Electronic Supplementary Information

Pd-catalyzed isocyanide insertion / nucleophilic attack by indole C-3 / desulfonylation in the same pot: A direct access to indoloquinolines of pharmacological interest

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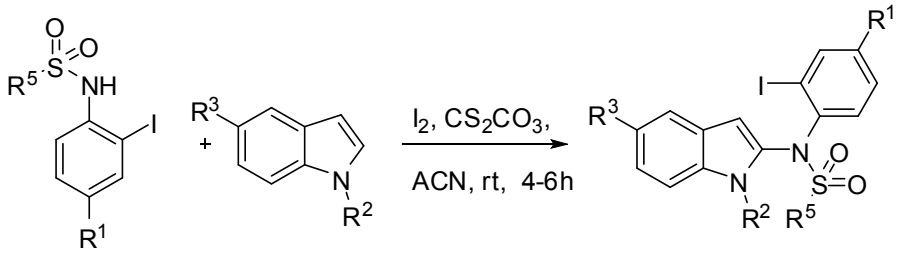
Experimental Section

Chemistry

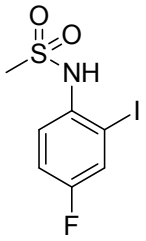
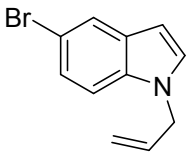
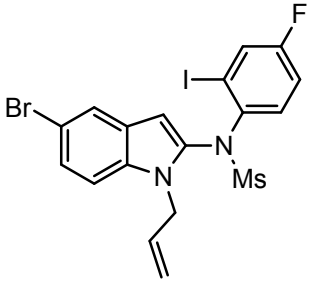
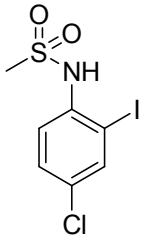
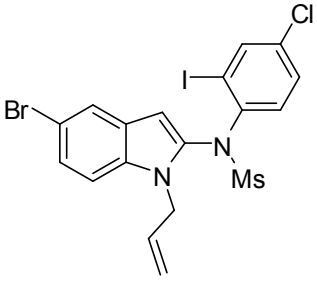
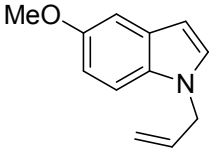
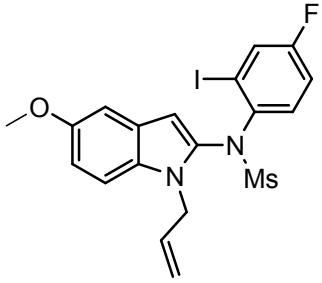
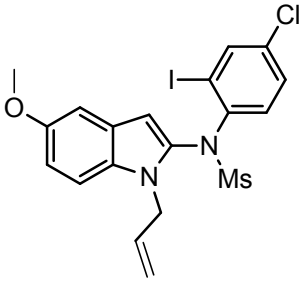
General methods: Unless stated otherwise, reactions were performed under nitrogen atmosphere using oven dried glassware. Reactions were monitored by thin layer chromatography (TLC) on silica gel plates (60 F254), visualizing with ultraviolet light or iodine spray. Flash chromatography was performed on silica gel (230-400 mesh) using distilled hexane, ethyl acetate. ^{1}H and ^{13}C NMR spectra were recorded in $CDCl_3$ solution by using a 400 MHz spectrometer. Proton chemical shifts (δ) are relative to tetramethylsilane (TMS, δ = 0.00) as internal standard and expressed in ppm. Spin multiplicities are given as s (singlet), d (doublet), dd (doublet of doublet), td (triplet of doublet), t (triplet) and m (multiplet) as well as bs (broad singlet). Coupling constants (*J*) are given in hertz. Infrared Melting points were determined using melting point apparatus and are uncorrected. MS spectra were obtained on aAgilent 6430 series Triple Quard LC-MS / MS spectrometer.

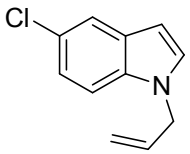
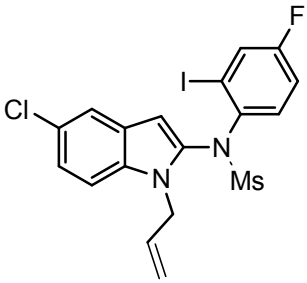
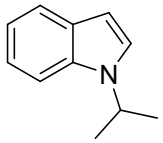
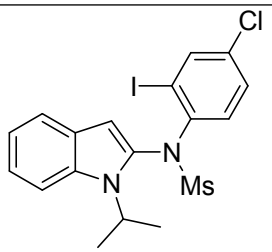
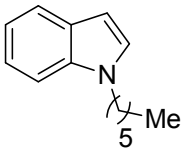
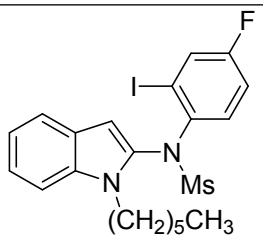
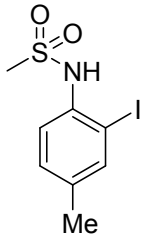
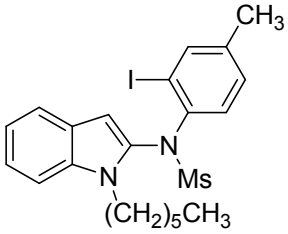
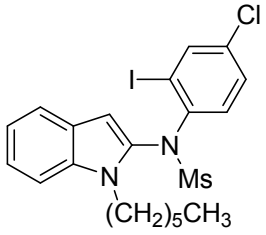
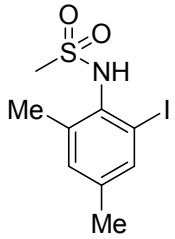
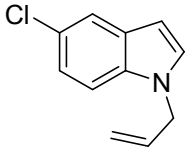
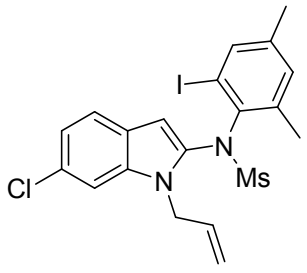
General procedure for the preparation of N-(1-substituted-5-substituted-1H-indol-2-yl)-N-(4-substituted-2-iodophenyl)methane-sulfonamide (1):

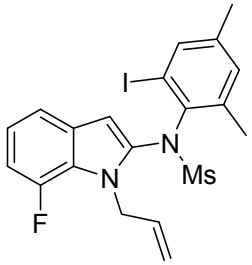
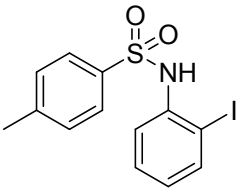
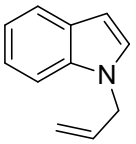
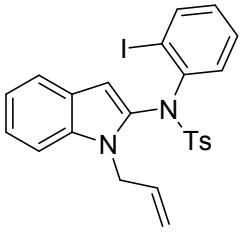


To a mixture of *N*-(2-iodophenyl)methane/4-methylbenzene/thiophene-2-sulfonamide derivative **S1** (1.0 mmol)**,** Cs_2CO_3 (1.5mmol), I_2 (1mmol) in acetonitrile (2.5 mL) was added indole derivative **S2** (1.2 mmol). Then the mixture was stirred at room temperature under nitrogen for 4-6 h. The progress of the reaction was monitored by TLC. Upon completion, the reaction was quenched with a saturation solution of $Na_2S_2O_3$ (5 mL) and extracted with ethyl acetate (3 × 30 mL). The combined organic phases were washed with brine (50 mL), dried over anhydrous sodium sulfate, filtered, and concentrated under a reduced pressure. The residue was purified by

column chromatography over silica gel using ethyl acetate–hexane to give the desired product (**1**).

