0E-05	-1	101.5	clorgyline	2.5E-08	1.5
MAO-B (h) recombinant enzyme	3477	1.0E-06	1	98.7	deprenyl	3.2E-08	1.4
MAO-B (h) recombinant enzyme	3477	1.0E-05	-1	101.2	deprenyl	3.2E-08	1.4
tyrosine hydroxylase	0214	1.0E-06	-16	116.2	3-iodo L-tyrosine	4.0E-07	1.1
tyrosine hydroxylase	0214	1.0E-05	-42	142.2	3-iodo L-tyrosine	4.0E-07	1.1
ATPase (Na+/K+)	2009	1.0E-06	1	98.7	ouabain	1.2E-06	1.4
ATPase (Na+/K+)	2009	1.0E-05	3	97.5	ouabain	1.2E-06	1.4
CENP-E (h)	2150	1.0E-06	-2	101.7	rose bengal	2.3E-06	2.7
CENP-E (h)	2150	1.0E-05	-1	101.1	rose bengal	2.3E-06	2.7
Eg5 (h)	2151	1.0E-06	-2	102.0	S-trityl-L-cysteine	3.6E-07	1.5
Eg5 (h)	2151	1.0E-05	-1	100.8	S-trityl-L-cysteine	3.6E-07	1.5
HDAC3 (h)	2083	1.0E-06	-3	103.4	trichostatin A	1.6E-08	1.6
HDAC3 (h)	2083	1.0E-05	-2	102.0	trichostatin A	1.6E-08	1.6
HDAC4 (h)	2493	1.0E-06	-9	109.1	trichostatin A	2.9E-06	1.0
HDAC4 (h)	2493	1.0E-05	-17	117.0	trichostatin A	2.9E-06	1.0
HDAC6 (h)	2495	1.0E-06	-11	110.6	trichostatin A	9.0E-09	1.4
HDAC6 (h)	2495	1.0E-05	-11	110.9	trichostatin A	9.0E-09	1.4
HDAC11 (h)	2663	1.0E-06	-11	110.9	scriptaid	7.4E-06	1.0
HDAC11 (h)	2663	1.0E-05	-16	115.8	scriptaid	7.4E-06	1.0
sirtuin 1 (h) (inhibitor effect)	2581	1.0E-06	15	85.1	suramin	5.6E-06	1.2
sirtuin 1 (h) (inhibitor effect)	2581	1.0E-05	-6	105.8	suramin	5.6E-06	1.2
sirtuin 2 (h) (inhibitor effect)	2582	1.0E-06	-5	105.1	suramin	1.9E-05	2.3
sirtuin 2 (h) (inhibitor effect)	2582	1.0E-05	-6	105.8	suramin	1.9E-05	2.3

(Bii). 27 Enzyme & Cell-Based Assay (Eurofin Cerep Diversity Panel, continued)	Catalog Ref	Test Concen-tration (M)	% Stimulat-ion Relative to Control	% Stimulat-ion Relative to Control	Reference Compound	EC_{50} Ref (M)	nH Ref
adenylyl cyclase (activator effect)	3002	1.0E-06	0	0.0	forskolin	6.6E-06	1.6
adenylyl cyclase (activator effect)	3002	1.0E-05	4	4.1	forskolin	6.6E-06	1.6
guanylyl cyclase (h) (activator effect)	3004	1.0E-06	0	0.2	sodium nitroprusside	6.1E-06	1.2
guanylyl cyclase (h) (activator effect)	3004	1.0E-05	0	0.0	sodium nitroprusside	6.1E-06	1.2

(Ci). Custom-selected extra Cerep Binding Assays	Catalog Ref	Test Concen-tration	% Inhibition of Control Specific Binding	1st / % of Control Specific Binding	Mean / % of Control Specific Binding	Reference Compound	IC_{50} Ref (M)	Ki Ref	nH Ref
M1 (h) (antagonist radioligand)	91	1.00E-05	-9	109	109	pirenzepine	2.50E-08	2.20E-08	1.4
mu (MOP) (h) (agonist radioligand)	118	1.00E-05	18	82.2	82.2	DAMGO	6.70E-10	2.70E-10	0.7
5-HT2B (h) (agonist radioligand)	1333	1.00E-05	54	46.4	46.4	(±)DOI	4.30E-09	2.10E-09	0.6

(Cii). Custom-selected extra Cerep Enzyme and Cell-Based Assays (continued)	Catalog Ref	Test Concen-tration	% Inhibition of Control Values	1st / % of Control Values	Mean / % of Control Values	Reference Compound	IC_{50} Ref	nH Ref
COX2 (h)	727	1.00E-05	11	88.7	88.7	NS 398	2.60E-08	1.4

(D) Percentage activity of HDACs after treatment with 10 μM WM-8014 tested in triplicate by Reaction Biology											
	HDAC1	HDAC2	HDAC3	HDAC4	HDAC5	HDAC6	HDAC7	HDAC8	HDAC9	HDAC10	HDAC11
WM-8014 10 µM	104 104 102	99 95 96	100 104 100	109 111 107	107 112 116	96 99 93	99 99 100	103 104 100	93 92 92	91 85 83	87 88 88
WM-8014 1 µM	105 105 104	100 99 101	103 103 103	119 115 119	106 106 108	99 107 104	84 94 90	106 106 99	100 99 104	92 96 94	89 92 89
Control IC_{50} (M): Trichostatin A	8.9E-09	1.6E-08	1.1E-08			2.4E-09		4.0E-07		3.8E-08	2.8E-08
Control IC_{50} (M): TMP269				3.2E-07	2.4E-07		7.8E-08		1.8E-08		

(E) Percentage activity of protein tyrosine phosphatases after treatment with 10 μM WM-8014 tested in triplicate by Reaction Biology							
	PTPN1/ PTP1B	PTPN2/ TC-PTP	PTPN12/ PTP-PEST	PTPN7/ LC-PTP	PTPRC/ CD45	PTPN6/ SHP1	PTPN11/ SHP2
WM-8014 10 µM	91 95 92	91 90 95	85 93 90	91 89 86	90 84 87	97 97 95	88 85 87
WM-8014 1 µM	106 104 105	100 105 101	106 101 109	105 103 100	87 84 85	101 99 99	93 96 91
Control PTP1B inhibitor IC_{50} (M)	1.1E-05	2.7E-05	8.3E-06	9.2E-06	5.2E-06	9.6E-06	1.3E-05

