

Synthesis and evaluation of 3-amino/guanidine substituted phenyl oxazoles as a novel class of LSD1 inhibitors with anti-proliferative properties

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1. Experimental Section:

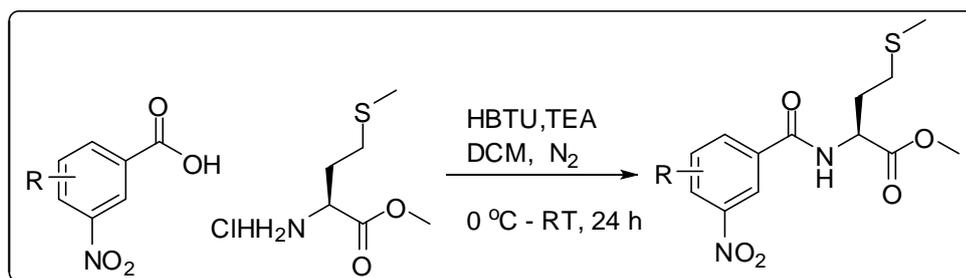
1.1 Chemistry

General methods

All reactions were carried out under an inert atmosphere with dry solvents, unless otherwise stated. Reactions were monitored by thin layer chromatography (TLC) on silica gel plates (60 F254), using UV light detection. Visualization of the spots on TLC plates was achieved either by UV light or by staining the plates in 2, 4 Di-Nitro Phenylhydrazine stain, Ninhydrin stain, KMNO₄ stain and charring on hot plate. Flash chromatography was performed on silica gel (230-400 mesh) using distilled hexane, ethyl acetate, dichloromethane. ¹H NMR and ¹³C NMR spectra were recorded in CDCl₃ or acetone-*d*₆ solution by using (Wormhole-vnmrs) 400 MHz spectrometers. Deuterium exchange studies were done in CD₃OD, D₂O. ¹H NMR and ¹³C NMR were recorded in CDCl₃ and CD₃OD solvents on 400 MHz spectrometer, respectively at ambient temperature. Chemical shifts are reported as δ values relative to internal CHCl₃ δ 7.26 or TMS δ 0.0 and CD₃OD δ 3.34 for ¹H NMR and CHCl₃ δ 77.0 and CD₃OD δ 49.05 for ¹³C NMR. ¹H NMR data is recorded as follows: chemical shift [multiplicity, coupling constant(s) J (Hz), relative integral] where multiplicity is defined as: s (singlet), d (doublet), t (triplet), q (quartet), dd (doublet of doublet), m (multiplet), bs (broad singlet). FTIR spectra were recorded on Bruker (Alpha) spectrometer. Mass spectra were recorded on Micro Mass VG-7070H mass spectrometer for ESI and are given in mass units (m/z). High resolution mass spectra (HRMS) [ESI+] were obtained using either a TOF or a double focusing spectrometer.

The compounds **1a**, **1b**, **1c** were commercially available. The methionine ester (**1d**) was synthesized from methionine and **7** was prepared from thiourea using literature protocols in quantitative yield.

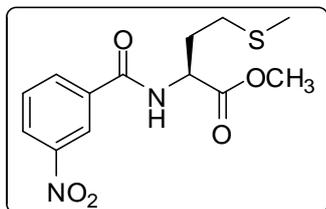
General procedure for synthesis of 2a-c:



HBTU (1.5 mmol) was added to a mixture of benzoic acid (**1a**, **1b**, **1c**) (1 mmol), methionine methylester hydrochloride (1.2 mmol) in dry DCM (10 mL) at 0 °C. The reaction mixture was stirred for 1 h to which dry triethylamine (10.0 mmol) was added drop wise at 0 °C. Stirring was continued at room temperature for another 12 h. The reaction mixture was cooled in an ice bath and acidified with 2N HCl. The residue was dissolved in ethyl acetate (750 mL), washed with water (2 x 250 mL), brine (1 x 100 mL), dried over Na₂SO₄, filtered and the solvent evaporated.

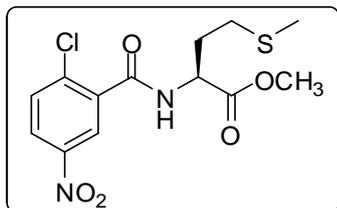
The residue was purified by flash chromatography (n-hexane/ ethylacetate 4:1) to afford the desired products (**2a**, **2b**, **2c**).

(S)-Methyl 4-(methylthio)-2-(3-nitrobenzamido) butanoate (2a):



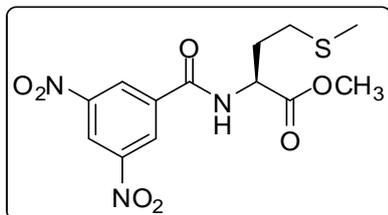
White semi solid; yield (87 %); $R_f = 0.60$ (30% EtOAc-n-Hexane); IR (cm^{-1}): 3325, 3056, 2956, 2876, 1735 (C=O of ester), 1690 (C=O of amide), 1540, 1340; ^1H NMR (300 MHz, CDCl_3) δ 2.13 (s, 3H), 2.16 -2.22 (m, 1H), 2.27 -2.34 (m, 1H), 2.62 (t, $J = 7.2$ Hz, 1H), 3.81 (s, 3H), 4.92- 4.97 (m, 1H), 7.33 (d, $J = 7.6$ Hz, 1H), 7.66 (d, $J = 8.0$ Hz, 1H), 8.17- 8.22 (dd, $J^1 = 7.4$ Hz, $J^2 = 1.6$ Hz, 1H), 8.36- 8.39 (dd, $J^1 = 7.4$ Hz, $J^2 = 1.6$ Hz, 1H), 8.67 (s, 1H); ^{13}C NMR (75 MHz, CDCl_3) δ 15.5, 30.2, 31.1, 52.4, 52.8, 122.1, 126.3, 126.3, 129.8, 133.2, 135.2, 148.2, 164.7 ; MS (EI, 70ev): m/z (%) $[\text{M}]^-$ 311.1 (100%).

(S)-Methyl 2-(2-chloro-5-nitrobenzamido)-4-(methylthio) butanoate (2b):



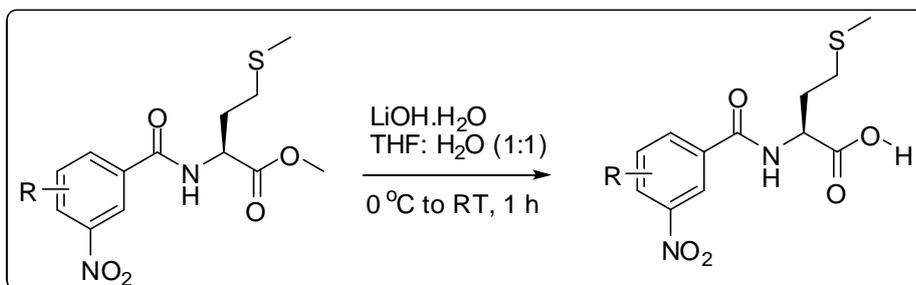
The title compound was prepared in 80% yield according to the general procedure as described above; Brown solid; $R_f = 0.55$ (30% EtOAc-n-Hexane); mp 265-267 °C; IR (cm^{-1}): 3295, 3042, 2955, 2865, 1728 (C=O of ester), 1678 (C=O of amide), 1520, 1334; ^1H NMR (300 MHz, CDCl_3) δ 2.13-2.2 (m, 4H), 2.31-2.38 (m, 1H), 2.56-2.67 (m, 2H), 3.82 (s, 3H), 4.93- 4.98 (m, 1H), 7.02 (d, $J = 7.2$ Hz, 1H), 7.62 (d, $J = 8.8$ Hz, 1H), 8.22-8.25 (dd, $J^1 = 8.8$ Hz, $J^2 = 2.6$ Hz, 1H), 8.53 (d, $J = 3.2$ Hz, 1H); ^{13}C NMR (75 MHz, CDCl_3) δ 15.5, 29.9, 31.3, 52.3, 52.8, 125.2, 125.8, 131.4, 135.7, 137.6, 146.4, 163.8, 171.6; MS (EI, 70ev): m/z (%) $[\text{M}]^+$ 347.1 (100%).

(S)-Methyl 2-(3, 5-dinitrobenzamido)-4-(methylthio)butanoate (2c):



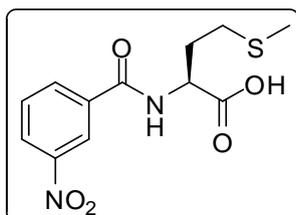
The title compound was prepared in 82% yield according to the general procedure as described above and isolated as yellow solid; $R_f = 0.7$ (30% EtOAc-n-Hexane); mp 285-288 °C; IR (cm^{-1}): 3310, 3010, 2975, 2855, 1732 (C=O of ester), 1660 (C=O of amide), 1515, 1324; ^1H NMR (300 MHz, CDCl_3) δ 2.16 (s, 3H), 2.17- 2.37 (m, 2H), 2.6-2.68 (m, 2H), 3.84 (s, 3H), 4.99 (td, $J^1 = 7.2$ Hz, $J^2 = 4.8$ Hz, 1H), 7.56 (d, $J = 7.2$ Hz, 1H), 9.00(d, $J = 2.0$ Hz, 1H), 9.18 (d, $J = 2.0$ Hz, 1H). ^{13}C NMR (75MHz, CDCl_3) δ 15.5, 30.2, 30.7, 38.6, 52.8, 52.9, 121.3, 127.3, 137.0, 148.6, 162.4, 172.2; MS (EI, 70ev): m/z (%) $[\text{M}]^+$ 358.2 (100%).

General procedure for synthesis of 3a-c:



LiOH.H₂O (1.5 mmol) was added to the solution of (*S*)-methyl 4-(methylthio)-2-(3-nitrobenzamido) butanoate **2a** (1.0 mmol) in THF (6 mL), H₂O (5 mL) at 0 °C. The reaction was stirred at 23 °C for 1 h. After completion of the reaction, the reaction mixture was concentrated to remove THF. The solution was cooled in an ice bath, acidified with 2N HCl, and extracted with EtOAc. The organic layer was collected, washed with brine, dried over anhydrous Na₂SO₄, filtered and concentrated. The residue was purified by flash chromatography (n-hexane/ ethylacetate 3:2) to afford the desired product **3a**.