S.No	Compound (**1**)	Compound (**2**)	Product (**3**)	Yield (%)
1	 **S1a**	 **S2a**	 **1a**	62
2	 **S1b**	**S2a**	 **1b**	58
3	**S1a**	 **S2b**	 **1c**	61
4	**S1b**	**S2b**	 **1d**	65

5	**S1a**	 **S2c**	 **1e**	68
6	**S1b**	 **S2d**	 **1m**	58
6	**S1a**	 **S2e**	 **1n**	63
7	 **S1c**	**S2e**	 **1o**	52
8	**S1c**	**S2e**	 **1p**	55
9	 **S1d**	 **S2f**		60

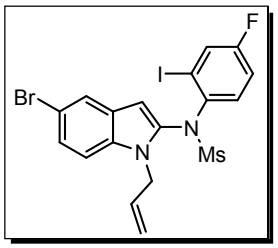
			1q	
10	**S1d**	 **S2g**	 **1r**	61
11	 **S1e**	 **S2h**	 **1t**	46

The compounds **1f**[1], **1h-1l**[1]**, 1s**[1] **and 1g**[2] were prepared according to the reported method.

References:

1. B. Prasad, B. Y. Sreenivas, D. Rambabu, G. R. Krishna, C. M. Reddy, K. L. Kumar and M. Pal, *Chem. Commun.* 2013, **49**, 3970.
2. B. Prasad, R. Adepu, A.K. Sharma and M. Pal, *Chem. Commun.* 2015, **51**, 1259.

***N*-(1-allyl-5-bromo-1*H*-indol-2-yl)-*N*-(4-fluoro-2-iodophenyl)methanesulfonamide (1a)**

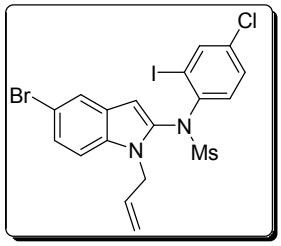


1a was prepared *via* the reaction of **S1a** with **S2a** according to the general procedure as mentioned above.

Bisque solid; yield: 62%; mp: 136.5-138.2 °C; R_f (10% EtOAc/*n*-hexane) 0.12; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.77 (d, *J* = 1.6 Hz, 1H), 7.68 (dd, *J* = 7.6, 2.8 Hz, 1H), 7.63 (dd, *J* = 8.8, 5.2 Hz, 1H), 7.34 (dd, *J* = 8.8, 1.6 Hz, 1H), 7.17-7.09 (m, 2H), 7.02 (s, 1H), 5.83-5.74 (m, 1H), 5.04 (m, 3H), 4.75 (d, *J* = 17.2 Hz, 1H), 3.27 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 162.5 (C-F *J* = 254.2 Hz), 137.6 (C-F *J* = 3.5Hz), 134.9, 133.6, 132.8, 131.8 (C-F *J* = 8.9 Hz), 128.0, 127.7 (C-

F J = 24.4 Hz), 126.1, 123.5, 116.5, 116.3, 113.7, 112.3, 100.9 (C-F J = 5.4 Hz), 100.7, 46.3, 39.5; MS (ES mass): m/z 549.1 (M+1).

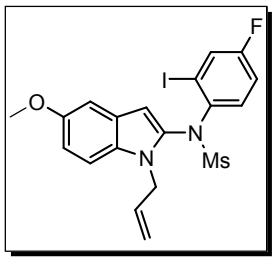
***N*-(1-allyl-5-bromo-1*H*-indol-2-yl)-*N*-(4-chloro-2-iodophenyl)methanesulfonamide (1b)**



1b was prepared *via* the reaction of **S1b** with **S2a** according to the general procedure as mentioned above.

Brown semi-solid; yield: 58%; R_f (10% EtOAc/*n*-hexane) 0.17; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.94 (s, 1H), 7.76 (s, 1H), 7.54 (d, J = 8.8 Hz, 1H), 7.34 (dd, J = 10.8, 9.2 Hz, 2H), 7.14 (d, J = 8.8 Hz, 1H), 7.01 (s, 1H), 5.80-5.73 (m, 1H), 5.04-5.00 (m, 3H), 4.73 (d, J = 17.2 Hz, 1H), 3.26 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 140.3, 140.0, 135.2, 134.6, 133.7, 132.7, 131.3, 129.5, 127.4, 126.1, 124.8, 123.5, 116.6, 113.8, 112.3, 101.0, 46.2, 39.6; MS (ES mass): m/z 567.2 (M+1).

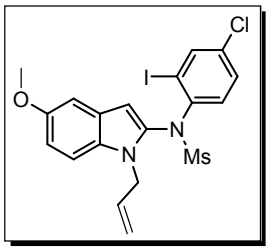
***N*-(1-allyl-5-methoxy-1*H*-indol-2-yl)-*N*-(4-fluoro-2-iodophenyl)methanesulfonamide (1c)**



1c was prepared *via* the reaction of **S1a** with **S2b** according to the general procedure as mentioned above.

Reddish brown semi-solid; yield: 61%; R_f (10% EtOAc/*n*-hexane) 0.09; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.65-7.49 (m, 2H), 7.50 (d, J = 8.4 Hz, 1H), 7.08-7.05 (m, 1H), 7.00 (s, 1H), 6.82 (dd, J = 8.4, 1.2 Hz, 1H), 6.70 (s, 1H), 5.85-5.75 (m, 1H), 5.06-4.98 (m, 3H), 4.77 (d, J = 17.2 Hz, 1H), 3.83 (s, 3H), 3.25 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 162.4 (C-F J = 253.8 Hz), 157.1, 138.0 (C-F J = 3.5 Hz), 135.9, 133.1, 132.7, 131.7, 131.6, 127.8 (C-F J = 24.5 Hz), 121.9, 120.0, 116.4, 116.2 (C-F J = 8.9 Hz), 110.7, 101.6, 93.9, 55.6, 46.0, 39.3; MS (ES mass): m/z 501.1 (M+1).

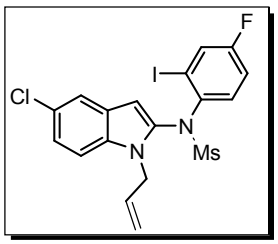
***N*-(1-allyl-5-methoxy-1*H*-indol-2-yl)-*N*-(4-chloro-2-iodophenyl)methanesulfonamide (1d)**



1d was prepared *via* the reaction of **S1b** with **S2b** according to the general procedure as mentioned above.

Reddish pink semi-solid; yield: 65%; R_f (10% EtOAc/*n*-hexane) 0.10; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.94 (d, *J* = 2.4 Hz, 1H), 7.55 (dd, *J* = 12.4, 8.4 Hz, 2H), 7.35 (dd, *J* = 8.8, 2.4 Hz, 1H), 7.01 (s, 1H), 6.83 (dd, *J* = 8.8, 2.4 Hz, 1H), 6.71 (s, 1H), 5.85-5.78 (m, 1H), 5.05 (d, *J* = 10.8 Hz, 1H), 5.02-4.98 (m, 2H), 4.78 (d, *J* = 17.2 Hz, 1H), 3.85 (s, 3H), 3.27 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 157.2 ,140.4, 140.2, 135.9, 134.9, 133.1, 132.4, 131.4, 129.4, 121.9, 120.0, 116.3, 110.7, 101.8, 101.1, 93.9, 55.6, 46.0, 39.4; MS (ES mass): *m*/*z* 517.1 (M+1).