(F) Percentage activity of 23 diverse kinases after treatment with 10 μM or 1 µM WM-8014 tested in triplicate by Reaction Biology with [ATP] Km shown (μM)								
	BMX/ETK [50]	BTK [50]	c-Kit [100]	CK1g1 [20]	CK1g2 [30]	CK1g3 [20]	EGFR [1]	ERK2 / MAPK1 [50]
WM-8014 10 µM	110 106 99	100 99 102	99 98 95	97 94 103	95 90 100	98 97 107	99 99 101	94 93 102
WM-8014 1 µM	107 106 109	107 105 103	95 95 100	105 101 112	95 95 104	97 92 106	99 99 100	100 98 109
Staurosporine control IC_{50} (M)	1.0E-08	2.3E-08	5.5E-08	9.8E-06	2.2E-06	2.4E-06	4.0E-08	3.2E-05
	FGFR1 [5]	FGFR2 [5]	FGFR3 [30]	GSK3b [10]	IKKb / IKBKB [30]	ITK [30]	JAK3 [2.5]	JNK1 [15]
WM-8014 10 µM	102 102 102	100 100 103	102 102 98	98 98 101	99 98 101	99 97 104	97 97 100	100 98 102
WM-8014 1 µM	101 100 98	100 99 98	100 100 100	104 104 106	100 100 98	99 96 102	107 103 94	100 99 97
Staurosporine control IC_{50} (M)	2.5E-09	1.2E-09	4.2E-08	4.2E-09	5.6E-07	1.4E-08	8.1E-11	5.0E-07
	JNK2 [20]	JNK3 [30]	KDR / VEGFR2 [20]	MEK1 [2.5]	NEK2 [10]	TAK1 [20]	TXK [30]	
WM-8014 10 µM	105 105 104	100 99 102	102 101 104	100 100 99	95 94 92	98 98 97	96 96 94	
WM-8014 1 µM	104 103 106	115 114 118	107 106 102	103 102 105	107 107 116	100 98 103	103 101 107	
Staurosporine control IC_{50} (M)	4.3E-06		1.8E-08	1.4E-08	6.0E-07	4.7E-08	3.4E-08	
JNKI VIII IC_{50} (M)		2.2E-07						

(G) Percentage activity of cathepsins and caspases after treatment with 10 μM or 1 μM WM-8014 tested in triplicate by Reaction Biology								
	Caspase 1	Caspase 2	Caspase 3	Caspase 4	Caspase 5	Caspase 6	Caspase 7	Caspase 8
WM-8014 10 µm	105 104 115	108 110 114	104 103 106	37 32 39	67 64 84	97 96 104	106 101 98	84 74 80
WM-8014 1 µm	99 93 104	103 101 100	111 105 111	73 60 66	80 83 90	111 109 113	116 104 119	110 104 102
Control Compound IC_{50} (M)	1.5E-08	2.5E-07	4.1E-10	2.2E-07	1.7E-09	2.6E-08	4.5E-10	1.8E-09
Control compound ID	IETD-CHO	IETD-CHO	DEVD-CHO	IETD-CHO	IETD-CHO	DEVD-CHO	DEVD-CHO	IETD-CHO
	Caspase 9	Caspase 10	Caspase 11	Caspase 14	Cathepsin B	Cathepsin C	Cathepsin H	Cathepsin L
WM-8014 10 µm	88 81 81	24 22 19	75 72 88	122 109 119	105 108 103	108 105 107	118 110 117	129 114 115
WM-8014 1 µm	101 91 94	54 48 51	81 75 87	89 77 91	105 104 102	105 106 106	118 106 114	127 125 126
Control Compound IC_{50} (M)	4.5E-08	3.8E-08	3.7E-07	6.8E-06	4.2E-09	4.3E-07	1.8E-07	2.5E-09
Control compound ID	IETD-CHO	IETD-CHO	IETD-CHO	WEHD-CHO	E64	E64	E64	E64

	Cathepsin S	Cathepsin V	Papain					
WM-8014 10 µm	115 102 110	227 193 228	133 108 124					
WM-8014 1 µm	113 107 111	84 81 87	122 100 120					
Control Compound IC_{50} (M)	2.1E-09	8.6E-09	3.4E-11					
Control compound ID	E64	E64	E64					

(H) Percentage activity of SIRTS after treatment with 10 μM or 1 μM WM-8014 tested in triplicate by Reaction Biology				
	SIRT 1	SIRT 2	SIRT 3	SIRT 5
WM-8014 10 µM	94 88 101	98 88 92	73 79 82	92 92 93
WM-8014 1 µM	97 100 103	92 96 94	86 89 85	96 98 96
Suramin IC_{50} (M)	9.5E-07	1.7E-05		
Nicotinamide IC_{50} (M)			1.6E-05	3.3E-05

(I)[c] Targets most affected (>50% effect at 10 μM)	% binding @ 10 μM	% binding @ 1μM
5HT2B *(h)* [agonist radioligand]	54	16
A1 *(h)* [antagonist radioligand]	56	11
A3 *(h)* [agonist radioligand]	87	44
MT1 (ML1A) *(h)* [agonist radioligand]	65	20
PPAR-g *(h)* [agonist radioligand]	76	22
Dopamine transporter *(h)* [antagonist radioligand]	56	7
	% activity @ 10μM	% activity @ 1μM
Caspase 4	37, 32, 39	73, 60, 66
Caspase 10	24, 22, 19	54, 48, 51

[a.] Eurofin Cerep 71 Receptor and 27 Enzyme Diversity Panel. The binding assay results are expressed as a percentage of control specific binding. The results of enzyme and uptake assays are expressed as a percentage of control specific activity.
[b.] Compounds were tested by Reaction Biology either in single does triplicate mode at 1 µM and 10 µM or 2 concentrations of 1 µM and 10µM according to Reaction Biology protocols.
[c.] Summary of receptor and enzyme activities affected by treatment with WM-8014 out of 158 tested targets. Note that biochemical inhibition of these enzymes occurs at a concentration that is at least 2 orders of magnitude higher than the IC_{50} of KAT6A inhibition in biochemical assays.

Supplementary Table 4: Data collection and refinement statistics of the WM-8014-MYST^{Cryst} and AcCoA-MYST^{Cryst} co-crystal structures.