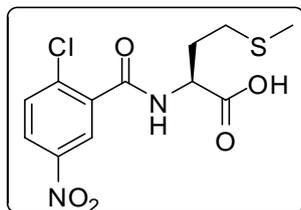
(*S*)-4-(Methylthio)-2-(3-nitrobenzamido) butanoic acid (**3a**):



White solid; yield (88 %); $R_f = 0.4$ (30% EtOAc-n-Hexane); mp 276-278 °C; IR (cm^{-1}): 3302, 2989, 2866, 1712 (C=O of acid), 1650 (C=O of amide), 1525, 1314; ^1H NMR (300 MHz, CDCl_3) δ 2.12 (s, 3H), 2.15 -2.34 (m, 2H), 2.65 (t, $J = 7.2$ Hz, 1H), 4.92- 4.97 (m, 1H), 7.28 (bs, 1H), 7.61-7.69 (m,

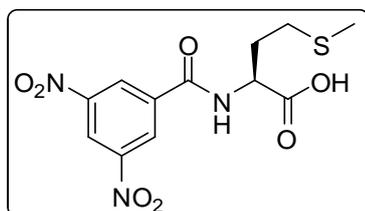
2H), 8.15 (d, $J = 7.8$ Hz, 1H), 8.33 (d, $J = 7.8$ Hz, 1H), 8.65 (s, 1H); ^{13}C NMR (75 MHz, CDCl_3) δ 15.4, 30.2, 30.6, 52.5, 122.2, 126.5, 129.9, 133.4, 134.9, 148.1, 165.6, 175.3; MS (EI, 70eV): m/z (%) $[\text{M}]^-$ 297.0 (100%)

(S)-2-(2-Chloro-5-nitrobenzamido)-4-(methylthio)butanoic acid (3b):



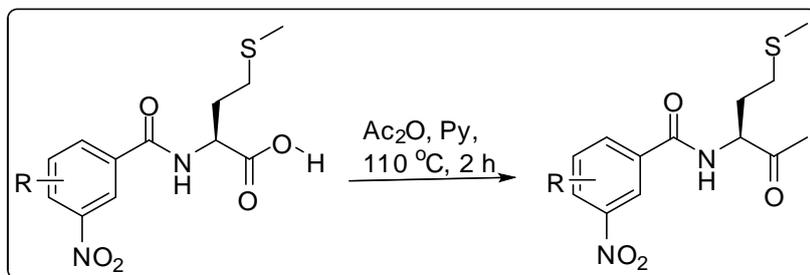
The title compound was synthesized in 94% yield according to the general procedure as described above for **3a** and isolated as brown solid; $R_f = 0.42$ (30% EtOAc-n-Hexane); mp 231-235 °C; IR (cm^{-1}): 3299, 3050, 2979, 2856, 1722 (C=O of acid), 1650 (C=O of amide), 1515, 1324; ^1H NMR (300 MHz, CDCl_3) δ 2.14 (s, 3H), 2.16-2.4 (m, 2H), 2.65-2.69 (m, 4H), 4.13-5.11 (m, 3H), 7.10 (d, $J = 7.6$ Hz, 1H), 7.62 (d, $J = 8.4$ Hz, 1H), 8.22-8.25 (dd, $J^1 = 8.4$ Hz, $J^2 = 2.8$ Hz, 1H), 8.53 (d, $J = 2.4$ Hz, 1H); ^{13}C NMR (75 MHz, CDCl_3) δ 15.5, 29.7, 30.1, 30.9, 52.4, 125.3, 126.0, 131.5, 135.4, 137.7, 146.5, 164.4, 175.2 MS (EI, 70eV): m/z (%) $[\text{M}]^+$ 333.1 (100%).

(S)-2-(3,5-Dinitrobenzamido)-4-(methylthio)butanoic acid (3c):



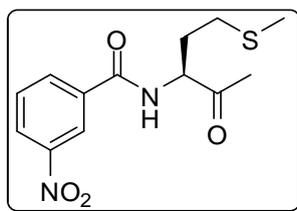
The title compound was synthesized in 90% yield according to the general procedure as described for **3a** as light brown solid; $R_f = 0.42$ (30% EtOAc-n-Hexane); mp 268-270 °C; IR (cm^{-1}): 3301, 3060, 2985, 2862, 1718 (C=O of acid), 1660 (C=O of amide), 1525, 1334; ^1H NMR (300 MHz, $\text{DMSO}-d_6$) δ 2.12 (s, 3H), 2.13- 2.33 (m, 2H), 2.57-2.70 (m, 2H), 4.80-4.86 (m, 2H), 9.11(d, $J = 2.0$ Hz, 1H), 9.21 (d, $J = 7.6$ Hz, 1H), 9.31(d, $J = 2.0$ Hz, 2H). ^{13}C NMR (75MHz, $\text{DMSO}-d_6$) δ 15.3, 30.4, 30.9, 52.6, 120.8, 127.8, 137.4, 148.3, 162.6, 173.3; MS (EI, 70eV): m/z (%) $[\text{M}]^+$ 344.1 (100%).

General procedure for synthesis of 4a-c:



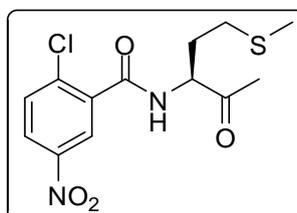
To a solution of (*S*)-4-(methylthio)-2-(3-nitrobenzamido)butanoic acid **3a** (4.9 mmol) in acetic anhydride (30.4 mmol, 3 mL) was added pyridine (58.8 mmol, 5 mL) and the mixture was heated at 90 °C for 2 h. After completion the reaction mixture was concentrated to remove the excess of acidic anhydride and pyridine and the residue was extracted with EtOAc (150 mL). The EtOAc layer was washed successively with 1.0 N HCl (50 mL), water (50 mL) and brine, dried over anhydrous Na₂SO₄, filtered and concentrated. The residue was purified by column chromatography (EtOAc/hexanes) to provide of the desired product **4a**.

(S)-N-(1-(Methyl thio)-4-oxopentan-3-yl)-3-nitrobenzamide (4a):



Light brown semi solid; $R_f = 0.6$ (30% EtOAc-n-Hexane); IR (cm⁻¹): 3100, 3060, 2985, 2862, 1720 (C=O of ketone), 1665 (C=O of amide), 1515, 1314; ¹H NMR (300 MHz, CDCl₃) δ 2.13 (s, 3H), 2.14 -2.17 (m, 2H), 2.34 (s, 3H), 2.41 -2.64 (m, 2H), 4.96 - 4.99 (m, 1H), 7.35 (d, $J = 6.4$ Hz, 1H), 7.67 (t, $J = 7.8$ Hz, 1H), 8.17 (d, $J = 7.8$ Hz, 1H), 8.37- 8.39 (dd, $J^1 = 8.0$ Hz, $J^2 = 1.6$ Hz, 1H), 8.67 (s, 1H); ¹³C NMR (75 MHz, CDCl₃) δ 15.7, 27.3, 30.1, 30.5, 58.9, 122.1, 126.4, 129.8, 133.1, 135.4, 148.3, 164.8, 205.7; MS (EI, 70ev): m/z (%) [M]⁺ 295.0 (100%).

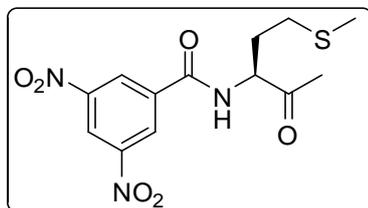
(S)-2-Chloro-N-(1-(methylthio)-4-oxopentan-3-yl)-5-nitrobenzamide (4b):



The title compound was synthesized as a yellow semi solid in 78% yield according to the general procedure described above for **4a**; $R_f = 0.65$ (30% EtOAc-n-Hexane); IR (cm⁻¹): 3200, 3050, 2976, 2862, 1710 (C=O of ketone), 1655 (C=O of amide), 1518, 1323; ¹H NMR (300 MHz, CDCl₃): 2.00-2.07 (m, 1H), 2.13 (s, 3H), 2.34 (s, 3H), 2.39- 2.46 (m, 1H), 2.53-2.61 (m, 2H), 4.96-5.00 (m, 1H),

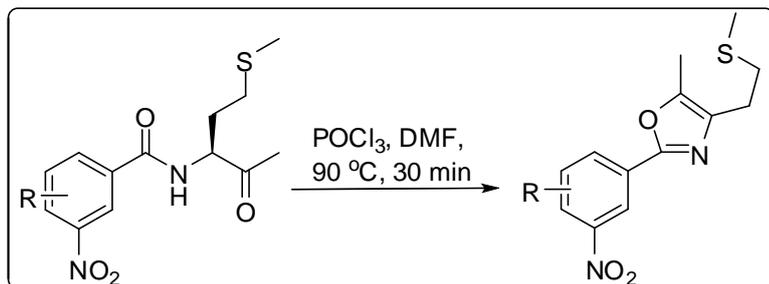
7.16 (d, $J = 4.8$ Hz, 1H), 7.62 (d, $J = 7.2$ Hz, 1H), 8.22-8.25 (dd, $J^1 = 7.2$ Hz, $J^2 = 2.4$ Hz, 1H), 8.51 (d, $J = 2.8$ Hz, 1H); ^{13}C NMR (75 MHz, CDCl_3): 15.5, 25.0, 30.9, 33.3, 126.7, 128.2, 131.7, 133.2, 140.2, 146.9, 156.4, 208.2 ; MS (EI, 70ev): m/z (%) $[\text{M}]^+$ 333.1 (100%).

(S)-N-(1-(Methylthio)-4-oxopentan-3-yl)-3,5-dinitrobenzamide (4c):



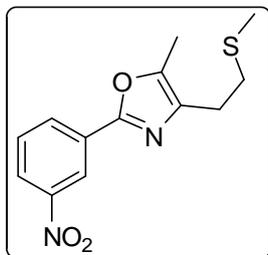
The title compound was synthesized as colorless semi solid in 74% yield according to the general procedure as described above for 4a; $R_f = 0.7$ (30% EtOAc-*n*-Hexane); IR (cm^{-1}): 3233, 3040, 2978, 2872, 1718 (C=O of ketone), 1656 (C=O of amide), 1520, 1321; ^1H NMR (300 MHz, CDCl_3) δ 2.03-2.19 (m, 5H), 2.25-2.36 (m, 2H), 2.55-2.64 (m, 2H), 4.90-5.00 (m, 1H), 7.85 (bs, 1H), 9.03 (d, $J = 1.6$ Hz, 2H), 9.17 (d, $J = 1.6$ Hz, 1H). ^{13}C NMR (75MHz, CDCl_3) δ 15.7, 27.3, 29.6, 59.1, 121.2, 127.5, 137.1, 148.5, 162.6, 205.6; MS (EI, 70ev): m/z (%) $[\text{M}]^+$ 288.4 (100%).

General procedure for synthesis of 5a-c:



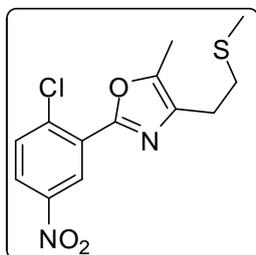
Phosphorus oxychloride (1 mL, 10 mmol) was added dropwise to a solution of (S)-N-(1-(Methylthio)-4-oxopentan-3-yl)-3-nitrobenzamide **4a** (3.3 mmol) in DMF (10 mL) at 0 °C. The mixture was heated to 90°C for 45 min and then cooled to 0 °C before being quenching with water and extracted with EtOAc. The combined organic phases were washed with water and brine (15 mL), dried over anhydrous Na_2SO_4 , filtered and the solvent evaporated. The residue was purified by column chromatography (EtOAc/hexanes) to provide of the desired products.

5-Methyl-4-(2-(methylthio) ethyl)-2-(3-nitrophenyl) oxazole (5a):



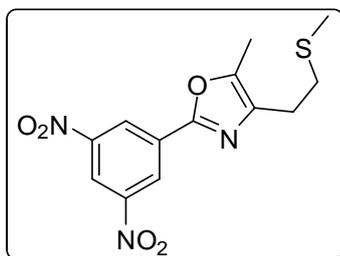
Brown semi solid; $R_f = 0.7$ (30% EtOAc-n-Hexane); Yield 84 % ; IR (cm^{-1}): 3060, 2955, 2862, 1520, 1321; $^1\text{H NMR}$ (300 MHz, CDCl_3) δ 2.15 (s, 3H), 2.39 (s, 3H), 2.78 -2.87 (m, 4H), 7.59 - 7.63 (t, $J = 8.0$ Hz, 1H), 8.23 (d, $J = 8.0$ Hz, 1H), 8.25 (d, $J = 1.6$ Hz, 1H), 8.8 (s, 1H) ; $^{13}\text{C NMR}$ (75 MHz, CDCl_3) δ 10.3, 15.7, 26.1, 33.5, 1207, 124.0, 129.3, 129.8, 131.4, 135.4, 145.5, 148.6, 157.1; MS (EI, 70ev): m/z (%) $[\text{M}]^+$ 279.0 (100%).