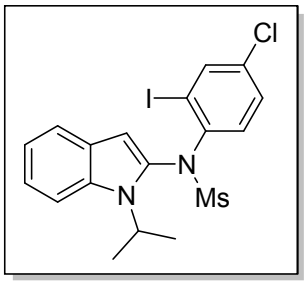
***N*-(1-allyl-5-chloro-1*H*-indol-2-yl)-*N*-(4-fluoro-2-iodophenyl)methanesulfonamide (1e)**



1e was prepared *via* the reaction of **S1a** with **S2c** according to the general procedure as mentioned above.

beige solid; yield: 68%; mp: 126.2-128.5 °C; R_f (10% EtOAc/*n*-hexane) 0.12; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.66 (dd, *J* = 7.6, 2.8 Hz, 1H), 7.61 (m, 2H), 7.19 (s, 2H), 7.15-7.10 (m,1H), 7.01 (s, 1H), 5.82-5.73 (m, 1H), 5.04-5.01 (m, 3H), 4.74 (d, *J* = 17.2 Hz, 1H), 3.26 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 162.5 (C-F *J* = 254 Hz), 137.6 (C-F *J* = 3.6Hz), 135.0, 133.4, 132.9, 131.8 (C-F *J* = 8.9 Hz), 127.9 (C-F *J* = 24.6 Hz), 126.8, 126.2, 123.5, 120.4, 116.5, 116.3, 111.9, 101.0, 100.8 (C-F *J* = 8.4 Hz), 46.3 ,39.5; MS (ES mass): *m*/*z* 505.1 (M+1).

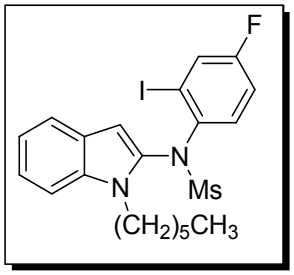
***N*-(4-chloro-2-iodophenyl)-*N*-(1-isopropyl-1*H*-indol-2-yl)methanesulfonamide (1m)**



1m was prepared *via* the reaction of **S1b** with **S2d** according to the general procedure as mentioned above.

Yellow solid; yield: 58%; mp: 104.5-106.5 °C; R_f (15% EtOAc-*n*-Hexane) 0.41; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.98 (d, *J* = 2.4 Hz, 1H), 7.65-7.62 (m, 2H), 7.58 (d, *J* = 8.4 Hz, 1H), 7.38 (dd, *J* = 8.8, 2.4 Hz, 1H), 7.27-7.22 (m, 1H), 7.14 (t, *J* = 8.0 Hz, 1H), 7.03 (s, 1H), 5.50-5.43 (m, 1H), 3.29 (s, 3H), 1.58 (d, *J* = 7.2 Hz, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 140.8, 140.5, 135.1, 133.3, 132.9, 131.3, 129.5, 126.6, 122.6, 121.4, 119.9, 112.5, 101.2, 100.8, 46.9, 39.0, 30.9, 21.1; MS (ES mass): *m*/*z* 489.2 (M+1).

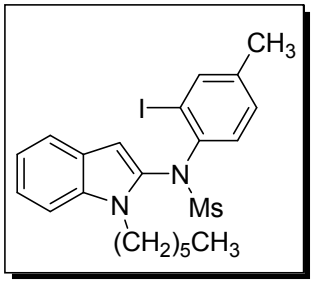
***N*-(4-fluoro-2-iodophenyl)-*N*-(1-hexyl-1*H*-indol-2-yl)methanesulfonamide (1n)**



1n was prepared *via* the reaction of **S1a** with **S2e** according to the general procedure as mentioned above.

Yellow solid; yield: 63%; mp: 96.5 to 97.5 °C; R_f (15% EtOAc-*n*-Hexane) 0.46; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.70 (dd, *J* = 7.6, 2.8 Hz, 1H), 7.66-7.60 (m, 2H), 7.31-7.25 (m, 2H), 7.17-7.12 (m, 1H), 7.11-7.08 (m, 1H), 7.07 (s, 1H), 4.28 (t, *J* = 8.0 Hz, 2H), 3.25 (s, 3H), 1.53-1.48 (m, 2H), 1.41-1.33 (m, 2H), 1.27-1.26 (m, 4H), 0.89-0.86 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 162.4, (C-F *J* = 253.8 Hz), 137.7 (C-F *J* = 3 Hz), 134.6, 133.7, 131.6 (C-F *J* = 5.3 Hz), 127.9 (C-F *J* = 22.6 Hz), 125.9, 122.9, 121.2, 120.2, 116.4 (C-F *J* = 20.2 Hz), 110.1, 101.2, 100.8 (C-F *J* = 8.5 Hz), 43.6, 39.3, 31.5, 29.9, 26.6, 22.5, 13.9; MS (ES mass): *m*/*z* 515.2 (M+1).

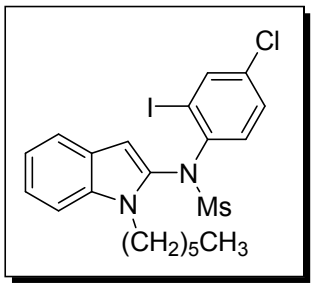
***N*-(1-hexyl-1*H*-indol-2-yl)-*N*-(2-iodo-4-methylphenyl)methanesulfonamide (1o)**



1o was prepared *via* the reaction of **S1c** with **S2e** according to the general procedure as mentioned above.

Semi solid; yield: 52%; R_f (15% EtOAc-*n*-Hexane) 0.42; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.80 (s, 1H), 7.63 (d, *J* = 7.6 Hz, 1H), 7.51 (d, *J* = 8.0 Hz, 1H), 7.28 (d, *J* = 8.0 Hz, 1H), 7.25-7.22 (m, 1H), 7.17-7.11 (m, 2H), 7.06 (s, 1H), 4.28 (t, *J* = 8.0 Hz, 2H), 3.25 (s, 3H), 2.30 (s, 3H), 1.52-1.45 (m, 2H), 1.39-1.33 (m, 2H), 1.27-1.23 (m, 4H), 0.89-0.85 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 141.5, 140.4, 138.8, 134.5, 134.0, 130.4, 130.1, 126.0, 122.7, 121.1, 120.1, 110.1, 101.1, 100.6, 43.7, 39.3, 31.5, 29.8, 26.6, 22.4, 20.4, 14.0; MS (ES mass): *m*/*z* 511.1 (M+1).

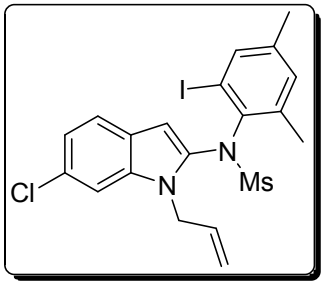
***N*-(4-chloro-2-iodophenyl)-*N*-(1-hexyl-1*H*-indol-2-yl)methanesulfonamide (1p)**



1p was prepared *via* the reaction of **S1b** with **S2e** according to the general procedure as mentioned above.

Semi solid; yield: 55%; R_f (15% EtOAc-*n*-Hexane) 0.42; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.98 (d, *J* = 2.4 Hz, 1H), 7.64 (d, *J* = 7.6 Hz, 1H), 7.56 (d, *J* = 8.4 Hz, 1H), 7.36-7.33 (m, 1H), 7.30-7.27 (m, 2H), 7.17-7.14 (m, 1H), 7.08 (s, 1H), 4.29-4.24 (t, *J* = 8.0 Hz, 2H), 3.26 (s, 3H), 1.51-1.46 (m, 2H), 1.39-1.35 (m, 2H), 1.27-1.25 (m, 4H), 0.90-0.87 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 140.3, 140.1, 134.9, 134.5, 133.5, 131.2, 129.5, 125.9, 123.0, 121.2, 120.3, 110.1, 101.3, 101.1, 43.6, 39.4, 31.5, 29.9, 26.6, 22.5, 14.0; MS (ES mass): *m*/*z* 531.5 (M+1).