Data collection	MYST^{Cryst} + WM-8014	MYST^{Cryst} + AcCoA
Temperature (K)	100	100
Space group	*P 2$_1$2$_1$ 2$_1$*	*P 2$_1$2$_1$ 2$_1$*
Unit cell parameters		
a, b, c (Å)	46.2, 56.9, 121.0	46.4, 57.9, 120.0
α, β, γ (^{o})	90.0, 90.0, 90.0	90.0, 90.0, 90.0
Data collection		
Resolution (Å)	46.23 - 1.85 (1.89 - 1.85)	46.38 - 1.95 (2.00 - 1.95)
No. of observations	136,644 (8,344)	173,236 (11,806)
No. of unique reflections	27,815 (1,669)	24,350 (1,662)
Redundancy	4.9 (5.0)	7.2 (7.4)
Data completeness (%)	99.4 (98.6)	99.9 (99.4)
I/σ_I	13.4 (2.2)	19.0 (3.0)
R_{merge} (%)	7.2 (69.8)	6.3 (61.8)
R_{meas} (%)	8.0 (77.9)	6.8 (66.6)
R_{pim} (%)	3.5 (33.8)	2.5 (24.7)
Refinement statistics		
Resolution (Å)	43.20 - 1.85	41.70 - 1.95
R_{work}	0.191	0.184
R_{free}	0.222	0.213
No. of reflections	27777	24293
Completeness (%)	99.1	99.8
No. of atoms		
Protein	2163	2211
Ligand	44	102
Water	144	154
R.m.s. deviations from ideality		
Bond angles (^{o})	1.059	1.223
Bond lengths (Å)	0.007	0.008
Average B-factor (Å^2)		
Protein	29.2	39.0
Ligand	21.7	39.6
Water	34.6	43.9
Ramachandran Plot		
Residues in most favored regions (%)	97.3	97.4
Outliers (%)	0	0.4

Values in parentheses are for the highest resolution shell.

Supplementary Table 5: Data collection and refinement statistics of the WM-1119-MYST^{Cryst} co-crystal structures.

Data collection	MYST^{Cryst} + WM-1119
Temperature (K)	100
Space group	*P* $2_1 2_1 2_1$
Unit cell parameters	
a, b, c (Å)	45.9, 56.1, 120.9
α, β, γ (°)	90.0, 90.0, 90.0
Data collection	
Resolution (Å)	45.89 - 2.13 (2.19 - 2.13)
No. of observations	129008 (9665)
No. of unique reflections	18176 (1409)
Redundancy	7.1 (6.9)
Data completeness (%)	99.7 (96.2)
I/σ_I	8.5 (1.8)
R_{merge} (%)	14.6 (86.5)
R_{meas} (%)	15.8 (93.4)
R_{pim} (%)	5.9 (34.9)
Refinement statistics	
Resolution (Å)	42.91 - 2.13
R_{work}	0.201
R_{free}	0.236
No. of reflections	18122
Completeness (%)	99.7
No. of atoms	
Protein	2285
Ligand	29
Water	135
R.m.s. deviations from ideality	
Bond angles (°)	0.650
Bond lengths (Å)	0.003
Average B-factor (Å^2)	
Protein	33.9
Ligand	22.5
Water	35.6
Ramachandran Plot	
Residues in most favored regions (%)	96.6
Outliers (%)	0.0

Supplementary Table 6: Pharmacological profiling of WM-1119. Profiling of WM-1119 against a panel of (A) 70 CEREP binding assay targets[a], (B) 28 CEREP enzyme and/or cell-assay targets[a], (C) 4 supplementary custom-selected safety CEREP targets[a], (D) Reaction Biology 11 histone deacetylases[b], (E) Reaction Biology 7 phosphatases[b], (F) Reaction Biology 23 diverse kinases[b], (G), Reaction Biology 19 Caspases/Cathepsins[b], (H) Reaction Biology 4 SIRTs[b]. Of these 159 different targets, no target was affected by more than 50% at 10 μM, demonstrating even less off-target activity that WM-8014 (Supplementary Table 3).

(A) Binding Assay (Eurofin Cerep 71 Receptor Diversity Panel)	Catalog Ref	Test Concentration. (M)	% Inhibition of Control Specific Binding	% of Control Specific Binding	Reference Compound	IC_{50} Ref (M)	Ki Ref (M)
A1 (h) (antagonist radioligand)	2	1E-06	-1	101.4	DPCPX	1.5E-09	9.4E-10
A1 (h) (antagonist radioligand)	2	1E-05	43	57.3	DPCPX	1.5E-09	9.4E-10
A2A (h) (agonist radioligand)	4	1E-06	6	94.3	NECA	3.6E-08	3.0E-08
A2A (h) (agonist radioligand)	4	1E-05	49	51.5	NECA	3.6E-08	3.0E-08
A3 (h) (agonist radioligand)	6	1E-06	-2	102.3	IB-MECA	2.1E-10	1.3E-10
A3 (h) (agonist radioligand)	6	1E-05	39	61.3	IB-MECA	2.1E-10	1.3E-10
alpha 1 (non-selective) (antagonist radioligand)	8	1E-06	-6	105.7	prazosin	4.0E-10	1.1E-10
alpha 1 (non-selective) (antagonist radioligand)	8	1E-05	0	100.3	prazosin	4.0E-10	1.1E-10
alpha 2 (non-selective) (antagonist radioligand)	11	1E-06	-16	116.1	yohimbine	1.1E-07	4.9E-08
alpha 2 (non-selective) (antagonist radioligand)	11	1E-05	6	94.3	yohimbine	1.1E-07	4.9E-08
beta 1 (h) (agonist radioligand)	18	1E-06	6	93.8	atenolol	3.6E-07	2.0E-07
beta 1 (h) (agonist radioligand)	18	1E-05	0	99.5	atenolol	3.6E-07	2.0E-07
beta 2 (h) (antagonist radioligand)	20	1E-06	5	95.1	ICI 118551	1.4E-09	4.8E-10
beta 2 (h) (antagonist radioligand)	20	1E-05	-5	104.7	ICI 118551	1.4E-09	4.8E-10
AT1 (h) (antagonist radioligand)	24	1E-06	-6	105.9	saralasin	1.2E-09	6.1E-10
AT1 (h) (antagonist radioligand)	24	1E-05	3	97.5	saralasin	1.2E-09	6.1E-10
AT2 (h) (agonist radioligand)	26	1E-06	6	93.7	angiotensin-II	5.5E-11	2.7E-11
AT2 (h) (agonist radioligand)	26	1E-05	-3	102.9	angiotensin-II	5.5E-11	2.7E-11
BZD (central) (agonist radioligand)	28	1E-06	-5	105.4	diazepam	5.8E-09	4.9E-09
BZD (central) (agonist radioligand)	28	1E-05	-12	112.4	diazepam	5.8E-09	4.9E-09
B1 (h) (agonist radioligand)	1189	1E-06	0	100.4	desArg10-KD	5.8E-10	1.1E-10
B1 (h) (agonist radioligand)	1189	1E-05	21	79.2	desArg10-KD	5.8E-10	1.1E-10
B2 (h) (agonist radioligand)	33	1E-06	-7	107.5	NPC 567	1.1E-08	5.9E-09
B2 (h) (agonist radioligand)	33	1E-05	-7	107.3	NPC 567	1.1E-08	5.9E-09
CB1 (h) (agonist radioligand)	36	1E-06	8	92.2	CP 55940	1.3E-09	1.2E-09
CB1 (h) (agonist radioligand)	36	1E-05	-8	107.6	CP 55940	1.3E-09	1.2E-09