2-(2-Chloro-5-nitrophenyl)-5-methyl-4-(2-(methylthio) ethyl) oxazole (5b):



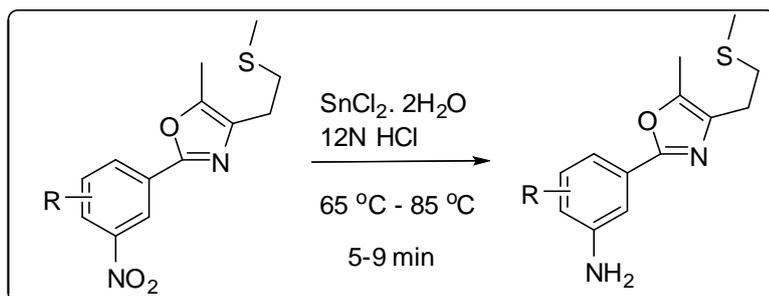
The title compound was synthesized as brown color semi solid in 80% yield according to the general procedure as described above for **5a**; $R_f = 0.7$ (30% EtOAc-n-Hexane); IR (cm^{-1}): 3070, 2978, 2856, 1510, 1318; $^1\text{H NMR}$ (300 MHz, CDCl_3) δ 2.15 (s, 3H), 2.41(s, 3H), 2.80-2.89 (m, 4H), 7.65 (d, $J = 8.4$ Hz, 1H), 8.14-8.16 (dd, $J^1 = 8.4$ Hz, $J^2 = 2.8$ Hz, 1H), 8.85 (d, $J = 2.8$ Hz, 1H); $^{13}\text{C NMR}$ (75 MHz, CDCl_3) δ 10.3, 15.7, 26.0, 33.5, 124.3, 125.8, 127.7, 132.3, 135.5, 138.4, 145.9, 146.4, 155.1MS (EI, 70ev): m/z (%) $[\text{M}]^+$ 313.1 (100%).

2-(3, 5-Dinitrophenyl)-5-methyl-4-(2-(methylthio) ethyl) oxazole (5c):



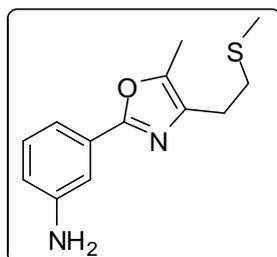
The title compound was synthesized as brown color semi solid in 88% yield according to the general procedure as described above for **5a**; $R_f = 0.72$ (30% EtOAc-n-Hexane); IR (cm^{-1}): 3048, 2978, 2856, 1530, 1325; $^1\text{H NMR}$ (300 MHz, CDCl_3) δ 2.15 (s, 3H), 2.43 (s, 3H), 2.80-2.89 (m, 4H), 9.02 (d, $J = 2.0$ Hz, 1H), 9.10 (d, $J = 2.0$ Hz, 2H). $^{13}\text{C NMR}$ (75MHz, CDCl_3) δ 10.4, 15.7, 25.9, 33.5, 118.6, 125.3, 130.9, 136.4, 146.9, 148.9, 155.2; MS (EI, 70ev): m/z (%) $[\text{M}]^+$ 324.2 (96%), 294.2(100%).

General procedure for synthesis of 6a-c:



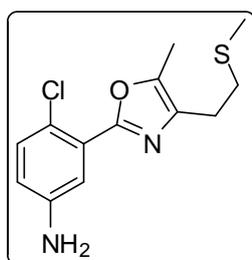
To a solution of 5-Methyl-4-(2-(methylthio) ethyl)-2-(3-nitrophenyl) oxazole (**5a**) (2.5 mmol) in 36% HCl (6 mL), stannouschloride monohydrate (12.5 mmol) was added. The mixture was heated to 100 °C for 15min, cooled to 0 °C the aqueous solution was basified with 2N NaOH and extracted three times with dichloromethane. The organic layers were combined, washed with brine solution and dried over anhydrous Na_2SO_4 and concentrated to give desired product **6a**.

3-(5-Methyl-4-(2-(methylthio) ethyl) oxazol-2-yl) aniline (**6a**):



Brown solid; $R_f = 0.4$ (30% EtOAc-n-Hexane); mp 292-294 °C; Yield (65 %); IR (cm^{-1}): 3390, 3350 (NH_2), 3020, 2910, 2858, 1650, 1110; ^1H NMR (300 MHz, CDCl_3) δ 2.12 (s, 3H), 2.33 (s, 3H), 2.74 - 2.85 (m, 4H), 6.70 - 6.73 (dd, $J^1 = 7.8$ Hz, $J^2 = 2.2$ Hz, 1H), 7.18 - 7.22 (m, 2H), 7.31 (t, $J = 1.6$ Hz, 1H), 7.32 - 7.37 (dt, $J^1 = 8.6$ Hz, $J^2 = 1.2$ Hz, 1H), 8.8 (s, 1H); ^{13}C NMR (75 MHz, CDCl_3) δ 10.2, 15.7, 26.1, 33.7, 112.1, 116.6, 116.9, 128.3, 129.7, 134.2, 144.4, 146.2, 159.6; MS (EI, 70ev): m/z (%) $[\text{M}]^+$ 249.3 (100%). HRMS $[\text{M}]^+$ Calcd. for $\text{C}_{13}\text{H}_{17}\text{N}_2\text{O}_5\text{S}$ 249.1062; Found: 249.1056.

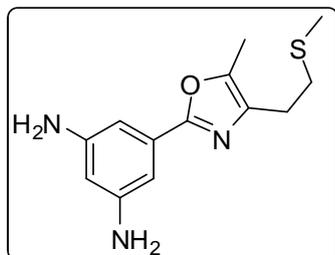
4-Chloro-3-(5-methyl-4-(2-(methylthio) ethyl) oxazol-2-yl) aniline (**6b**):



The title compound was synthesized as yellow color semi solid in 76% yield according to the general procedure as described above for **6a**; $R_f = 0.42$ (30% EtOAc-n-Hexane); IR (cm^{-1}): 3400, 3375

(NH₂), 3010, 2962, 2892, 1620, 1220; ¹H NMR (300 MHz, CDCl₃) δ 2.14 (s, 3H), 2.36 (s, 3H), 2.77-2.86 (m, 4H), 3.5 (bs, 2H), 6.63-6.66 (dd, *J*¹ = 8. Hz, *J*² = 2.8 Hz, 1H), 7.22-7.26 (dd, *J*¹ = 8.0 Hz, *J*² = 2.8 Hz, 2H). ¹³C NMR (75MHz, CDCl₃) δ 10.23, 15.6, 26.1, 33.6, 116.5, 117.6, 121.2, 126.8, 131.6, 134.3, 144.7, 157.6; MS (EI, 70ev): *m/z* (%) [M]⁺ 283.1 (100%). HRMS [M]⁺ Calcd. for C₁₃H₁₆N₂OSCl 283.0672; Found: 283.0675.

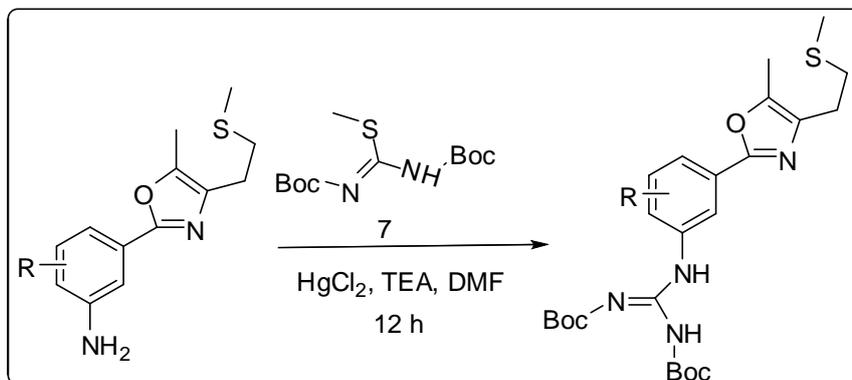
5-(5-Methyl-4-(2-(methylthio) ethyl) oxazol-2-yl) benzene-1, 3-diamine (6c):



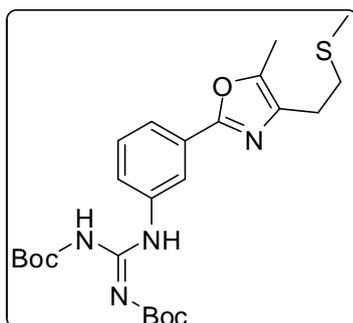
The title compound was synthesized as dark brown color semi solid in 76% yield according to the general procedure as described above for **6a**; *R_f* = 0.2 (30% EtOAc-n-Hexane); IR (cm⁻¹): 3425, 3396 (NH₂), 3030, 2954, 2892, 1600, 1550, 1190; ¹H NMR (300 MHz, CDCl₃) δ 2.12 (s, 3H), 2.32 (s, 3H), 2.73-2.83 (m, 4H), 3.69-3.78 (bs, 4H), 6.07 (t, *J* = 2.0 Hz, 1H), 6.74 (d, *J* = 2.0 Hz, 2H). ¹³C NMR (75MHz, CDCl₃) δ 15.7, 26.2, 29.7, 33.7, 40.9, 58.8, 71.63, 87.1, 103.1, 103.6, 125.9, 128.4, 128.5, 129.4, 134.3, 143.8, 147.7, 159.7; MS (EI, 70ev): *m/z* (%) [M]⁺ 264.1 (100%), 216.2 (78%) HRMS [M]⁺ Calcd. for C₁₃H₁₈N₃OS 264.1171; Found: 264.1174.

General procedure for synthesis of 8a-c:

To a solution of 3-(5-Methyl-4-(2-(methylthio) ethyl) oxazol-2-yl) aniline **6a** (0.4 mmol) in dryDMF (6 mL), N₁, N₂-bis (tert-butoxycarbonyl)-S-methylisothiurea (0.45 mmol), triethylamine (0.1 mL, 0.4 mmol) and HgCl₂ (or AgNO₃) (0.48 mmol) was added. The suspension was stirred at room temperature for 12 h then concentrated in vacuo. The crude reaction mixture was taken up in DCM (20 mL), filtered through a pad of celite on a sintered glass funnel, washed with saturated aqueous NH₄Cl (5 mL) and organic layer was washed with brine (50 mL), and dried over Na₂SO₄ and concentrated to give desired product (**8a**).