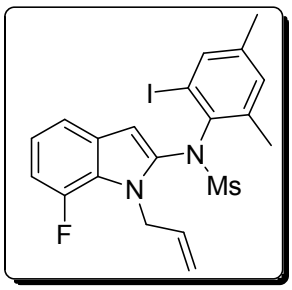
***N*-(1-allyl-5-bromo-1*H*-indol-2-yl)-*N*-(2-iodo-4,6-dimethylphenyl)methanesulfonamide (1q)**



1q was prepared *via* the reaction of **S1d** with **S2f** according to the general procedure as mentioned above.

white solid; yield: 60%; mp: 118.5-119.5 °C; R_f (10% EtOAc/*n*-hexane) 0.13; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.67 (s, 1H), 7.57 (d, *J* = 1.6 Hz, 1H), 7.14-7.10 (m, 2H), 7.05 (d, *J* = 8.4 Hz, 1H), 6.95 (s, 1H), 5.35-5.26 (m, 1H), 4.83-4.76 (m, 2H), 4.50 (dd, *J* = 13.2, 4.0 Hz, 2H), 3.38 (s, 3H), 2.30 (s, 3H), 2.25 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 141.2, 140.3, 139.4, 136.1, 135.4, 133.5, 131.8, 127.2, 126.1, 113.4, 122.4, 119.9, 115.3, 110.9, 109.9, 104.3, 98.2, 46.4, 39.5, 20.5, 20.2. MS (ES mass): *m*/*z* 515.0 (M+1).

_N_-(1-allyl-7-fluoro-1_H_-indol-2-yl)-_N_-(2-iodo-4,6-dimethylphenyl)methanesulfonamide (1r)

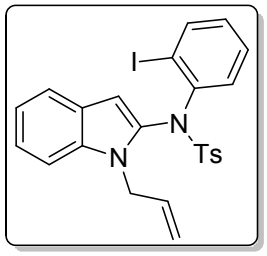


1r was prepared *via* the reaction of **S1d** with **S2g** according to the general procedure as mentioned above.

White solid; yield: 61%; mp: 107.0-108.2 °C R_f (10% EtOAc/*n*-hexane) 0.10; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.68 (s, 1H), 7.36 (d, *J* = 7.6 Hz, 1H), 7.23 (d, *J* = 2.4 Hz, 1H), 7.02 (dt, *J* = 8.0, 4.8 Hz, 1H), 6.96 (s, 1H), 6.85 (dd, *J* = 16, 7.6 Hz, 1H), 5.48-5.39 (m, 1H), 4.91-4.84 (m, 1H), 4.76-4.69 (m, 2H), 4.40 (d, *J* = 15.6 Hz, 1H), 3.37 (s, 3H), 2.32 (s, 3H), 2.26 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 150.8 (C-F *J* = 212 Hz), 148.2, 141.2, 140.2, 139.5, 136.1, 135.2, 133.5 (C-F *J* = 5.8 Hz), 129.9 (C-F *J* = 5.3 Hz), 123.0, 120.5 (C-F *J* = 6.7 Hz), 116.4 (C-F *J* =

3.5 Hz), 114.2, 108.2 (C-F J = 18.6 Hz), 104.3, 100.4 (C-F J = 1.7 Hz), 47.9, 39.5, 20.6, 20.2. MS (ES mass): m/z 499.8 (M+1).

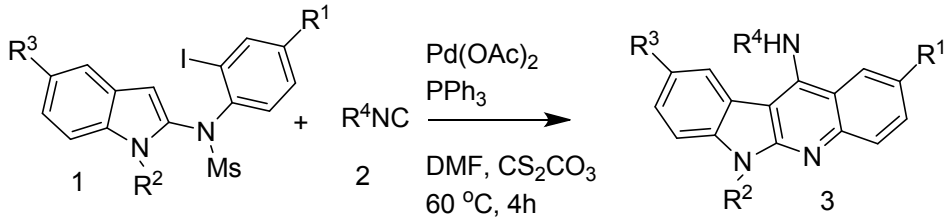
N-(1-allyl-1H-indol-2-yl)-N-(2-iodophenyl)-4-methylbenzenesulfonamide (1s)



1s was prepared *via* the reaction of **S1e** with **S2h** according to the general procedure as mentioned above.

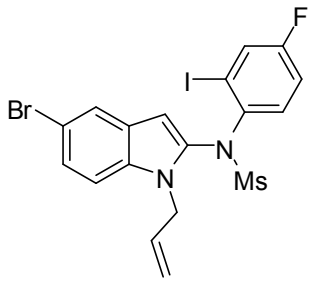
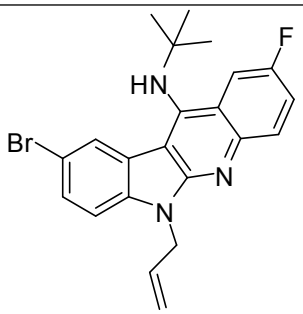
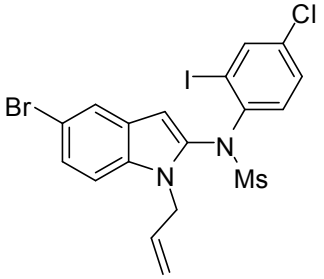
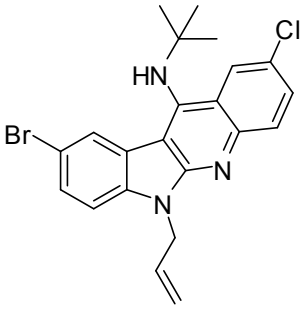
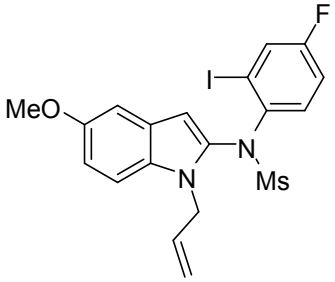
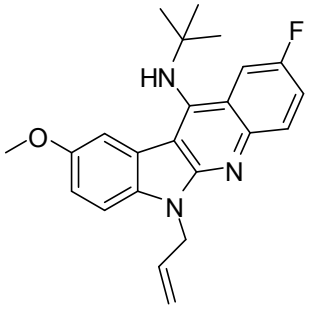
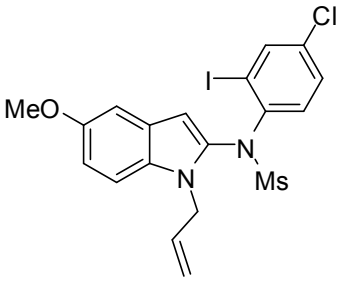
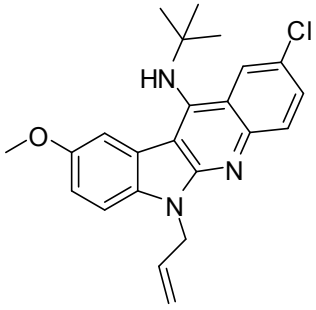
White solid; yield: 46%; mp: 169.5-171.5 °C; R_f (10% EtOAc/n-hexane) 0.31; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 7.96 (d, J = 7.6 Hz, 1H), 7.60 (d, J = 8.0 Hz, 2H), 7.52 (d, J = 7.6 Hz, 1H), 7.33-7.27 (m, 5H), 7.23-7.19 (m, 1H), 7.10 (t, J = 7.6 Hz, 1H), 7.03-6.98 (m, 1H), 6.32 (s, 1H), 5.93-5.83 (m, 1H), 5.21 (s, 2H), 5.06 (dd, J = 10.4, 1.2 Hz, 1H), 4.91 (dd, J = 17.6, 1.2 Hz, 1H), 2.47 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 144.7, 142.7, 141.1, 134.8, 134.7, 134.2, 134.1, 130.4, 129.9, 129.5 (2C), 129.3 (2C), 128.8, 125.8, 122.7, 120.9, 120.1, 116.3, 111.1, 101.7, 101.0, 46.9, 21.7; MS (ES mass): m/z 529.1 (M+1).