CB2 (h) (agonist radioligand)	37	1E-06	-5	104.8	WIN 55212-2	2.0E-09	1.3E-09
CB2 (h) (agonist radioligand)	37	1E-05	13	87.2	WIN 55212-2	2.0E-09	1.3E-09
CCK1 (CCKA) (h) (agonist radioligand)	39	1E-06	-10	110.3	CCK-8s	1.4E-10	1.1E-10
CCK1 (CCKA) (h) (agonist radioligand)	39	1E-05	-21	120.7	CCK-8s	1.4E-10	1.1E-10
CCK2 (CCKB) (h) (agonist radioligand)	41	1E-06	-9	108.8	CCK-8s	6.9E-11	2.8E-11
CCK2 (CCKB) (h) (agonist radioligand)	41	1E-05	8	92.2	CCK-8s	6.9E-11	2.8E-11
CRF1 (h) (agonist radioligand)	1467	1E-06	1	99.1	sauvagine	4.0E-10	2.5E-10
CRF1 (h) (agonist radioligand)	1467	1E-05	2	98	sauvagine	4.0E-10	2.5E-10
D1 (h) (antagonist radioligand)	44	1E-06	2	98.2	SCH 23390	4.4E-10	1.8E-10
D1 (h) (antagonist radioligand)	44	1E-05	0	99.6	SCH 23390	4.4E-10	1.8E-10
D2S (h) (antagonist radioligand)	46	1E-06	-4	103.7	(+)butaclamol	2.1E-09	7.0E-10
D2S (h) (antagonist radioligand)	46	1E-05	18	82.1	(+)butaclamol	2.1E-09	7.0E-10
D3 (h) (antagonist radioligand)	48	1E-06	-2	102.1	(+)butaclamol	2.5E-09	5.6E-10
D3 (h) (antagonist radioligand)	48	1E-05	6	93.6	(+)butaclamol	2.5E-09	5.6E-10
D4.4 (h) (antagonist radioligand)	49	1E-06	-12	112.4	clozapine	7.5E-08	2.9E-08
D4.4 (h) (antagonist radioligand)	49	1E-05	-3	102.9	clozapine	7.5E-08	2.9E-08
ETA (h) (agonist radioligand)	54	1E-06	6	93.9	endothelin-1	8.1E-11	4.0E-11
ETA (h) (agonist radioligand)	54	1E-05	3	96.9	endothelin-1	8.1E-11	4.0E-11
ETB (h) (agonist radioligand)	56	1E-06	4	96.3	endothelin-3	3.0E-11	1.7E-11
ETB (h) (agonist radioligand)	56	1E-05	-9	108.9	endothelin-3	3.0E-11	1.7E-11
GABA (non-selective) (agonist radioligand)	57	1E-06	-18	117.7	GABA	2.5E-08	1.5E-08
GABA (non-selective) (agonist radioligand)	57	1E-05	5	95.2	GABA	2.5E-08	1.5E-08
AMPA (agonist radioligand)	64	1E-06	-5	104.6	L-glutamate	3.6E-07	3.3E-07
AMPA (agonist radioligand)	64	1E-05	-4	104.3	L-glutamate	3.6E-07	3.3E-07
kainate (agonist radioligand)	65	1E-06	-1	100.5	kainic acid	1.3E-08	1.0E-08
kainate (agonist radioligand)	65	1E-05	1	99.1	kainic acid	1.3E-08	1.0E-08
NMDA (antagonist radioligand)	66	1E-06	6	94.3	CGS 19755	3.7E-07	3.0E-07
NMDA (antagonist radioligand)	66	1E-05	5	94.8	CGS 19755	3.7E-07	3.0E-07
H1 (h) (antagonist radioligand)	870	1E-06	-14	113.7	pyrilamine	2.2E-09	1.4E-09
H1 (h) (antagonist radioligand)	870	1E-05	-9	108.6	pyrilamine	2.2E-09	1.4E-09
H2 (h) (antagonist radioligand)	1208	1E-06	4	96.3	cimetidine	3.4E-07	3.3E-07
H2 (h) (antagonist radioligand)	1208	1E-05	-11	110.7	cimetidine	3.4E-07	3.3E-07
H3 (h) (agonist radioligand)	1332	1E-06	22	78.4	(R)alpha -Me-histamine	1.5E-09	3.7E-10
H3 (h) (agonist radioligand)	1332	1E-05	-4	103.5	(R)alpha -Me-histamine	1.5E-09	3.7E-10

I2 (antagonist radioligand)	81	1E-06	-16	115.8	idazoxan	1.2E-08	7.7E-09
I2 (antagonist radioligand)	81	1E-05	10	90.1	idazoxan	1.2E-08	7.7E-09
CysLT1 (LTD4) (h) (agonist radioligand)	86	1E-06	-12	112.4	LTD4	6.5E-10	2.9E-10
CysLT1 (LTD4) (h) (agonist radioligand)	86	1E-05	-11	111	LTD4	6.5E-10	2.9E-10
MC4 (h) (agonist radioligand)	420	1E-06	-17	117.5	NDP-alpha -MSH	3.4E-10	3.1E-10
MC4 (h) (agonist radioligand)	420	1E-05	-5	105.5	NDP-alpha -MSH	3.4E-10	3.1E-10
MT1 (ML1A) (h) (agonist radioligand)	1538	1E-06	-6	105.5	melatonin	8.9E-11	7.1E-11
MT1 (ML1A) (h) (agonist radioligand)	1538	1E-05	-1	100.6	melatonin	8.9E-11	7.1E-11
M (non-selective) (antagonist radioligand)	89	1E-06	17	83.2	atropine	1.1E-09	1.8E-10
M (non-selective) (antagonist radioligand)	89	1E-05	2	98.1	atropine	1.1E-09	1.8E-10
NK1 (h) (agonist radioligand)	100	1E-06	4	95.6	[Sar9,Met(O2)11]-SP	9.9E-11	4.4E-11
NK1 (h) (agonist radioligand)	100	1E-05	6	94	[Sar9,Met(O2)11]-SP	9.9E-11	4.4E-11
NK2 (h) (agonist radioligand)	102	1E-06	-15	114.6	[Nleu10]-NKA (4-10)	3.1E-09	1.7E-09
NK2 (h) (agonist radioligand)	102	1E-05	-9	108.8	[Nleu10]-NKA (4-10)	3.1E-09	1.7E-09
NK3 (h) (antagonist radioligand)	104	1E-06	-17	116.6	SB 222200	5.3E-09	2.9E-09
NK3 (h) (antagonist radioligand)	104	1E-05	5	94.6	SB 222200	5.3E-09	2.9E-09
Y (non-selective) (agonist radioligand)	105	1E-06	-19	119.2	NPY	2.4E-10	1.6E-10
Y (non-selective) (agonist radioligand)	105	1E-05	-4	103.9	NPY	2.4E-10	1.6E-10
N neuronal alpha 4beta 2 (h) (agonist radioligand)	3029	1E-06	0	99.7	nicotine	3.4E-09	1.1E-09
N neuronal alpha 4beta 2 (h) (agonist radioligand)	3029	1E-05	-9	109.5	nicotine	3.4E-09	1.1E-09
opioid (non-selective) (antagonist radioligand)	112	1E-06	12	88.2	naloxone	9.0E-10	6.5E-10
opioid (non-selective) (antagonist radioligand)	112	1E-05	-4	104	naloxone	9.0E-10	6.5E-10
NOP (ORL1) (h) (agonist radioligand)	358	1E-06	-1	101.5	nociceptin	2.1E-10	9.8E-11
NOP (ORL1) (h) (agonist radioligand)	358	1E-05	-11	111.3	nociceptin	2.1E-10	9.8E-11
PPARgamma (h) (agonist radioligand)	641	1E-06	-32	132	rosiglitazone	7.7E-09	4.1E-09
PPARgamma (h) (agonist radioligand)	641	1E-05	-5	105.5	rosiglitazone	7.7E-09	4.1E-09
PCP (antagonist radioligand)	124	1E-06	-6	105.6	MK 801	8.5E-09	4.8E-09
PCP (antagonist radioligand)	124	1E-05	-1	101	MK 801	8.5E-09	4.8E-09
EP2 (h) (agonist radioligand)	1955	1E-06	-2	101.9	PGE2	2.8E-09	1.4E-09
EP2 (h) (agonist radioligand)	1955	1E-05	6	94.3	PGE2	2.8E-09	1.4E-09
IP (PGI2) (h) (agonist radioligand)	2230	1E-06	12	88.5	iloprost	1.5E-08	8.6E-09
IP (PGI2) (h) (agonist radioligand)	2230	1E-05	6	94.2	iloprost	1.5E-08	8.6E-09
P2X (agonistradioligand)	127	1E-06	-5	104.7	alpha ,beta - MeATP	8.0E-09	3.7E-09