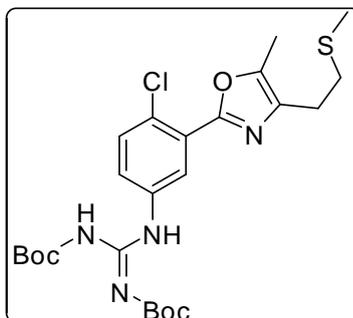


(E)-Tert-butyl (tert-butoxycarbonylamino) (3-(5-methyl-4-(2-(methylthio) ethyl) oxazol-2-yl) phenylamino) methylenecarbamate (8a):



Colorless semi solid; $R_f = 0.8$ (30% EtOAc-n-Hexane); Yield (78%); IR (cm^{-1}): 3261(NH), 3154, 2978, 2926, 1721(amide), 1644 (C=N); ^1H NMR (300 MHz, CDCl_3) δ 1.52(s, 9H), 1.53(s, 9H), 2.14 (s, 3H), 2.35 (s, 3H), 2.75 -2.85 (m, 4H), 7.40 (t, $J = 8.0$ Hz, 1H), 7.71(d, $J = 8.0$ Hz, 1H), 7.92 (t, $J = 2.2$ Hz, 1H), 8.06 (s, 1H), 10.45(bs, 1H), 11.63(bs, 1H) ; ^{13}C NMR (75 MHz, CDCl_3) δ 10.2, 15.7, 26.2, 33.7, 112.3, 116.3, 116.6, 128.5, 129.6, 134.4, 143.9, 146.5, 159.6; MS (EI, 70ev): m/z (%) $[\text{M}]^+$ 491.23 (100%). HRMS $[\text{M}]^+$ Calcd. for $\text{C}_{24}\text{H}_{35}\text{N}_4\text{O}_5\text{S}$ 491.2328; Found: 491.2316.

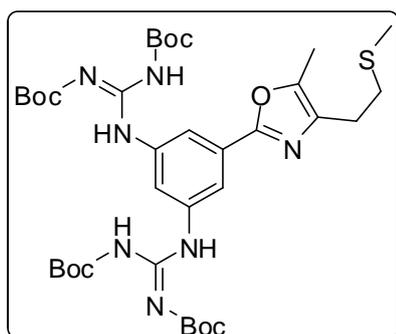
(E)-tert-butyl (tert-butoxycarbonylamino) (4-chloro-3-(5-methyl-4-(2 (methylthio) ethyl) oxazol-2-yl) phenylamino) methylenecarbamate (8b):



The title compound was synthesized as colorless solid in 72% yield according to the general procedure as described above for **8a**; $R_f = 0.8$ (30% EtOAc-n-Hexane); mp 310-312 °C

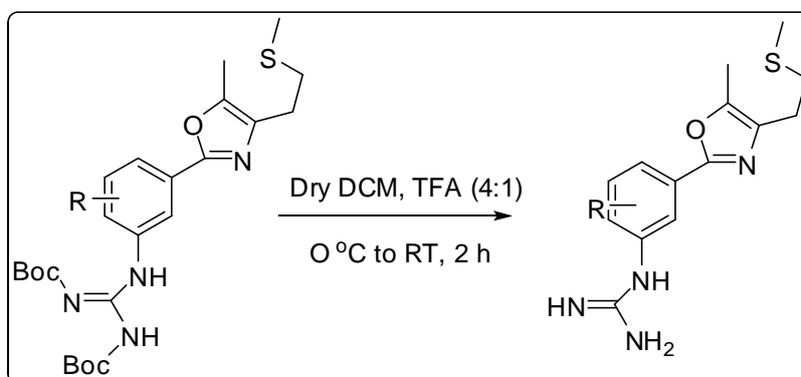
IR (cm⁻¹): 3321(NH), 3124, 2978, 2862, 1700 (amide), 1665 (C=N); ¹H NMR (300 MHz, CDCl₃) δ 1.51 (s, 9H), 1.53 (s, 9H), 2.14 (s, 3H), 2.37 (s, 3H), 2.80-2.86 (m, 4H), 7.43 (d, *J* = 8.8 Hz, 1H), 7.87-7.90 (dd, *J*¹ = 8.8 Hz, *J*² = 2.4 Hz, 1H), 8.04 (d, *J* = 2.4 Hz, 1H). ¹³C NMR (75MHz, CDCl₃) δ 10.3, 15.7, 26.2, 28.0, 28.1, 29.7, 33.6, 79.9, 84.0, 123.2, 124.2, 126.7, 127.4, 131.6, 134.7, 135.8, 144.9, 153.2, 153.4, 156.9, 163.2; MS (EI, 70ev): *m/z* (%) [M]⁺ 542.3 (10%), 525.3 (100%) HRMS [M]⁺ Calcd. for C₂₄H₃₄N₄O₆SCl 541.1874; Found: 541.1865.

Tert-butyl(5-(5-methyl-4-(2-(methylthio)ethyl)oxazol-2-yl)-1,3-phenylene)bis(azanediyl)bis((tert-butoxycarbonylamino) methan-1-yl-1-ylidene) dicarbamate (8c):



The title compound was synthesized as colorless solid in 80% yield according to the general procedure as described above for **8a**; *R_f* = 0.85 (30% EtOAc-n-Hexane); mp 369-372 °C · IR (cm⁻¹): 3361(NH), 3133, 2958, 2862, 1710(amide), 1660 (C=N); ¹H NMR (300 MHz, CDCl₃): 1.52 (s, 3H), 2.13 (s, 3H), 2.34 (s, 3H), 2.74-2.85 (m, 4H), 8.12 (d, *J* = 1.6 Hz, 2H), 8.16 (s, 1H), 10.48 (s, 2H), 11.54 (bs, 2H). ¹³C NMR (75MHz, CDCl₃): 10.1, 15.7, 26.2, 28.1, 33.6, 79.6, 83.8, 115.4, 116.5, 128.7, 134.7, 137.9, 144.3, 153.3, 158.8, 163.2; MS (EI, 70ev): *m/z* (%) [M]⁺ 748.36 (68%), 348.15 (100%).

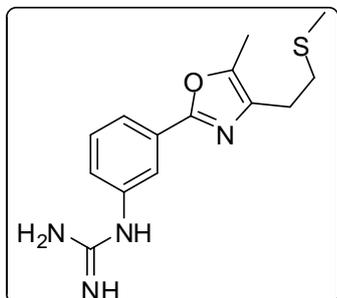
General procedure for the synthesis of 9a-c:



A solution containing (E)-Tert-butyl (tert-butoxycarbonylamino) (3-(5-methyl-4-(2-(methylthio) ethyl) oxazol-2-yl) phenylamino) methylenecarbamate **8a** (0.023 mmol) in DCM (1 ml), 1mL of TFA was added at 0 °C, and then stirred at RT for 1 h. The solvent was removed under high vacuum to generate the

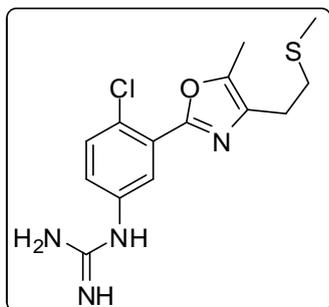
trifluoroacetate salt. This salt was redissolved in 20 mL of an aqueous solution of NaHCO₃ and washed with DCM (3 × 15 mL). The organic layer was washed with water (2 × 10 mL), followed by brine wash, dried over anhydrous Na₂SO₄ and concentrated to give the corresponding free guanidine (**9a**).

1-(3-(5-Methyl-4-(2-(methylthio) ethyl) oxazol-2-yl) phenyl) guanidine (9a):



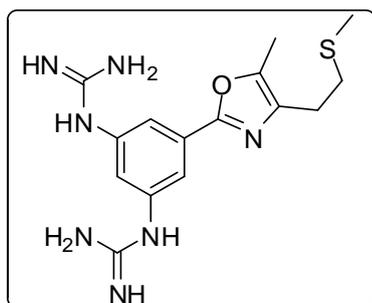
Color less semi solid; $R_f = 0.3$ (5% MeOH-CH₂Cl₂); Yield (85%); IR (cm⁻¹): 3340(NH), 3178, 2963, 2854, 1680 (C=N); ¹H NMR (300 MHz, CDCl₃) δ 2.15 (s, 3H), 2.33 (s, 3H), 2.74 -2.84 (m, 4H), 2.86- 3.40(bs, 4H), 6.98 (d, $J = 8.0$ Hz, 1H), 7.33 (t, $J = 8.0$ Hz, 1H), 7.56 (s, 1H), 7.61 (d, $J = 8.0$ Hz, 1H); ¹³C NMR (75 MHz, CDCl₃) δ 8.6, 14.3, 29.3, 33.1, 122.2, 124.4, 126.6, 128.7, 130.4, 134.5, 135.7, 145.7, 156.6, 158.6; MS (EI, 70ev): m/z (%) [M]⁺ 291.3 (100%). HRMS [M]⁺ Calcd. for C₁₄H₁₈N₄OS 291.3228; Found: 291.3216.

1-(4-Chloro-3-(5-methyl-4-(2-(methylthio) ethyl) oxazol-2-yl) phenyl) guanidine (9b):



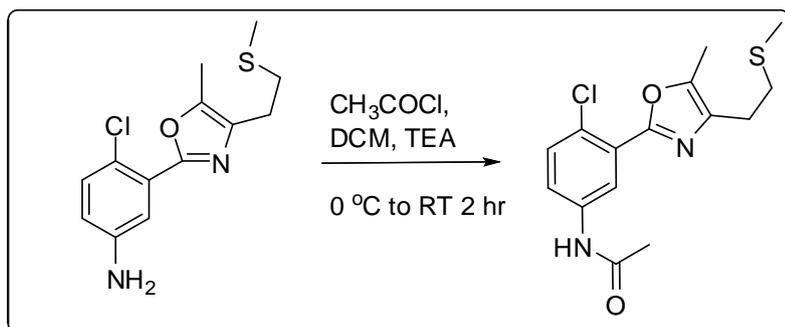
The title compound was synthesized as colorless semi solid in 80% yield according to the general procedure as described above for **9a**; $R_f = 0.3$ (5% MeOH-CH₂Cl₂); Yield (74%); IR (cm⁻¹): 3420(NH), 3178, 2978, 2921, 2834, 1690 (C=N); ¹H NMR (300 MHz, CDCl₃) δ 2.18 (s, 3H), 2.38 (s, 3H), 2.71 -2.89 (m, 4H), 7.10-7.30 (m, 2H), 7.52 (d, $J = 8.0$ Hz, 1H), 7.78 (s, 1H), 10.00 (s, 1H), 10.29 (bs, 2H) ; ¹³C NMR (75 MHz, CDCl₃) δ 10.2, 15.5, 29.7, 33.4, 126.7, 127.2, 128.2, 131.7, 132.9, 133.2, 146.9, 156.4 ; MS (EI, 70ev): m/z (%) [M]⁺ 291.3 (100%). MS (EI, 70ev): m/z (%) [M]⁺ 324.3 (100%). HRMS [M]⁺ Calcd. for C₁₄H₁₇ClN₄OS 324.3134; Found: 324.3146.

1,1'-(5-(5-Methyl-4-(2-(methylthio) ethyl) oxazol-2-yl)-1,3-phenylene)diguanidine (9c):



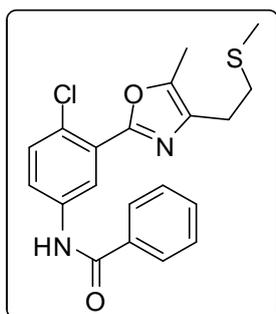
The title compound was synthesized as white color solid in 80% yield according to the general procedure as described above for **9a**; mp 308-311 °C; $R_f = 0.3$ (15% MeOH-CH₂Cl₂); Yield (80%); IR (cm⁻¹): 3390 (NH), 3156, 2985, 2911, 2865, 1676 (C=N); ¹H NMR (300 MHz, CD₃OD) δ 2.06 (s, 3H), 2.33 (s, 3H), 2.74-2.77 (m, 4H), 7.24 (s, 1H), 7.79 (s, 2H); ¹³C NMR (75MHz, CD₃OD) δ 10.1, 15.5, 26.6, 34.4, 121.7, 124.0, 131.8, 136.5, 138.7, 147.6, 158.1, 158.9; MS (EI, 70ev): m/z (%) [M]⁺ 348.1 (100%) HRMS [M]⁺ Calcd. for C₁₅H₂₂N₇OS 348.1607; Found: 348.1612.