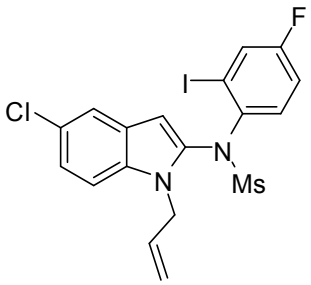
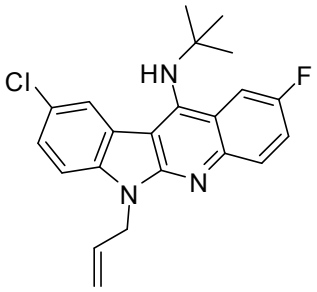
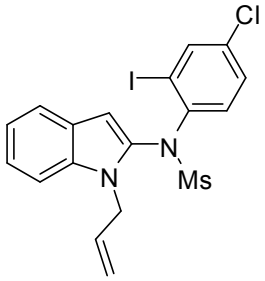
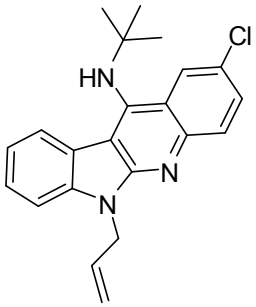
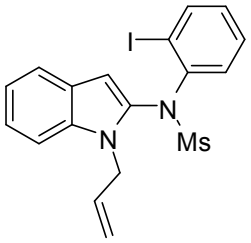
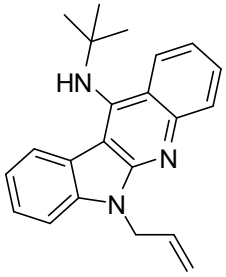
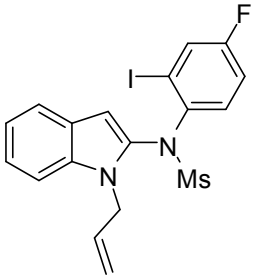
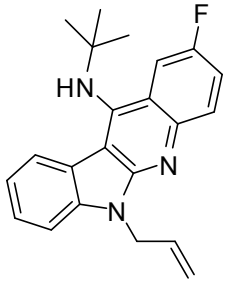
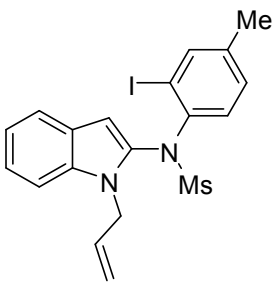
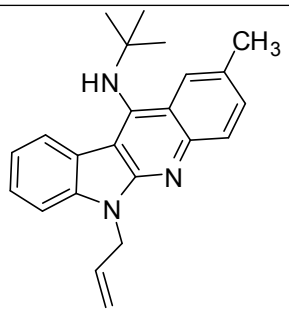
General Procedure for the preparation of indolo[2,3-*b*]quinolin-11-amines (3)

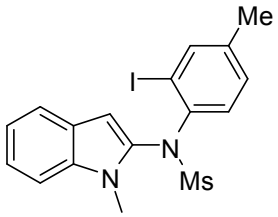
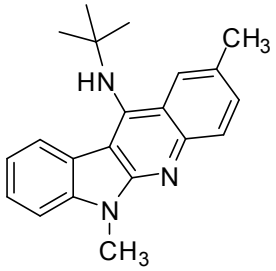
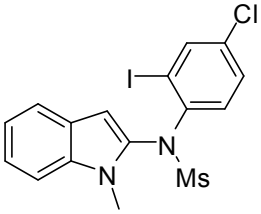
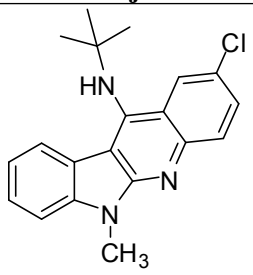
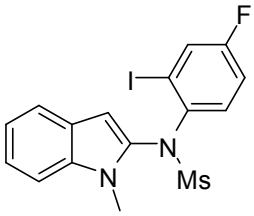
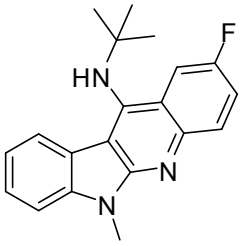
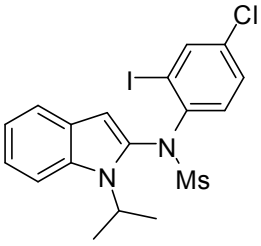
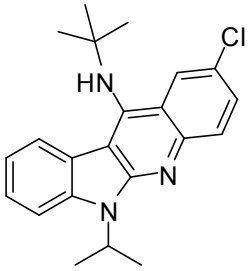
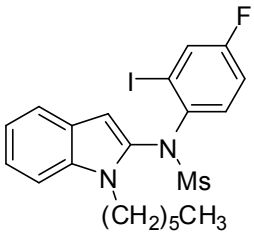
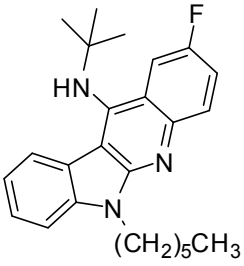


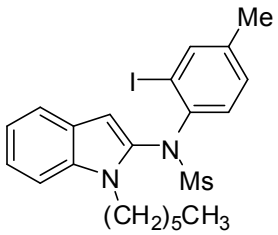
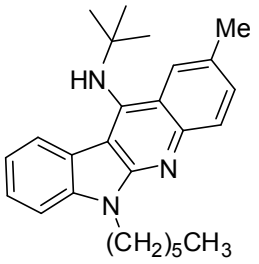
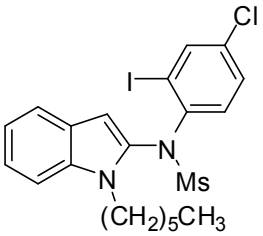
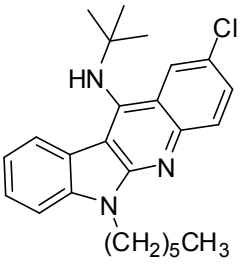
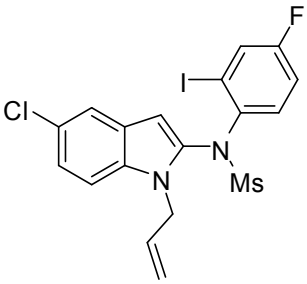
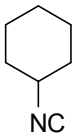
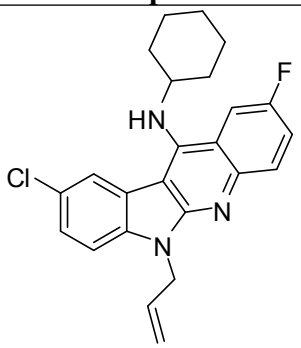
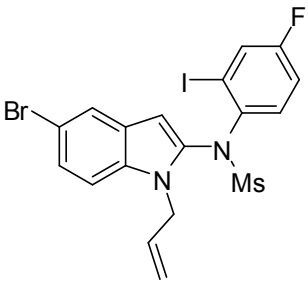
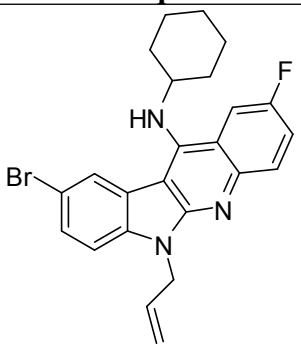
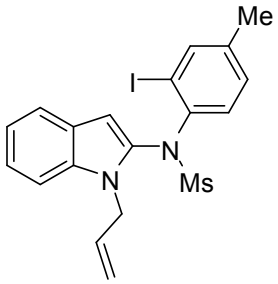
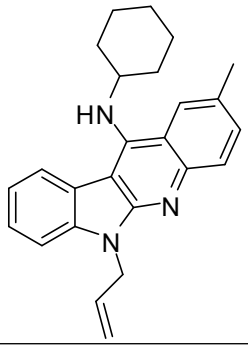
To mixture of N-(4-substituted-2-iodophenyl)-N-(1-alkyl-1H-indol-2-yl)methane-sulfonamide (**1**) (0.3 mmol), $Pd_2(dba)_3$ (5 mol%), PPh_3 (11 mol%) and Cs_2CO_3 in anhydrous DMF (3 mL) was added slowly isocyanide (0.9 mmol) for 10 minutes then stirred at 60 °C under nitrogen atmosphere for 4 h. The progress of the reaction was monitored by TLC. Upon completion, the mixture was cooled to room temperature, and filtered to remove the solid materials. The filtrate was extracted with ethyl acetate (3 x 15 mL). The organic layers were collected, combined, dried over anhydrous Na_2SO_4, filtered and concentrated under a reduced pressure. The residue was