P2X (agonist radioligand)	127	1E-05	-5	105.5	alpha ,beta - MeATP	8.0E-09	3.7E-09
P2Y (agonist radioligand)	128	1E-06	21	79.3	dATPalpha S	3.1E-08	1.5E-08
P2Y (agonist radioligand)	128	1E-05	9	91.2	dATPalpha S	3.1E-08	1.5E-08
sigma (non-selective) (h) (agonist radioligand)	3500	1E-06	-5	105.2	haloperidol	5.3E-08	4.2E-08
sigma (non-selective) (h) (agonist radioligand)	3500	1E-05	-5	104.9	haloperidol	5.3E-08	4.2E-08
GR (h) (agonist radioligand)	469	1E-06	-18	118.4	dexamethasone	2.6E-09	1.3E-09
GR (h) (agonist radioligand)	469	1E-05	-19	119.2	dexamethasone	2.6E-09	1.3E-09
ER (non-selective) (h) (agonist radioligand)	152	1E-06	4	95.6	17-beta -estradiol	5.2E-10	1.7E-10
ER (non-selective) (h) (agonist radioligand)	152	1E-05	3	97.2	17-beta -estradiol	5.2E-10	1.7E-10
PR (h) (agonist radioligand)	2341	1E-06	7	93.2	promegestone	9.0E-10	7.2E-10
PR (h) (agonist radioligand)	2341	1E-05	-5	104.8	promegestone	9.0E-10	7.2E-10
AR (h) (agonist radioligand)	933	1E-06	0	99.5	testosterone	4.0E-09	1.8E-09
AR (h) (agonist radioligand)	933	1E-05	8	91.6	testosterone	4.0E-09	1.8E-09
TRH1 (h) (agonist radioligand)	1616	1E-06	1	98.9	TRH	3.0E-08	2.0E-08
TRH1 (h) (agonist radioligand)	1616	1E-05	2	97.9	TRH	3.0E-08	2.0E-08
V1a (h) (agonist radioligand)	159	1E-06	-4	104	[d(CH2)51,Tyr(Me)2]-AVP	6.0E-10	3.7E-10
V1a (h) (agonist radioligand)	159	1E-05	13	87.3	[d(CH2)51,Tyr(Me)2]-AVP	6.0E-10	3.7E-10
V2 (h) (agonist radioligand)	497	1E-06	-8	107.9	AVP	1.7E-09	1.2E-09
V2 (h) (agonist radioligand)	497	1E-05	8	92	AVP	1.7E-09	1.2E-09
Ca2+ channel (L, dihydropyridine site) (antagonist radioligand)	161	1E-06	0	100.2	nitrendipine	1.2E-10	7.8E-11
Ca2+ channel (L, dihydropyridine site) (antagonist radioligand)	161	1E-05	-9	108.7	nitrendipine	1.2E-10	7.8E-11
Ca2+ channel (L, diltiazem site) (benzothiazepines) (antagonist radioligand)	162	1E-06	5	94.7	diltiazem	7.1E-08	5.5E-08
Ca2+ channel (L, diltiazem site) (benzothiazepines) (antagonist radioligand)	162	1E-05	-19	119.4	diltiazem	7.1E-08	5.5E-08
Ca2+ channel (L, verapamil site) (phenylalkylamine) (antagonist radioligand)	163	1E-06	3	97	D 600	2.8E-08	1.4E-08
Ca2+ channel (L, verapamil site) (phenylalkylamine) (antagonist radioligand)	163	1E-05	-18	117.6	D 600	2.8E-08	1.4E-08
KATP channel (antagonist radioligand)	165	1E-06	-19	119.1	glibenclamide	2.0E-10	6.5E-11
KATP channel (antagonist radioligand)	165	1E-05	-23	123.5	glibenclamide	2.0E-10	6.5E-11
KV channel (antagonist radioligand)	166	1E-06	-5	104.9	alpha -dendrotoxin	1.2E-10	9.8E-11
KV channel (antagonist radioligand)	166	1E-05	0	99.7	alpha -dendrotoxin	1.2E-10	9.8E-11