N-(4-Chloro-3-(5-methyl-4-(2-(methylthio) ethyl) oxazol-2-yl)phenyl)acetamide (10):



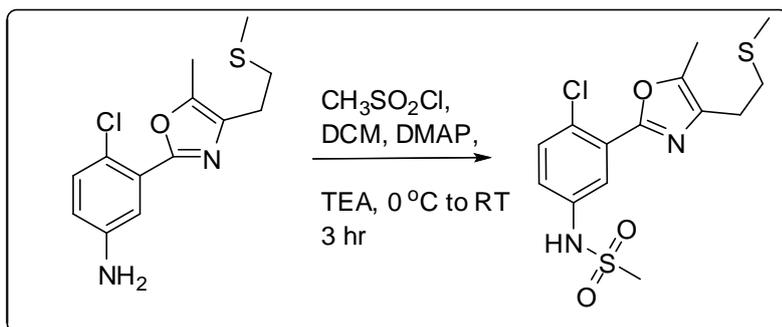
Acetyl chloride (1.2 mmol) was added to a mixture of 4-chloro-3-(5-methyl-4-(2-(methylthio) ethyl) oxazol-2-yl) aniline (**6b**) (1 mmol), triethylamine(2.4 mmol) in dry dichloromethane (10 mL) at 0 °C. The reaction mixture was stirred for 2 hr followed by drop wise addition of 2N HCl. The residue was dissolved in dichloromethane (50 mL), washed with water (2 x 250 mL), brine (1 x 100 mL), dried over Na₂SO₄, filtered and the solvent evaporated. The residue was purified by flash chromatography (n-hexane/ ethylacetate 4:1) to afford the desired product (**10**) as yellow colour semi solid in 90% yield; $R_f = 0.42$ (30% EtOAc-n-Hexane); IR (cm⁻¹): 3345, 3066, 2966, 2866, 1685 (C=O of amide), 1340; ¹H NMR (300 MHz, CDCl₃) δ 1.88 (s, 3H), 2.15 (s, 3H), 2.50 (s, 3H), 2.87-2.98 (m, 4H), 7.12 (d, $J = 8.1$ Hz, 1H), 7.64 (dd, $J^1 = 8.1$ Hz, $J^2 = 2.8$ Hz, 2H), 7.82 (s, 1H), 9.86 (bs, 1H) . ¹³C NMR (75MHz, CDCl₃) δ 10.3, 15.6, 23.6, 29.6, 33.6, 119.8, 127.6, 131.8, 131.9, 134.9, 138.0, 145.3, 156.1, 169.6; MS (EI, 70ev): m/z (%) [M]⁺ 323.15 (100%).

***N*-(4-Chloro-3-(5-methyl-4-(2-(methylthio) ethyl) oxazol-2-yl)phenyl)benzamide (11):**



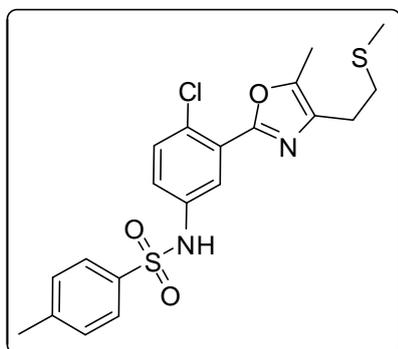
The title compound was synthesized as colourless semi solid in 86% yield according to the procedure as described above for **10** (instead of acetyl chloride benzoyl chloride was taken); $R_f = 0.42$ (10% EtOAc-n-Hexane); IR (cm^{-1}): 3195, 3052, 2965, 2875, 1679 (C=O of amide), 1314; ^1H NMR (300 MHz, CDCl_3) δ 2.16 (s, 3H), 2.48 (s, 3H), 2.96-3.08 (m, 4H), 7.36 (d, $J= 8.2$ Hz, 1H), 7.47-7.52 (m, 2H), 7.63-7.74 (m, 2H), 8.06 (d, $J= 7.2$ Hz, 1H), 8.85 (bs, 1H). ^{13}C NMR (75MHz, CDCl_3) δ 11.2, 15.1, 29.8, 33.8, 121.1, 125.1, 126.9, 127.9, 128.6, 130.7, 132.0, 136.9, 138.8, 158.6, 166.6; MS (EI, 70ev): m/z (%) $[\text{M}]^-$ 385.27 (100%).

***N*-(4-Chloro-3-(5-methyl-4-(2-(methylthio)ethyl)oxazol-2-yl)phenyl)methanesulfonamide (12):**



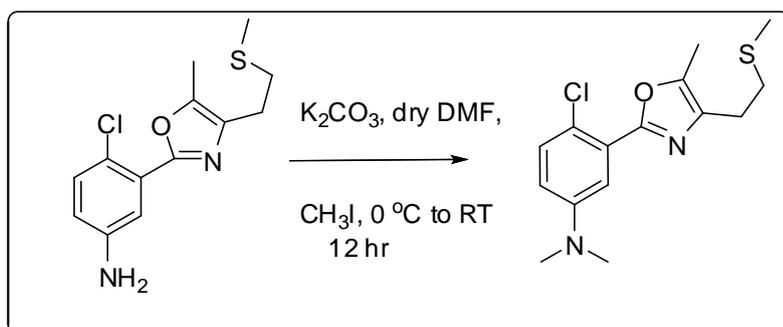
Methane sulfonyl chloride (1.2 mmol) was added to a mixture of 4-chloro-3-(5-methyl-4-(2-(methylthio) ethyl) oxazol-2-yl) aniline (**6b**) (1 mmol), triethylamine (2.4 mmol) and DMAP (0.1 mmol) in dry dichloromethane (10 mL) at 0 °C. The reaction mixture was stirred for 3 hr followed by drop wise addition of 2N HCl. The residue was dissolved in dichloromethane (50 mL), washed with water (2 x 150 mL), brine (1 x 50 mL), dried over Na_2SO_4 , filtered and the solvent evaporated. The residue was purified by flash chromatography (n-hexane/ ethylacetate 3.5:1.5) to afford the desired product (**12**) as brown colour semi solid in 90% yield; $R_f = 0.30$ (30% EtOAc-n-Hexane); IR (cm^{-1}): 3290, 3050, 2985, 2865, 1680 (C=O of amide), 1326; ^1H NMR (300 MHz, CDCl_3) δ 2.25 (s, 3H), 2.35 (s, 3H), 2.75 (s, 3H), 2.87-3.02 (m, 4H), 5.96 (bs, 1H), 6.85 (d, $J= 8.4$ Hz, 1H), 7.13 (d, $J= 8.6$ Hz, 1H), 7.40-7.46 (m, 1H). ^{13}C NMR (75MHz, CDCl_3) δ 11.5, 15.6, 28.6, 36.6, 39.3, 115.5, 121.1, 125.0, 130.4, 136.7, 138.6, 144.5, 158.6; MS (EI, 70ev): m/z (%) $[\text{M}]^+$ 360.97 (100%).

***N*-(4-Chloro-3-(5-methyl-4-(2-(methylthio)ethyl)oxazol-2-yl)phenyl)-4-methylbenzene sulphonamide (13):**



The title compound was synthesized as yellow colour semi solid in 81% yield according to the procedure as described above for **12** (instead of methane sulfonyl chloride 4-toluene sulfonyl chloride was taken); $R_f = 0.6$ (10% EtOAc-n-Hexane); IR (cm^{-1}): 3210, 3010, 2965, 2875, 1690 (C=O of amide), 1314; ^1H NMR (300 MHz, CDCl_3) δ 2.20 (s, 3H), 2.32 (s, 3H), 2.41(s, 3H), 2.85-2.96 (m, 4H), 6.15 (bs, 1H), 6.91 (d, $J = 8.4$ Hz, 1H), 6.98 (d, $J = 7.2$ Hz, 1H), 7.21-7.26 (m, 1H), 7.41 (d, $J = 8.2$ Hz, 2H), 7.62 (d, $J = 8.2$ Hz, 1H). ^{13}C NMR (75MHz, CDCl_3) δ 10.3, 15.6, 20.9, 25.9, 35.9, 110.5, 121.7, 124.5, 128.7, 130.2, 130.9, 136.2, 136.9, 139.9, 157.7; MS (EI, 70ev): m/z (%) $[\text{M}]^+$ 437.23 (100%).

4-Chloro-*N,N*-dimethyl-3-(5-methyl-4-(2-(methylthio)ethyl)oxazol-2-yl)aniline (14):



Methyl iodide (6.0 mmol) was added to a mixture of 4-chloro-3-(5-methyl-4-(2-(methylthio)ethyl) oxazol-2-yl) aniline (**6b**) (1 mmol), anhydrous K_2CO_3 (6.0 mmol) in dry dimethyl formamide (10 mL) at 0 °C. The reaction mixture was stirred for 12 hr followed by drop wise addition of 1N HCl at 0 °C. The residue was dissolved in dichloromethane (2 x 50 mL), washed with water (2 x 150 mL), brine (1 x 50 mL), dried over Na_2SO_4 , filtered and the solvent evaporated. The residue was purified by flash chromatography (n-hexane/ ethylacetate 3:2) to afford the desired product (**14**) as brown colour semi solid in 65% yield; $R_f = 0.20$ (40% EtOAc-

n-Hexane); IR (cm^{-1}): 3050, 2972, 2872, 1630, 1290; ^1H NMR (300 MHz, CDCl_3) δ 2.14 (s, 3H), 2.45 (s, 3H), 3.05-3.16 (m, 4H), 3.55 (s, 6H), 6.82 (d, $J= 8.4$ Hz, 1H), 6.96 (d, $J= 7.8$ Hz, 1H), 7.15 (m, 1H). ^{13}C NMR (75MHz, CDCl_3) δ 10.3, 15.6, 26.0, 31.8, 45.9, 111.7, 117.5, 124.5, 125.2, 128.7, 136.9, 145.3, 148.2, 155.6; MS (EI, 70ev): m/z (%) $[\text{M}]^+$ 311.17 (100%).

1.2 Pharmacology

1.2.1 Evaluation of cell viability

The MTT assay was used for evaluating the effect of the compounds on cell viability. RPMI medium supplemented with 10% FBS and antibiotics was used to culture HeLa and MDA-MB-231 cells. MTT dye prepared in 1X PBS was used for cell viability assays. HeLa and MDA-MB-231 cells in monolayer were trypsinised and the cell count was adjusted to 3×10^6 cells/ml using medium containing 10% fetal calf serum. Cells were pre-incubated at a concentration of 1×10^6 cells/ml in culture medium for 3h at 37°C and 6.5% CO_2 . Cells were then seeded at a concentration of 5×10^4 cells/well in 100 μl culture medium and the plates incubated at 37°C in 5% CO_2 incubator for 24 h. After 24h, 100 μl of previously diluted compounds of different concentrations of test compounds in media were added and incubated at 37°C in 5% CO_2 for 72 h and cells were periodically checked for granularity, shrinkage and swelling. After 72 h, 10 ml of MTT dye was added to each well. The plates were gently shaken and incubated for 4h at 37°C in 5% CO_2 . The supernatant was removed and 100 μl of 1:1 DMSO: Ethanol was added and the plates were gently shaken to solubilize the formed formazan. The absorbance was measured using a microplate reader at a wavelength of 550 nm. The data was plotted with log (compound concentration) on the x-axis and % viability of cells on the y-axis.