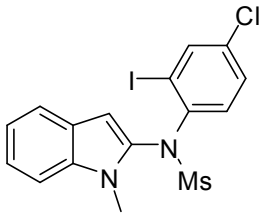
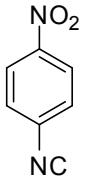
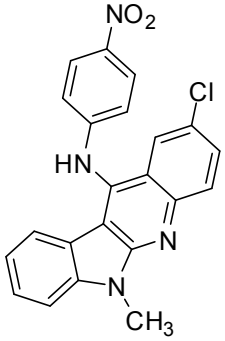
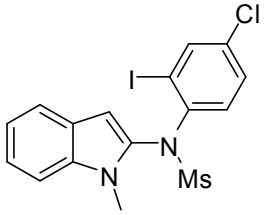
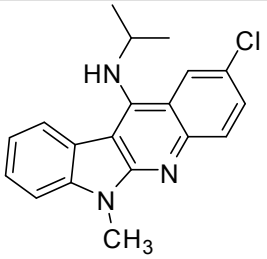
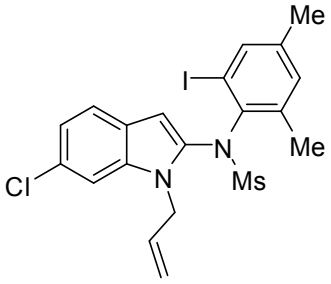
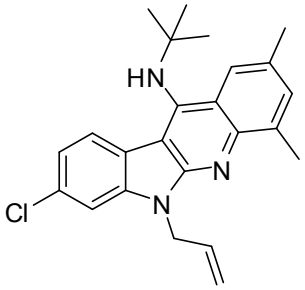
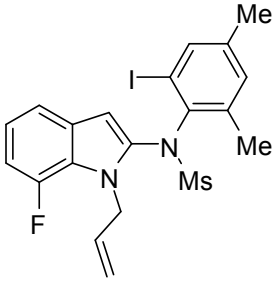
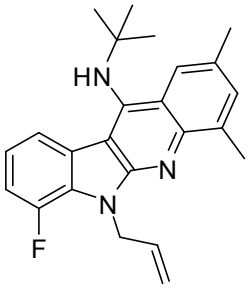
purified by column chromatography over silica gel using ethyl acetate–hexane to give the desired product **3**.

Entry	Compound (**1**)	Compound (**2**)	Product (**3**)	Yield (%)
1	 **1a**	 **2a**	 **3a**	65
2	 **1b**	**2a**	 **3b**	68
3	 **1c**	**2a**	 **3c**	60
4	 **1d**	**2a**	 **3d**	65

5	 **1e**	**2a**	 **3e**	62
6	 **1f**	**2a**	 **3f**	60
7	 **1g**	**2a**	 **3g**	64
8	 **1h**	**2a**	 **3h**	65
9	 **1i**	**2a**	 **3i**	67

10	 **1j**	**2a**	 **3j**	64
11	 **1k**	**2a**	 **3k**	65
12	 **1l**	**2a**	 **3l**	65
13	 **1m**	**2a**	 **3m**	62
14	 **1n**	**2a**	 **3n**	58

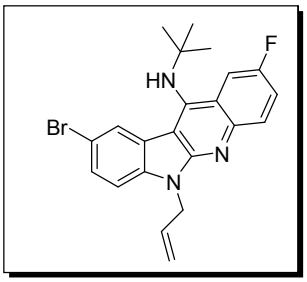
15	 **1o**	**2a**	 **3o**	66
16	 **1p**	**2a**	 **3p**	63
17	 **1e**	 **2b**	 **3q**	68
18	 **1a**	**2b**	 **3r**	68
19	 **1i**	**2b**		47

			3s	
20	 **1k**	 **2c**	 **3t**	26
23	 **1k**	 **2d**	 **3u**	57
21	 **1q**	**2a**	 **4a**	60
22	 **1r**	**2a**	 **4b**	58

[a]All the reactions were carried out using **1** (0.3 mmol), **2** (0.9 mmol), $Pd(OAc)_2$ (5 mol%), PPh_3 (11 mol%) and Cs_2CO_3 (0.9 mmol) in DMF (3 mL) at 60 °C for 4 h under nitrogen. [b]Isolated yield.

Analytical data of products:

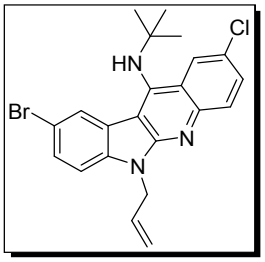
6-allyl-9-bromo-*N*-tert-butyl-2-fluoro-6*H*-indolo[2,3-*b*]quinolin-11-amine (3a)



3a was prepared *via* the reaction of **1a** with **2a** according to the general procedure as mentioned above.

Orange solid; yield: 65%; mp: 130.2-131.5 °C; R_f (10% EtOAc/*n*-hexane) 0.44; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.52 (s, 1H), 8.04-7.96 (m, 2H), 7.58 (d, *J* = 7.6 Hz, 1H), 7.45 (td, *J* = 8.4, 2.8 Hz, 1H), 7.25 (d, *J* = 5.6 Hz, 1H), 6.09-5.99 (m, 1H), 5.26 (d, *J* = 29.6 Hz, 1H), 5.17 (d, *J* = 20.4 Hz, 1H), 5.18-5.10 (m, 2H), 3.90-3.89 (m, 1H), 1.37 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 159.1 (C-F *J* = 240.3 Hz), 152.6, 147.2 (C-F *J* = 5.1Hz), 145.0, 140.2, 132.3, 129.7 (C-F *J* = 6.8 Hz), 129.6, 125.7, 124.2 (C-F *J* = 8.4 Hz), 122.4, 119.0 (C-F *J* = 25.6Hz), 117.0, 112.6, 112.1, 110.6, 108.7 (C-F *J* = 23.3 Hz), 58.1, 43.7, 31.4 (3C); ^{19}F NMR (376 MHz, $CDCl_3$) δ -119.3 (m); HPLC: 99.21%, column: X Bridge C-18 75*4.6 mm 3.5µ, mobile phase A: 0.1 % TFA in water mobile phase B: CH_3CN (gradient) T/B% : 0/20, 3/20, 8/40, 15/95, 20/95, 25/20,30/20; flow rate: 1 mL/min; UV 290 nm, retention time 4.40 min; MS (ES mass): *m*/*z* 428.2 (M+1).

6-allyl-9-bromo-*N*-tert-butyl-2-chloro-6*H*-indolo[2,3-*b*]quinolin-11-amine (3b)

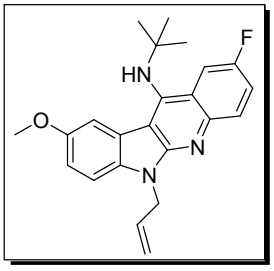


3b was prepared *via* the reaction of **1b** with **2a** according to the general procedure as mentioned above.