SKCa channel (antagonist radioligand)	167	1E-06	12	88.3	apamin	2.5E-11	1.2E-11
SKCa channel (antagonist radioligand)	167	1E-05	-10	110.2	apamin	2.5E-11	1.2E-11
Na+ channel (site 2) (antagonist radioligand)	169	1E-06	-3	103.4	veratridine	4.0E-06	3.6E-06
Na+ channel (site 2) (antagonist radioligand)	169	1E-05	-14	114.3	veratridine	4.0E-06	3.6E-06
Cl- channel (GABA-gated) (antagonist radioligand)	170	1E-06	2	98.3	picrotoxinin	2.9E-07	2.4E-07
Cl- channel (GABA-gated) (antagonist radioligand)	170	1E-05	6	93.8	picrotoxinin	2.9E-07	2.4E-07
norepinephrine transporter (h) (antagonist radioligand)	355	1E-06	-15	114.5	protriptyline	5.6E-09	4.1E-09
norepinephrine transporter (h) (antagonist radioligand)	355	1E-05	-5	105.2	protriptyline	5.6E-09	4.1E-09
dopamine transporter (h) (antagonist radioligand)	52	1E-06	16	83.8	BTCP	1.2E-08	6.4E-09
dopamine transporter (h) (antagonist radioligand)	52	1E-05	-8	107.7	BTCP	1.2E-08	6.4E-09
GABA transporter (antagonist radioligand)	60	1E-06	-8	108.1	nipecotic acid	1.9E-06	1.8E-06
GABA transporter (antagonist radioligand)	60	1E-05	-8	107.6	nipecotic acid	1.9E-06	1.8E-06
choline transporter (CHT1) (h) (antagonist radioligand)	1552	1E-06	8	91.5	hemicholinium-3	1.0E-08	5.8E-09
choline transporter (CHT1) (h) (antagonist radioligand)	1552	1E-05	11	89.1	hemicholinium-3	1.0E-08	5.8E-09
5-HT transporter (h) (antagonist radioligand)	439	1E-06	-5	104.6	imipramine	3.5E-09	1.6E-09
5-HT transporter (h) (antagonist radioligand)	439	1E-05	-8	108.2	imipramine	3.5E-09	1.6E-09
Serotonin (5-Hydroxytryptamine) 5-HT1, Non-Selective	271010	1E-06	-3	103.2	Serotonin (5-HT)	3.2E-09	1.2E-09
Serotonin (5-Hydroxytryptamine) 5-HT1, Non-Selective	271010	1E-05	3	96.6	Serotonin (5-HT)	3.2E-09	1.2E-09

(Bi). 28 Enzyme & Cell-Based Assay (Eurofin Cerep Diversity Panel)	Catalog Ref	Test Concentration (M)	% Inhibition of Control Values	% of Control Values	Reference Compound	IC_{50} Ref (M)	nH Ref
COX1(h)	4173	1E-06	-11	110.9	Diclofenac	8.5E-09	1.3
COX1(h)	4173	1E-05	-31	130.5	Diclofenac	8.5E-09	1.3
5-lipoxygenase (h)	772	1E-06	18	81.5	NDGA	3.8E-07	2.2
5-lipoxygenase (h)	772	1E-05	16	84.4	NDGA	3.8E-07	2.2
PDE1B (h)	4070	1E-06	3	97	Nitrendipine	8.1E-06	1.2
PDE1B (h)	4070	1E-05	-6	106.3	Nitrendipine	8.1E-06	1.2
PDE2A1 (h)	4071	1E-06	-8	107.9	BAY60-7550	1.6E-09	0.9
PDE2A1 (h)	4071	1E-05	-9	109.2	BAY60-7550	1.6E-09	0.9
PDE3A (h)	4072	1E-06	3	97.4	milrinone	4.5E-07	1
PDE3A (h)	4072	1E-05	11	89.4	milrinone	4.5E-07	1
PDE4D2 (h)	4077	1E-06	7	92.8	Ro 20-1724	1.6E-07	0.7
PDE4D2 (h)	4077	1E-05	20	79.8	Ro 20-1724	1.6E-07	0.7
PDE5 (h) (non-selective)	204	1E-06	6	94.5	dipyridamole	8.4E-07	0.5

PDE5 (h) (non-selective)	204	1E-05	22	78.1	dipyridamole	8.4E-07	0.5
phosphatase 1B (h) (PTP1B)	2593	1E-06	3	96.6	Ammonium molybdate	3.0E-07	1.9
phosphatase 1B (h) (PTP1B)	2593	1E-05	4	96	Ammonium molybdate	3.0E-07	1.9
phosphatase CDC25A (h)	4570	1.00E-06	9	90.9	NSC 663284	2.3E-07	1.8
phosphatase CDC25A (h)	4570	1.00E-05	1	99.1	NSC 663284	2.3E-07	1.8
phosphatase CDC25B (h)	4571	1.00E-06	5	95.2	NSC 663284	5.4E-07	2.3
phosphatase CDC25B (h)	4571	1.00E-05	10	89.5	NSC 663284	5.4E-07	2.3
PKCalpha (h)	348	1E-06	-23	123.1	Bis 10	2.0E-09	1
PKCalpha (h)	348	1E-05	-8	107.6	Bis 10	2.0E-09	1
acetylcholinesterase (h)	363	1E-06	0	99.7	galanthamine	7.9E-07	0.9
acetylcholinesterase (h)	363	1E-05	-1	100.8	galanthamine	7.9E-07	0.9
COMT (catechol- O-methyl transferase)	457	1E-06	-19	119.3	Ro 41-0960	4.2E-08	2
COMT (catechol- O-methyl transferase)	457	1E-05	-4	104.1	Ro 41-0960	4.2E-08	2
GABA transaminase	461	1E-06	26	73.5	O-(Carboxyméthyl) hydroxylamine hémihydrochloride	1.2E-07	1.4
GABA transaminase	461	1E-05	-8	108.4	O-(Carboxyméthyl) hydroxylamine hémihydrochloride	1.2E-07	1.4
MAO-A (h)	191	1E-06	5	94.5	clorgyline	2.4E-08	1.4
MAO-A (h)	191	1E-05	-6	106.1	clorgyline	2.4E-08	1.4
MAO-B (h) recombinant enzyme	3477	1E-06	-3	102.5	deprenyl	8.6E-08	1.6
MAO-B (h) recombinant enzyme	3477	1E-05	-1	101	deprenyl	8.6E-08	1.6
tyrosine hydroxylase	214	1E-06	5	95	3-iodo L-tyrosine	6.7E-07	1.2
tyrosine hydroxylase	214	1E-05	-3	102.8	3-iodo L-tyrosine	6.7E-07	1.2
ATPase (Na+/K+)	2009	1E-06	-6	106.2	ouabain	2.6E-07	1.3
ATPase (Na+/K+)	2009	1E-05	-3	103.4	ouabain	2.6E-07	1.3
CENP-E (h)	2150	1E-06	5	95	rose bengal	3.1E-07	1.7
CENP-E (h)	2150	1E-05	3	96.7	rose bengal	3.1E-07	1.7
Eg5 (h)	2151	1E-06	2	97.7	S-trityl-L-cysteine	2.9E-07	1.2
Eg5 (h)	2151	1E-05	13	87.2	S-trityl-L-cysteine	2.9E-07	1.2
HDAC3 (h)	2083	1E-06	-9	109.5	trichostatin A	3.6E-09	0.8
HDAC3 (h)	2083	1E-05	21	78.6	trichostatin A	3.6E-09	0.8
HDAC4 (h)	2493	1E-06	1	99.1	trichostatin A	3.0E-06	1
HDAC4 (h)	2493	1E-05	0	99.9	trichostatin A	3.0E-06	1
HDAC6 (h)	2495	1E-06	-2	101.8	trichostatin A	9.0E-09	1.5
HDAC6 (h)	2495	1E-05	-2	102	trichostatin A	9.0E-09	1.5
HDAC11 (h)	2663	1E-06	20	80.3	scriptaid	3.8E-06	0.7
HDAC11 (h)	2663	1E-05	36	64.1	scriptaid	3.8E-06	0.7
sirtuin 1 (h) (inhibitor effect)	2581	1E-06	-2	102	suramin	8.5E-06	1.4
sirtuin 1 (h) (inhibitor effect)	2581	1E-05	-2	102.3	suramin	8.5E-06	1.4
sirtuin 2 (h) (inhibitor effect)	2582	1E-06	-1	101.4	suramin	5.7E-05	6.2
sirtuin 2 (h) (inhibitor effect)	2582	1E-05	-5	104.9	suramin	5.7E-05	6.2