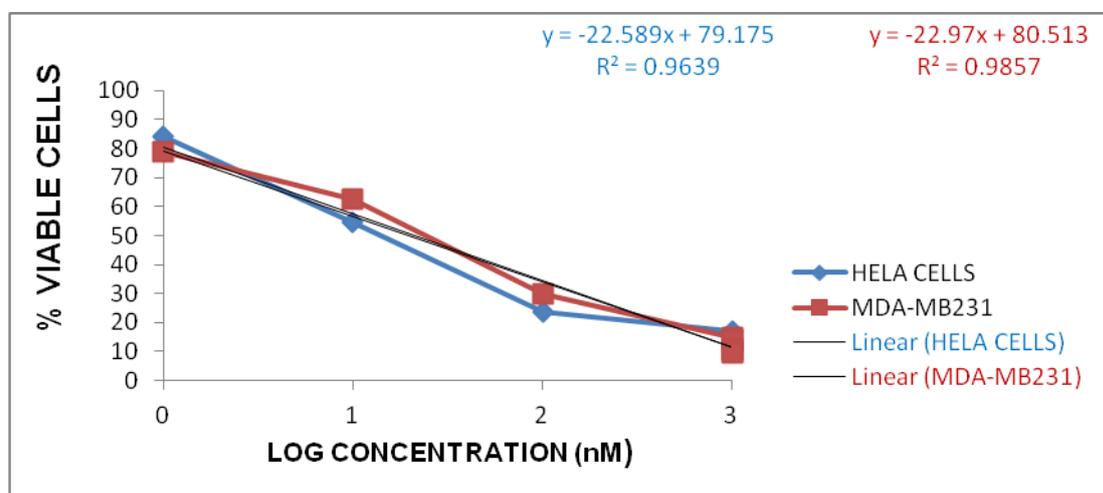


Figure S1: Log Concentration Vs % Viable Cells for Compound **6a** against HeLa and MDA-MB231 Cells by MTT assay

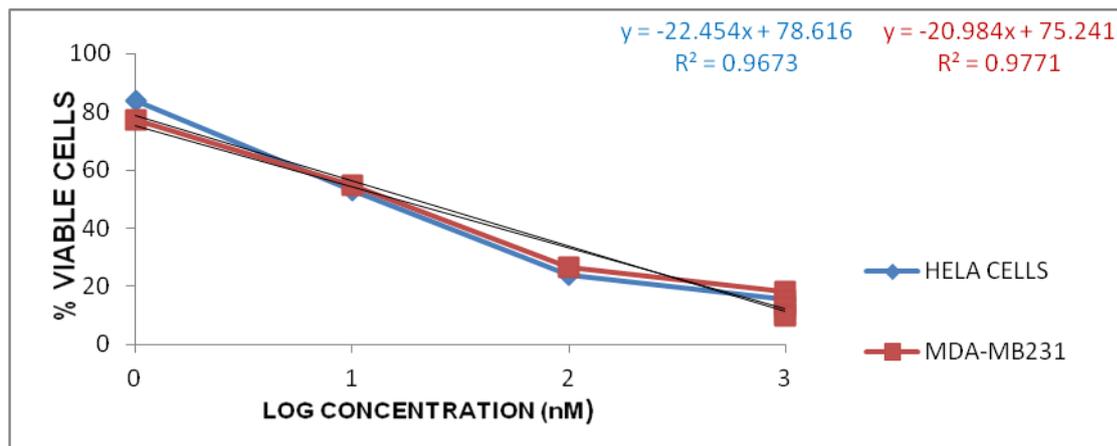


Figure S2: Log Concentration Vs % Viable Cells for Compound **6b** against HeLa and MDA-MB231 Cells by MTT assay

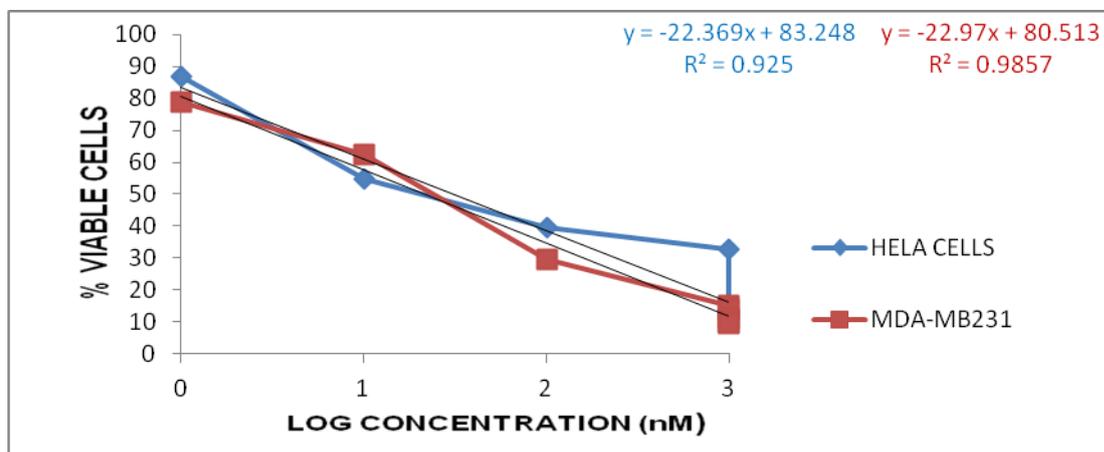


Figure S3: Log Concentration Vs % Viable Cells for Compound **9a** against HeLa and MDA-MB231 Cells by MTT assay

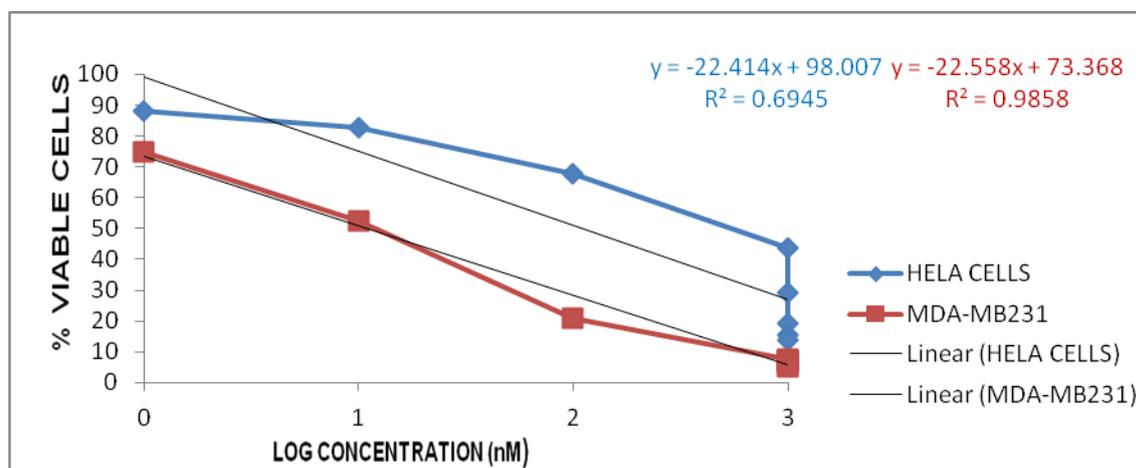


Figure S4: Log Concentration Vs % Viable Cells for Compound **9c** against HeLa and MDA-MB231 Cells by MTT assay

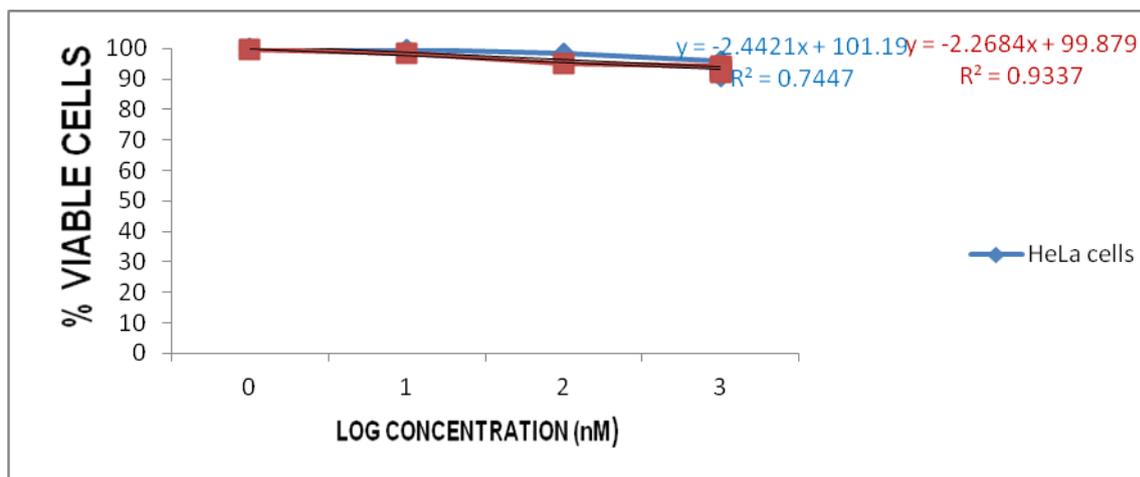
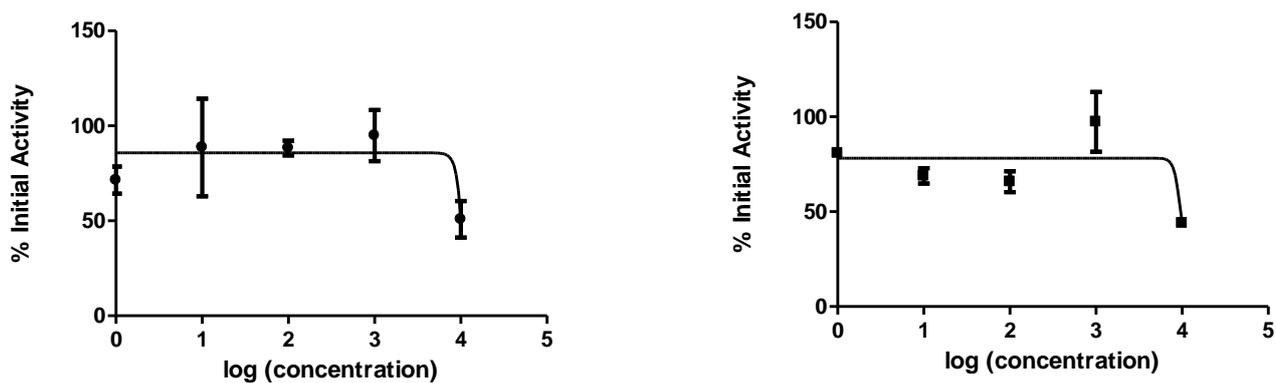


Figure S5: Log Concentration Vs % Viable Cells for 10% DMSO Control against HeLa and MDA-MB231 Cells by MTT assay

1.2.2 Evaluation of *in vitro* LSD1 activity

The effect of the compounds was evaluated in an *in vitro* LSD1 enzyme assay. The compounds were dissolved in 100% DMSO to a stock concentration of 10mM and used in the assay as per the manufacturer's instructions (Cayman Chemical Company, LSD1 screening kit Catalog # 700120). Briefly, the assay is based on the generation of the product Resorufin by the enzyme Horse radish peroxidase in the presence of hydrogen peroxide, which is produced by the demethylation of a methylated histone H3K4 peptide by LSD1. The amount of Resorufin generated is measured spectrophotometrically at 595/60 nm and is an indicator of LSD1 activity. The assay was performed in a 96-well plate format with triplicates of all samples. Absorbance values were normalized to background (lacking the substrate peptide) and % initial activity was calculated using the formula $100 - [(I-S)/S] * 100$, where I was the absorbance of the Initial activity samples (without inhibition) and S was the absorbance of the test samples (with various compounds). The data was analyzed and IC₅₀ values calculated using non-linear regression analysis in the software GraphPad Prism. The data was plotted with log (compound concentration) on the x-axis and % initial activity on the y-axis.