Yellow solid; yield: 68%; mp: 167-168.8 °C; R_f (10% EtOAc/n-hexane) 0.46; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.49 (s, 1H), 8.33 (d, *J* = 2.4 Hz, 1H), 7.98 (d, *J* = 8.8 Hz, 1H), 7.60 (dd, *J* = 8.8, 1.6 Hz, 2H), 7.27 (m, 1H), 6.05 (m, 1H), 5.22 (d, *J* = 10.8 Hz, 1H), 5.17-5.10 (m, 3H), 3.94

(s, 1H), 1.37 (s, 9H); ^{13}C NMR (100 MHz, CDCl3) δ: 153.0, 147.0, 146.4, 140.1, 132.0, 129.6, 129.6, 129.1, 127.3, 125.6, 124.4, 124.2, 122.5, 117.0, 112.4, 112.2, 110.7, 58.1, 43.7, 31.5(3C) ; HPLC: 99.21%, column: X Bridge C-18 75*4.6 mm 3.5µ, mobile phase A: 0.1 % TFA in water mobile phase B: CH3CN (gradient) T/B% : 0/20, 2/20, 10/95, 20/95, 22/20, 25/20; flow rate: 1 mL/min; UV 290 nm, retention time 4.52 min; MS (ES mass): m/z 444.1 (M+3).

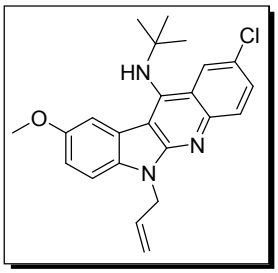
6-allyl-*N*-tert-butyl-2-fluoro-9-methoxy-6*H*-indolo[2,3-*b*]quinolin-11-amine (3c)



3c was prepared *via* the reaction of **1c** with **2a** according to the general procedure as mentioned above.

White solid; yield: 60%; mp: 116.2-117.9°C; R_f (10% EtOAc/*n*-hexane) 0.71; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.25 (d, *J* = 8.4 Hz, 1H), 8.04-8.00 (m, 2H), 7.44-7.40 (m, 1H), 6.88 (d, *J* = 7.6 Hz, 2H), 6.13-6.03 (m, 1H), 5.23 (d, *J* = 10.0 Hz, 1H), 5.18 (d, *J* = 17.6 Hz, 1H), 5.12 (d, *J* = 4.4 Hz, 2H), 3.95 (s, 3H), 3.91 (s, 1H), 1.37 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 159.7, 159.0 (C-F, *J* = 239 Hz), 153.2, 144.9 (C-F, *J* = 4.5 Hz), 143.9, 143.4, 132.6, 129.3 (C-F, *J* = 8.3 Hz), 124.6 (C-F, *J* = 8.3 Hz), 123.8, 118.0 (C-F, *J* = 25.8 Hz), 116.8, 114.0, 113.8, 108.8 (C-F, *J* = 23.3 Hz), 106.8, 94.6, 57.7, 55.5, 43.6, 31.4 (3C); ^{19}F NMR (376 MHz, $CDCl_3$) δ -119.9 (m); HPLC: 96.2%, column: X Bridge C-18 75*4.6 mm 3.5µ, mobile phase A: 0.1 % TFA in water mobile phase B: CH_3CN (gradient) T/B% : 0/20, 0.5/20, 2/95, 8/95, 10/20, 12/20; flow rate: 1 mL/min; UV 280 nm, retention time 3.68 min; MS (ES mass): *m*/*z* 378.3 (M+1).

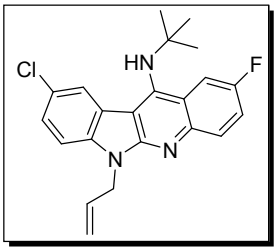
6-allyl-*N*-tert-butyl-2-chloro-9-methoxy-6*H*-indolo[2,3-*b*]quinolin-11-amine (3d)



3d was prepared *via* the reaction of **1d** with **2a** according to the general procedure as mentioned above.

Orange solid; yield: 65%; mp: 148.5-150.2 °C; R_f (10% EtOAc/*n*-hexane) 0.75; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.35 (d, *J* = 2 Hz, 1H), 8.21 (d, *J* = 8.8 Hz, 1H), 7.96 (d, *J* = 8.8 Hz, 1H), 7.55 (dd, *J* = 8.8, 2.4 Hz, 1H), 6.87 (m, 2H), 6.11-6.01 (m, 1H), 5.22 (d, *J* = 10.4 Hz, 1H), 5.13 (m, 3H), 3.93 (s, 3H), 3.90 (s, 1H), 1.35 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 159.7, 153.6, 145.4, 144.7, 143.3, 132.5, 128.9, 128.6, 126.9, 124.8, 124.3, 123.6, 116.9, 114.1, 113.6, 106.9, 94.8, 57.6, 55.6, 43.6, 31.4 (3C); HPLC: 99.08%, column: X Bridge C-18 75*4.6 mm 3.5µ, mobile phase A: 0.1 % TFA in water mobile phase B: CH_3CN (gradient) T/B% : 0/20, 2/20, 10/95, 20/95, 22/20, 25/20; flow rate: 1 mL/min; UV 290 nm, retention time 3.59 min; MS (ES mass): *m*/*z* 394.3 (M+1).

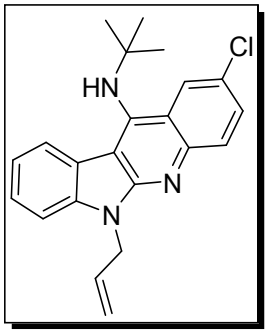
6-allyl-*N*-tert-butyl-2-chloro-9-fluoro-6*H*-indolo[2,3-*b*]quinolin-11-amine (3e)



3e was prepared *via* the reaction of **1e** with **2a** according to the general procedure as mentioned above.

Yellow solid; yield: 62%; mp: 110-111.2 °C; R_f (10% EtOAc/*n*-hexane) 0.43; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.35 (s, 1H), 8.04-7.96 (m, 2H), 7.46 (d, *J* = 8.0 Hz, 2H), 7.29 (d, *J* = 8.8 Hz, 1H), 6.09-6.00 (m, 1H), 5.21 (d, *J* = 10.4 Hz, 1H), 5.15-5.01 (m, 3H), 3.88 (s, 1H), 1.37 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 159.0 (C-F *J* = 239.9 Hz), 152.8, 147.1(C-F *J* = 5.2 Hz), 145.0, 139.8, 132.4, 129.6 (C-F *J* = 8.5 Hz), 126.9, 124.8, 124.2 (C-F *J* = 8 Hz), 122.7, 121.8, 119.0 (C-F *J* = 25.3 Hz), 117.0, 112.8, 112.7, 110.1, 108.7 (C-F *J* = 23.3 Hz), 58.0, 43.7, 31.4 (3C); ^{19}F NMR (376 MHz, $CDCl_3$) δ -119.4 (m); HPLC: 96.81%, column: X Bridge C-18 75*4.6 mm 3.5µ, mobile phase A: 0.1 % TFA in water mobile phase B: CH_3CN (gradient) T/B% : 0/20, 2/20, 10/95, 20/95, 22/20, 25/20; flow rate: 1 mL/min; UV 275 nm, retention time 3.86 min; MS (ES mass): *m*/*z* 382.2 (M+1).

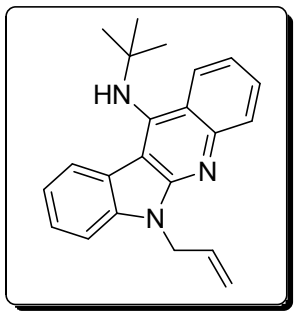
6-allyl-*N*-tert-butyl-2-chloro-6*H*-indolo[2,3-*b*]quinolin-11-amine (3f)



3f was prepared *via* the reaction of **1f** with **2a** according to the general procedure as mentioned above.