(Bii). 28 Enzyme & Cell-Based Assay (Eurofin Cerep Diversity Panel, continued)	Catalog Ref	Test Concen-tration (M)	% Stimulat-ion Relative to Control	% Stimulat-ion Relative to Control	Reference Compound	EC_{50} Ref (M)	nH Ref
adenylyl cyclase (activator effect)	3002	1E-06	0	-0.1	forskolin	1.3E-05	1.2
adenylyl cyclase (activator effect)	3002	1E-05	0	0.4	forskolin	1.3E-05	1.2
guanylyl cyclase (h) (activator effect)	3004	1E-06	0	-0.3	sodium nitroprusside	1.1E-05	1
guanylyl cyclase (h) (activator effect)	3004	1E-05	5	5	sodium nitroprusside	1.1E-05	1

(Ci). Custom-selected extra Cerep Binding Assays	Catalog Ref	Test Concen-tration	% Inhibition of Control Specific Binding	1st / % of Control Specific Binding	Mean / % of Control Specific Binding	Reference Compound	IC_{50} Ref (M)	Ki Ref	nH Ref
M1 (h) (antagonist radioligand)	91	1E-05	1	99.2	99.2	pirenzepine	3.3E-08	2.9E-08	1.4
mu (MOP) (h) (agonist radioligand)	118	1E-05	6	93.7	93.7	DAMGO	9.1E-10	3.7E-10	1
5-HT2B (h) (agonist radioligand)	1333	1E-05	20	79.8	79.8	(±)DOI	6.8E-09	3.4E-09	1.1

(Cii). Custom-selected extra Cerep Enzyme and Cell-Based Assays (continued)	Catalog Ref	Test Concen-tration	% Inhibition of Control Values	1st / % of Control Values	Mean / % of Control Values	Reference Compound	IC_{50} Ref	nH Ref
COX2(h)	4186	1E-05	39	60.7	60.7	NS398	2.4E-07	1.2

(D) Percentage activity of HDACs after treatment with 1 μM and 10 μM WM-1119 tested in triplicate by Reaction Biology											
	HDAC1	HDAC2	HDAC3	HDAC4	HDAC5	HDAC6	HDAC7	HDAC8	HDAC9	HDAC10	HDAC11
WM-1119 10 μM	94.00 96.32 94.89	113.84 109.40 107.07	96.24 99.66 97.22	98.17 98.09 99.62	146.24 152.06 153.64	101.72 105.23 101.91	88.51 83.45 87.65	95.18 97.11 99.02	110.85 108.40 108.28	84.33 83.97 85.68	101.55 100.21 97.87
WM-1119 1 μM	100.17 99.80 99.37	112.68 110.81 115.08	91.87 90.23 92.28	95.53 94.61 94.19	117.78 119.76 118.15	99.91 99.82 99.04	89.82 89.40 89.25	98.06 97.84 97.36	105.64 96.89 96.31	82.58 86.04 83.26	94.88 96.57 95.14
Control IC_{50} (M): Trichostatin A	1.38E-08	5.60E-08	3.17E-08			3.16E-09		8.13E-07		4.48E-08	5.28E-06
Control IC_{50} (M): TMP269				1.94E-07	1.53E-07		5.70E-08		1.29E-08		

(E) Percentage activity of protein tyrosine phosphatases after treatment with 1 μM and 10 μM WM-1119 tested in triplicate by Reaction Biology, % Enzyme Activity (relative to DMSO controls)							
	PTP1B	PTPN2/ TC-PTP	PTPN12/ PTP-PEST	PTPN7/ LC-PTP	PTPRC/ CD45	PTPN6/ SHP1	PTPN11/ SHP2
WM-1119 10 μM	106 105 103	95 99 100	102 104 97	81 86 86	108 111 110	107 101 111	117 116 115
WM-1119 1 μM	102 100 95	100 99 94	100 96 97	94 77 77	105 105 104	113 109 103	114 117 113
Control PTP1B Inhibitor	8.6E-06	5.0E-06	3.1E-06	3.1E-06	3.9E-06	1.7E-06	1.8E-05

(F) Percentage activity of 23 diverse kinases after treatment with 1 μM and 10 μM WM-1119 tested in triplicate by Reaction Biology with [ATP] Km shown (μM)								
	BMX/ETK [50]	BTK [50]	c-Kit [100]	CK1g1 [20]	CK1g2 [30]	CK1g3 [20]	EGFR [15]	ERK2 / MAPK1 [50]
WM-1119 10 μM	110 110 110	103 102 100	102 96 98	97 101 108	105 108 106	97 101 96	104 104 105	98 99 100
WM-1119 1 μM	104 105 103	102 103 99	95 94 97	97 96 99	106 108 99	101 107 106	104 104 106	107 101 100
Staurosporine control IC_{50} (M)	6.2E-09	2.8E-08	5.6E-08	1.8E-05	7.8E-06	4.2E-06	5.4E-08	3.3E-08
	FGFR1 [5]	FGFR2 [5]	FGFR3 [30]	GSK3b [10]	IKKb / IKBKB [30]	ITK [30]	JAK3 [2.5]	JNK1 [15]
WM-1119 10 μM	105 107 109	98 108 103	109 107 110	103 105 110	108 111 111	104 104 111	108 110 108	95 102 99
WM-1119 1 μM	107 103 103	113 101 98	109 110 101	107 110 107	112 105 103	107 106 104	110 107 109	104 109 103
Staurosporine control IC_{50} (nM)	5.7E-09	2.3E-09	1.8E-08	6.8E-09	7.2E-07	1.7E-08	7.4E-11	7.9E-07
	JNK2 [20]	JNK3 [30]	KDR / VEGFR2 [20]	MEK1 [2.5]	NEK2 [20]	TAK1 [20]	TXK [30]	
WM-1119 10 μM	104 100 103	106 104 104	102 99 100	99 98 104	88 76 86	99 96 101	97 100 94	
WM-1119 1 μM	104 105 99	100 102 97	103 106 97	103 100 101	93 130 109	101 95 99	101 101 98	
Staurosporine control IC_{50} (nM) or JNKI VIII*	2.8E-06	6.2E-07	2.2E-08	1.2E-08	4.1E-07	3.7E-08	2.1E-08	