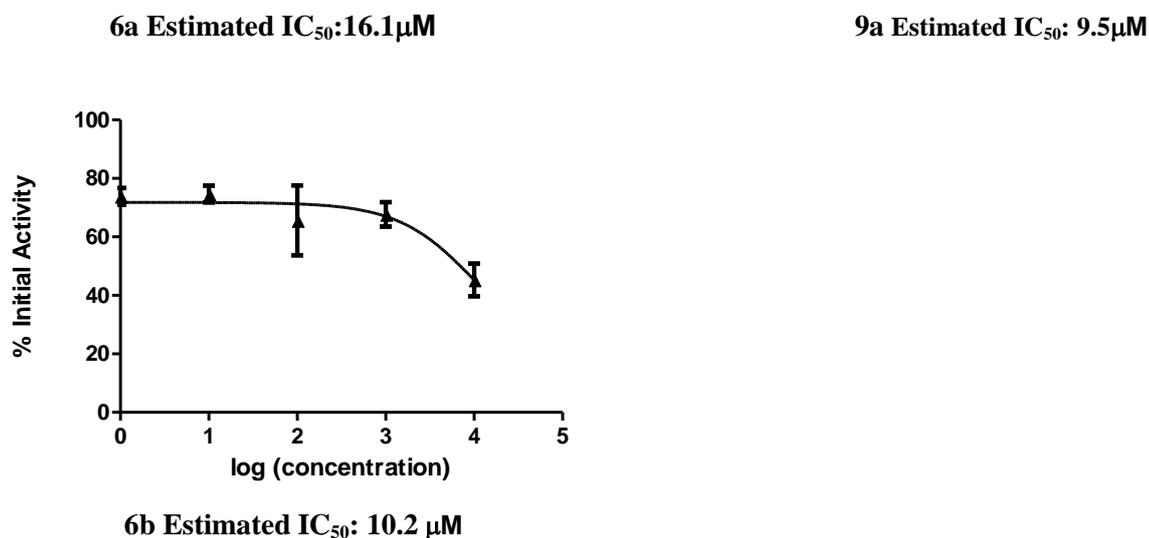


Figure S6: Log Concentration Vs % Initial activity for compounds **6a**, **6b** and **9a** against LSD1

1.2.3 Evaluation of *in vivo* apoptosis and toxicity using zebrafish embryos

All procedures using zebrafish were in accordance with ethical guidelines for animal use and followed those published by the NIH. An indigenous wild type adult zebrafish strain from India was used for this study (obtained from Vikrant Aquaculture, Mumbai, India). They were maintained in a recirculation system using purified ELIX System (Millipore, Billerica, US) grade water containing 200 mg/l sea salt at 28 °C under a 14:10 h light and dark cycle. Fish were fed three times daily with a combination of freshly hatched live brine shrimp and dry food. Males and females were kept in separate tanks for four days before they were allowed to spawn. On day five, 300-400 embryos were obtained by natural mating of both adult sexes (3:2 female and male ratios) in a breeding tank setup. Embryos were collected and raised in 60 mm Petri dishes containing E3 medium (5 mM NaCl, 0.17 mM KCl, 0.33 mM CaCl₂, and 0.33 mM MgSO₄) for 24 h. Compound stocks were diluted in E3 medium to obtain final concentrations of 10 μM in 0.1% DMSO. Zebrafish embryos at 24 h post fertilization were dechorionated with 500 μg/ml Pronase K for 10 minutes. Six embryos per well were placed in 24-well plates containing E3 medium and compounds added to a final concentration of 10 μM (in 0.1% DMSO). The embryos were grown in the presence of the compounds for 24 h, 48 h and 72 h. Control embryos were incubated in 0.1% DMSO. Apoptosis was assessed at the respective time points using acridine orange staining. Briefly, embryos were washed twice with 1X PBS (phosphate buffer saline) for 5min each, followed by incubation in acridine orange solution (5 μg/ml in E3 medium) for 1h at room temperature. Embryos were then washed with 1X PBS three times for 5min each and visualized under an upright fluorescence microscope (Zeiss Axioscope) at an excitation wavelength of 502 nm and an emission wavelength of 525 nm. Acridine orange is a fluorescent dye that binds to DNA and exhibits increased fluorescence in

fragmented nuclei, a hallmark of apoptosis^[1-2]. Embryos were also observed using visible light microscopy at regular intervals after compound treatment to document general toxicity-related effects such as developmental delays, deformations, edema and death.

1.3 Molecular Modeling Studies

1.3.1 Docking method

We carried out Molecular modeling (Docking analysis and Molecular dynamics simulations) studies to reveal the binding mode and ligand receptor complex stability. The docking studies of most potent molecules were performed using the Schrodinger software suite (Maestro, version 9.2, Schrodinger, LLC: New York, NY 2012). The compounds were sketched in 3D format using build panel and were prepared for docking using ligprep application. The Protein (PDB ID: 2Z3Y)^[1-2] for docking study was retrieved from protein data bank (PDB). The protein was prepared by giving preliminary treatment like adding hydrogen, adding missing residues, refining the loop with prime and finally minimized by using OPLS 2005 force field. Grids for molecular docking were generated with bound co-crystallized ligand. Compounds were docked using Glide (Glide version 5.7, Schrodinger, LLC: New York, NY 2012) in extra-precision mode, with up to three poses saved per molecule.

Table S1: Contribution of glide XP terms in docking score

Compound	GScore	LipophilicEvdW	HBond	Electro	PhobEn	LowMW
6a	-5.43	-3.9	-0.7	-0.2	-0.5	-0.5
6b	-6.50	-4.8	-0.8	-0.4	-0.3	-0.5
9a	-6.74	-5.0	-0.7	-0.5	-0.5	-0.5

LipophilicEvdW: Chemscore lipophilic pair term and fraction of the total protein-ligand vdw energy

HBond: Rewards for hydrogen bonding interaction between ligand and protein

Electro: Electrostatic reward

PhobEn: Hydrophobic enclosure reward

LowMW: Reward for ligands with low molecular weight

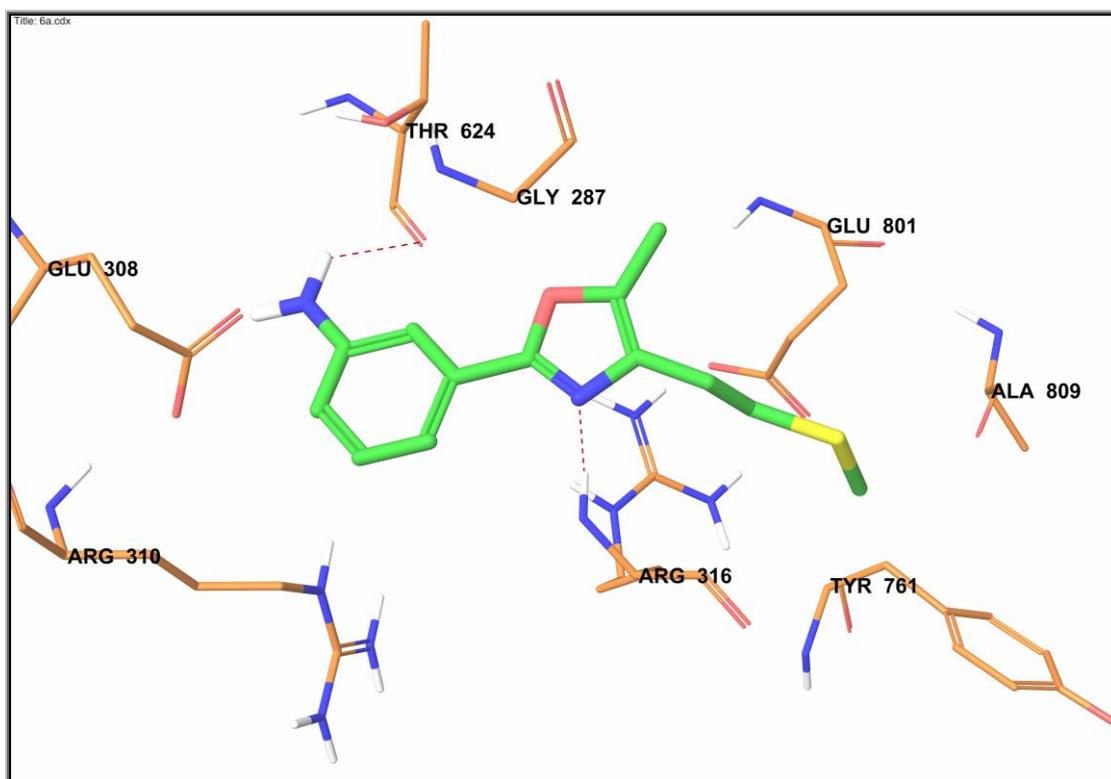
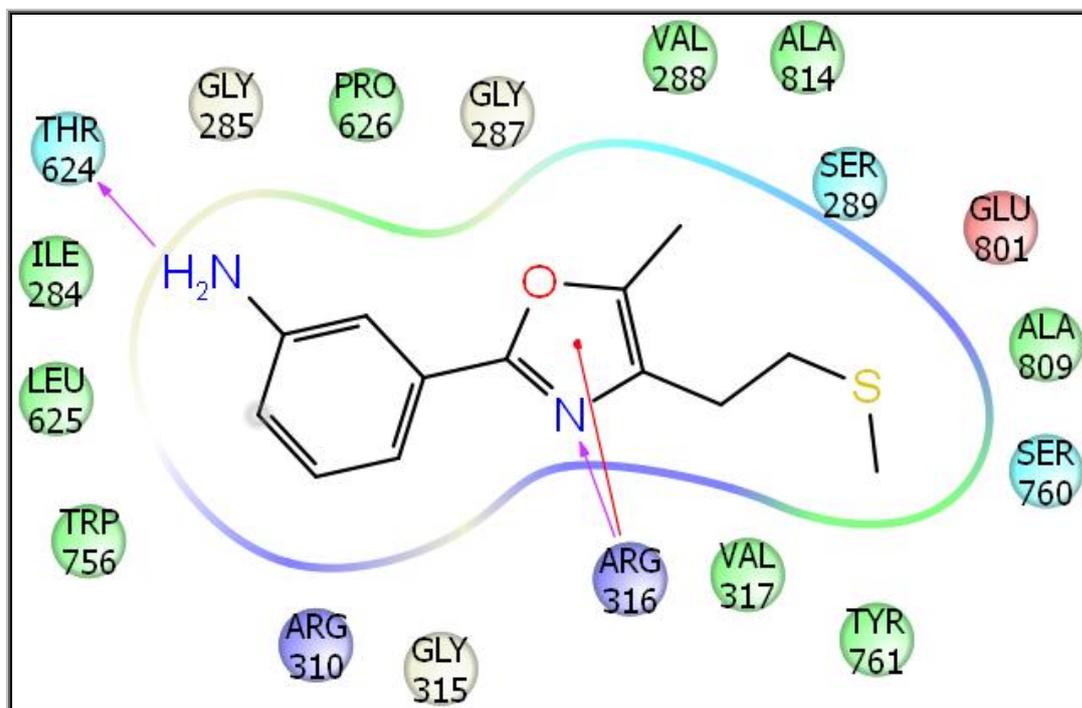


Figure S7: Binding orientation and interactions of **6a** at the LSD1 inhibitors binding site.