(G) Percentage activity of cathepsins and caspases after treatment with 1 μM and 10 μM WM-1119 tested in triplicate by Reaction Biology								
	Caspase 1	Caspase 2	Caspase 3	Caspase 4	Caspase 5	Caspase 6	Caspase 7	Caspase 8
WM-1119 10 μM	98 95 94	104 104 107	109 105 103	94 91 90	103 105 96	101 100 97	104 102 99	94 95 97
WM-1119 1 μM	92 96 96	103 101 99	105 105 98	95 94 92	110 112 109	104 103 99	102 102 98	105 106 102
Control Compound IC50 (M)	7.7E-08	1.1E-06	1.5E-09	2.2E-06	4.8E-08	2.2E-08	2.9E-09	3.3E-09
Control compound ID	IETD-CHO	IETD-CHO	DEVD-CHO	IETD-CHO	IETD-CHO	DEVD-CHO	DEVD-CHO	IETD-CHO
	Caspase 9	Caspase 10	Caspase 11	Caspase 14	Cathepsin B	Cathepsin C	Cathepsin H	Cathepsin L
WM-1119 10 μM	94 96 92	98 100 99	94 91 93	108 109 106	112 110 109	98 97 96	107 101 105	92 80 94
WM-1119 1 μM	103 104 97	106 107 103	100 94 91	110 113 105	109 110 105	96 96 93	106 107 107	104 108 107
Control Compound IC50 (M)	8.8E-08	1.5E-08	8.5E-07	1.8E-08	1.2E-08	6.2E-07	2.6E-08	4.0E-09
Control compound ID	IETD-CHO	IETD-CHO	IETD-CHO	WEHD-CHO	E64	E64	E64	E64

	Cathepsin S	Cathepsin V	Papain					
WM-1119 10 µM	96 100 102	98 94 90	91 98 96					
WM-1119 1 µM	98 96 95	96 96 88	104 104 99					
Control Compound IC50 (M)	2.3E-09	3.8E-09	7.8E-10					
Control compound ID	E64	E64	E64					

(H) Percentage activity of SIRTS after treatment with 1 µM and 10 μM WM-1119 tested in triplicate by Reaction Biology				
	SIRT 1	SIRT 2	SIRT 3	SIRT 5
WM-1119 10 µM	99 103 99	107 107 106	114 114 112	98 97 109
WM-1119 1 µM	99 103 101	105 104 105	115 118 112	136 110 110
IC_{50} (M): Surami	2.1E-06	1.6E-05		
IC_{50} (M): Nicotinimide			3.5E-05	2.2E-05

[a.] Eurofin Cerep 71 Receptor and 27 Enzyme Diversity Panel. The binding assay results are expressed as a percentage of control specific binding. The results of enzyme and uptake assays are expressed as a percentage of control specific activity.
[b.] Compounds were tested by Reaction Biology either in single does triplicate mode at 1 µM and 10 µM according to Reaction Biology protocols.

Supplementary Table 7: Oligonucleotide primers used in quantitative PCRs following oligo(dT) primed reverse transcriptase reactions.

Gene	Primer Sequence
Arf	F 5' GCCGCACCGGAATCCT 3' R 5' TTGAGCAGAAGAGCTGCTACGT 3'
Cdc6	F 5' AGGAGCCAGACAGTCCTCAA 3' R 5' GGGTCAAAAGCAGCAAAGAG 3'
E2f2	F 5' GATGGAGTCCTGGACCTGAA 3' R 5' CTGCCTACCCACTGGATGTT 3'
Ezh2	F 5' ATCTGAGAAGGGACCGGTTT 3' R 5' TCAGGGTCTTTAACGGGATG 3'
Gapdh	F 5' TTCACCACCATGGAGAAGGC 3' R 5' CCCTTTTGGCTCCACCCT 3'
Hsp90ab1	F 5' ACCTGGGAACCATTGCTAAG 3' R 5' AGAATCCGACACCAAACTGC 3'
Ink4a	F 5' CGTACCCCGATTCAGGTGAT 3' R 5' TTGAGCAGAAGAGCTGCTACGT 3'
Kat6a	F 5' CTTACACGGATGCCAAAAGG 3' R 5' GTTTTATCTGTGCCGCCTTC 3'
Kat6b	F 5' GTGCTTTTCCGTCCTCACTCC R 5' CACGATTTGACTCTTTAGTCCCC
Kat7	F 5' TTTTGGCCGCTATGAACTG 3' R 5' GGAGGATTGTCTGGCTCTTC 3'
Melk	F 5' TGGCTCTCTCCCAGTAGCAT 3' R 5' GAGTCTTGCTTTGCCACTCC 3'
Cdkn1a	F 5' GTGGGTCTGACTCCAGCCC 3' R 5' CCTTCTCGTGAGACGCTTAC 3'
Skp2	F 5' GCGCTAAAACAGGAGTCTGG 3' R 5' CCTGAAGGTGCTTCCTATGC 3'
drCdkn2a (Ink4a)	F 5' CGAGGATGAACTGACCACAGC 3' R 5' CAACAGCCAAAGGTGCGTTAC 3'
drCdkn1a	F 5' CAAGCCAAGAAGCGTCTAGTG 3' R 5' AACGGTGTCGTCTCTGGTTC 3'

Supplementary Table 8: Oligonucleotide primers used in quantitative PCRs following chromatin immunoprecipitation.

Gene	Primer Sequence
Melk	F 5' GCTGCTGGAACTTGAATCCT 3' R 5' AGTCTAGCAAAGCCGGAACA 3'
E2f2	F 5' ACGGGAACTAGAGGGGTGAA 3' R 5' GGACACTCGTGTGCTCTGAC 3'
Ezh2	F 5' AGAGGCGCTTGATAGTGCTG 3' R 5' GACTCCACTGCCTTCGATGT 3'

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