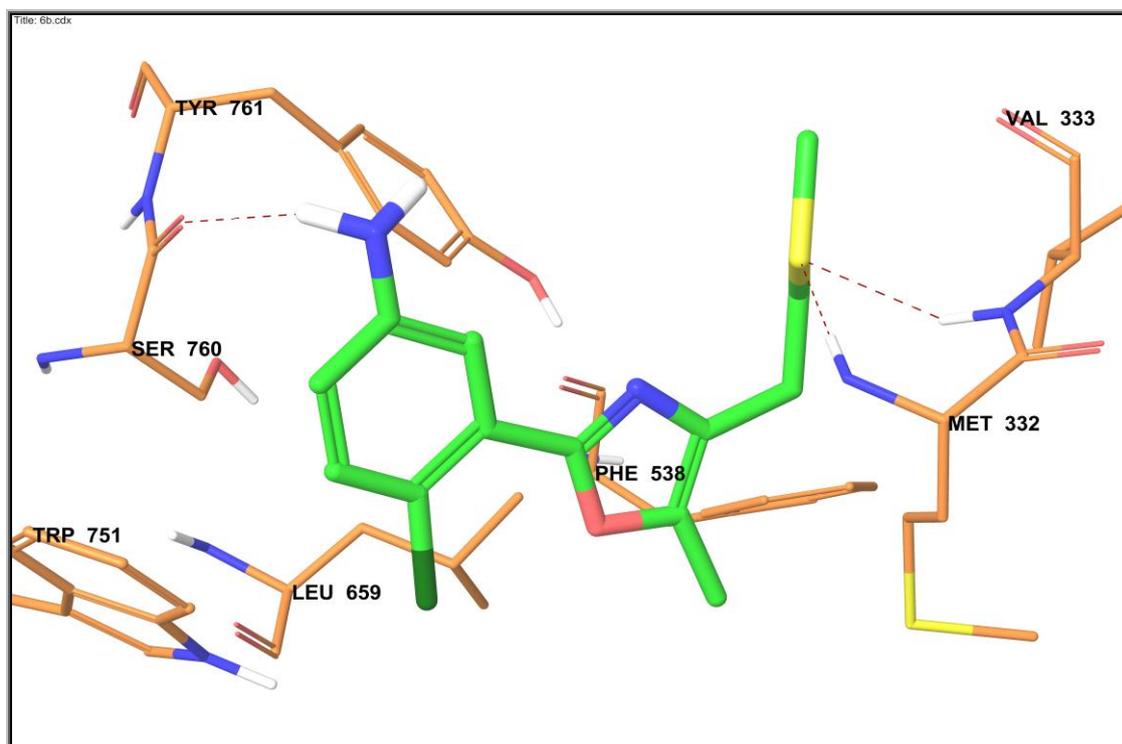
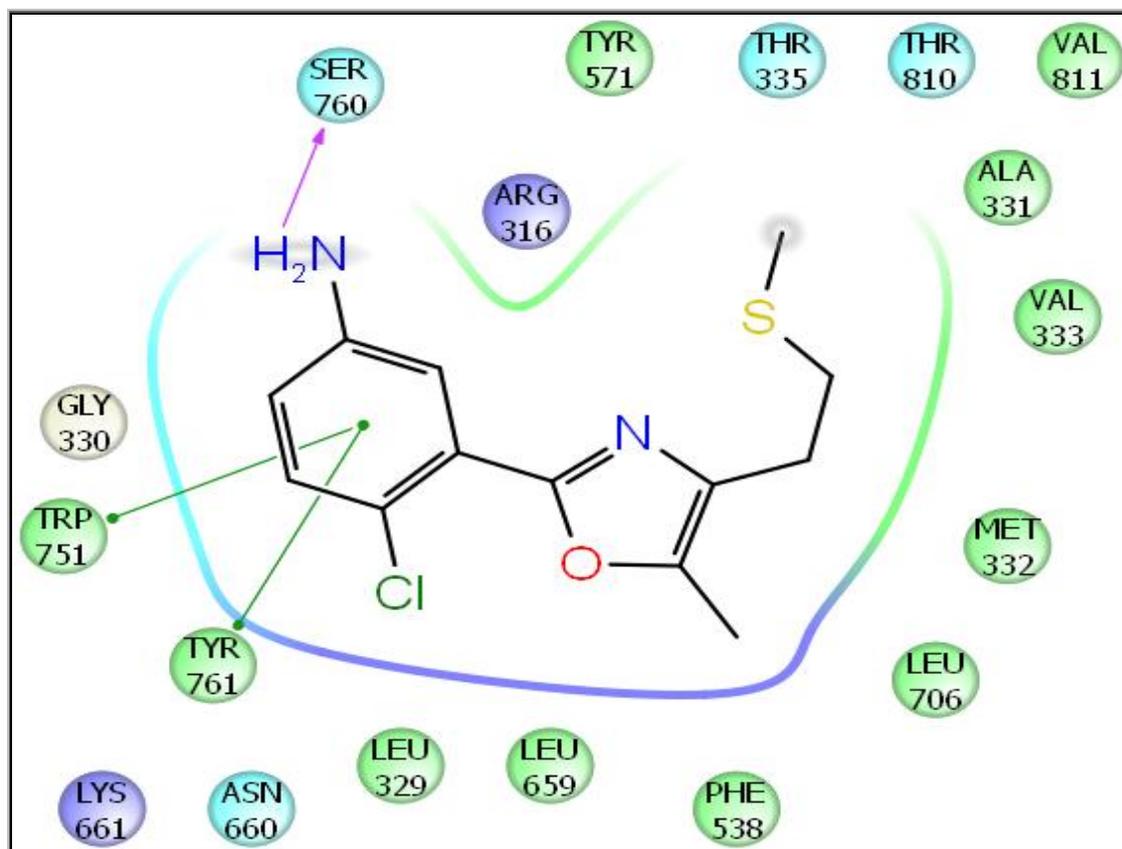


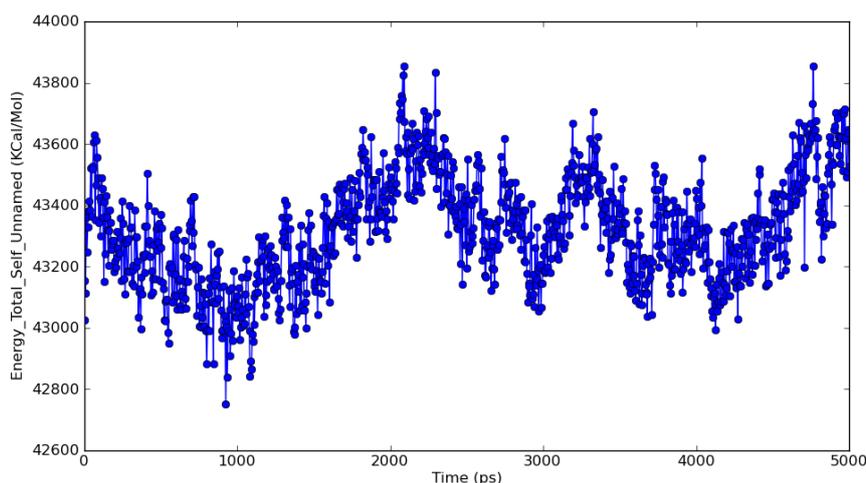
Figure S8: Binding orientation and interactions of **6b** at the LSD1 inhibitors binding site.

1.3.2 Molecular Dynamics Simulations Protocols:

Molecular dynamics (MD) simulations of **9a** at the binding site of protein were performed using the Desmond package incorporated in the Maestro^[3]. The system was built by applying OPLS-AA force field in an explicit solvent with the single point charges (SPC) water model (OPLS-AA/SPC). The initial coordinates for the MD calculations were taken from the docking experiments. The SPC water molecules were then added and system was neutralized by adding Na⁺ counter-ion to balance the net charges of the system. After the construction of the solvent environment, the complex system was composed of approximately 126117 atoms. Before equilibration and long production MD simulations, the systems were minimized and pre-equilibrated using the default relaxation routine implemented in Desmond.

The MD simulations were run for 5 ns and during the MD simulations, the equations of motion were integrated with a 2 fs time step in the NVT ensemble. The SHAKE algorithm was applied to all hydrogen atoms; the van der Waals (VDW) cutoff was set to 9 Å. The temperature was maintained at 300 K, employing the Nosé–Hoover thermostat method with a relaxation time of 1ps. The trajectory recording interval was kept for every 10 ps during entire MD runs.

(A)



(B)

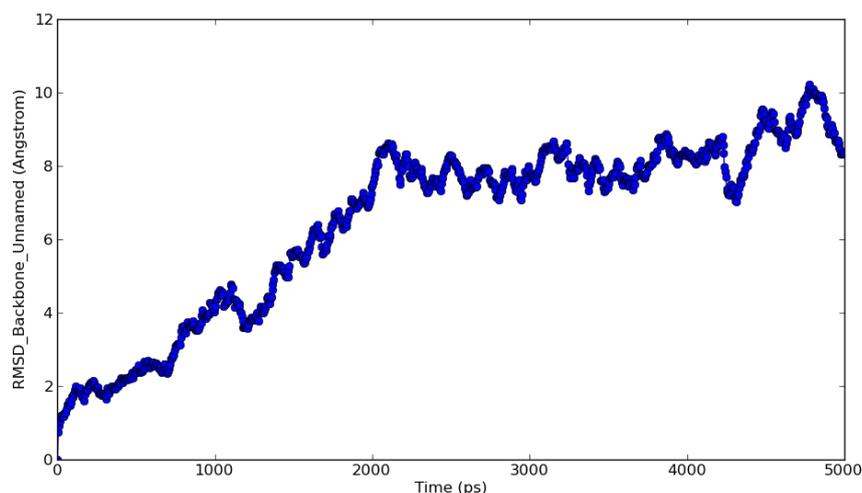


Figure S9: Time dependence of the total energy (**A**) and protein backbone RMSD (**B**) relative to the initial minimised complex of **9a** during MD simulations.

2. References

- 1) Tucker, B., and Lardelli, M. (2007) A rapid apoptosis assay measuring relative acridine orange fluorescence in zebrafish embryos. *Zebrafish*. 4, 113-116.
- 2) Mimasu, S., Sengoku, T., Fukuzawa, S., Umehara, T., and Yokoyama, S. (2008) Crystal structure of histone demethylase LSD1 and tranylcypramine at 2.25 Å. *Biochem Biophys Res Commun*. 366, 15-22.
- 3) Desmond Molecular Dynamics System, version 3.0, D. E. Shaw Research, New York, NY 2011. Maestro-Desmond Interoperability Tools, version 3.0, Schrödinger, New York, NY 2011