Beige solid; yield: 60%; mp: 107-108.4 °C; R_f (15% EtOAc-*n*-Hexane) 0.64; ^{1}H NMR (400 MHz, $CDCl_3$): 8.39 (d, *J* = 2.4 Hz, 1H), 8.35 (d, *J* = 7.6 Hz, 1H), 7.98 (d, *J* = 8.8 Hz, 1H), 7.58 (dd, *J* = 9.2, 2.8 Hz, 1H), 7.55-7.49 (m, 1H), 7.40 (d, *J* = 8.0 Hz, 1H), 7.30-7.27 (m, 1H), 6.11-6.04 (m, 1H), 5.21 (dd, *J* = 10.4, 1.2 Hz, 1H), 5.18-5.14 (m, 3H), 4.10 (s, 1H), 1.38 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 153.1, 146.6, 146.2, 141.5, 132.6, 129.2, 128.9, 127.1, 126.8, 124.6, 124.4, 122.6, 120.6, 119.7, 116.9, 113.1, 109.5, 57.8, 43.7, 31.5 (3C); HPLC: 98.6%, column: Symmetry C-18 75*4.6 mm, 3.5µm, mobile phase A: 0.1 % Triflouro acetic acid in water mobile phase B: CH_3CN (gradient) T/B%: 0/20, 1/20, 6/98, 10/98, 12/20, 15/20; flow rate: 1.0 mL/min; UV 275 nm retention time 6.25 min; MS (ES mass): *m*/*z* 364.0 (M+1).

6-allyl-*N*-tert-butyl-2-6*H*-indolo[2,3-*b*]quinolin-11-amine (3g)

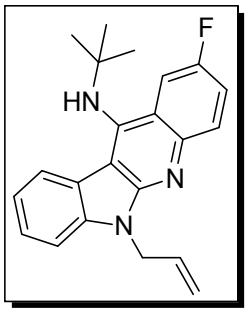


3g was prepared *via* the reaction of **1g** with **2a** according to the general procedure as mentioned above.

Yellow solid; yield: 64%; mp: 96.5-97.5 °C; R_f (15% EtOAc-*n*-Hexane) 0.61; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.40 (t, *J* = 7.6 Hz, 2H), 8.06 (d, *J* = 8.0 Hz, 1H), 7.67 (t, *J* = 7.6 Hz, 1H), 7.49 (t, *J* = 8.0 Hz, 1H), 7.40-7.36 (m, 2H), 7.30-7.26 (m, 1H), 6.14-6.04 (m, 1H), 5.22-5.15 (m, 4H), 4.15 (s, 1H), 1.39 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 159.7, 153.1, 147.5, 141.4, 132.8,

128.6, 127.4, 126.7, 125.4, 123.7, 122.6, 121.2, 121.1, 119.5, 116.7, 112.6, 109.3, 57.9, 43.6, 31.5 (3C); HPLC: 98.8%, column: X-BRIDGE C-18 150*4.6 mm, 5µ, mobile phase A: 5mM Ammonium Formate in water mobile phase B: CH_3CN (gradient) T/%B: 0/70, 2/70, 12/98, 20/98, 22/70, 25/70; flow rate: 1.0 mL/min; UV 280 nm retention time 7.72 min; MS (ES mass): *m*/*z* 330.0 (M+1).

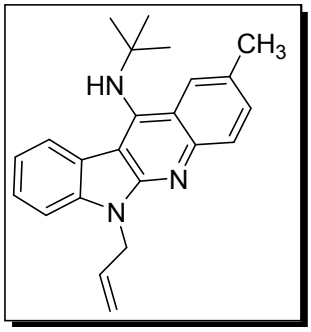
6-allyl-*N*-tert-butyl-2-fluoro-6*H*-indolo[2,3-*b*]quinolin-11-amine (3h)



3h was prepared *via* the reaction of **1h** with **2a** according to the general procedure as mentioned above.

Yellow solid; yield: 65%; mp: 153-154.1 °C; R_f (15% EtOAc-*n*-Hexane) 0.58; ^{1}H NMR (400 MHz, $CDCl_3$): 8.37 (d, *J* = 7.7 Hz, 1H), 8.06-8.02 (m, 2H), 7.54-7.50 (m, 1H), 7.47-7.42 (m, 1H), 7.40 (d, *J* = 8.0 Hz, 1H), 7.39 (t, *J* = 7.2 Hz, 1H), 6.13-6.05 (m, 1H), 5.23-5.19 (m, 2H), 5.16-5.15 (m, 2H), 4.06 (bs, 1H), 1.39 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 158.9 (C-F, *J* = 239.3Hz), 152.7, 146.8 (C-F, *J* = 5.0 Hz), 144.8, 141.6, 132.7, 129.4 (C-F, *J* = 8.5 Hz), 127.1, 124.2 (C-F, *J* = 8.4 Hz), 122.8, 120.5, 119.5, 118.6 (C-F, *J* = 25.5Hz), 116.8, 113.3, 109.3 (C-F *J* = 27.3Hz), 108.8, 57.8, 43.6, 31.5 (3C); HPLC: 98.9%, column: Symmetry C-18 75*4.6 mm, 3.5µm, mobile phase A: 0.1 % TFA in water mobile phase B: CH_3CN (gradient) T/%B: 0/20, 2/20, 9/98, 14/98, 16/20, 20/20; flow rate: 1.0 mL/min; UV 270 nm retention time 7.49 min; MS (ES mass): *m*/*z* 348.0 (M+1).

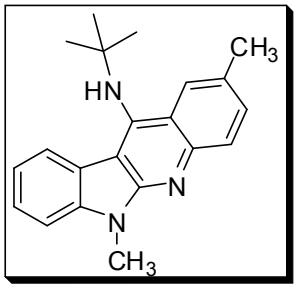
6-allyl-*N*-tert-butyl-2-methyl-6*H*-indolo[2,3-*b*]quinolin-11-amine (3i)



3i was prepared *via* the reaction of **1i** with **2a** according to the general procedure as mentioned above.

Beige solid; yield: 67%; mp: 131-132.5 °C; R_f (15% EtOAc-*n*-Hexane) 0.68; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.37 (d, *J* = 7.6 Hz, 1H), 8.16 (s, 1H), 7.95 (d, *J* = 8.8 Hz, 1H), 7.51-7.46 (m, 2H), 7.38 (d, *J* = 7.9 Hz, 1H), 7.33-7.28 (m, 1H), 6.13-6.03 (m, 1H), 5.21-5.14 (m, 4H), 4.09 (bs, 1H), 2.57 (s, 3H), 1.38 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 146.9, 146.4, 141.4, 132.9, 130.8, 130.6, 127.2, 126.5, 124.3, 123.6, 122.6, 121.1, 119.3, 116.7, 112.7, 111.6, 109.2, 57.7, 43.6, 31.5 (3C), 21.6; HPLC: 96.2%, column: Symmetry C-18 75*4.6 mm, 3.5µm, mobile phase A: 0.1 % Triflouro acetic acid in water mobile phase B: CH_3CN (gradient) T/%B: 0/20, 2/20, 9/98, 14/98, 16/20, 20/20; flow rate: 1.0 mL/min; UV 270 nm retention time 6.84 min; MS (ES mass): *m*/*z* 344.0 (M+1).

***N*-tert-butyl-6-isopropyl-2-methyl-6*H*-indolo[2,3-*b*]quinolin-11-amine (3j)**

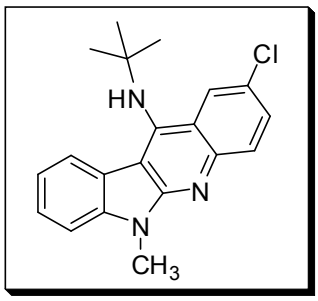


3j was prepared *via* the reaction of **1j** with **2a** according to the general procedure as mentioned above.

Yellow solid; yield: 64%; mp: 147-148.5 °C; R_f (15% EtOAc-*n*-Hexane) 0.65; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.37 (d, *J* = 7.6 Hz, 1H), 8.16 (s, 1H), 7.97 (d, *J* = 8.4 Hz, 1H), 7.54-7.50 (m, 2H), 7.39 (d, *J* = 8.0 Hz, 1H), 7.28 (d, *J* = 7.6 Hz, 1H), 4.09 (bs, 1H), 3.97 (s, 3H), 2.57 (s, 3H), 1.38 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 147.0, 143.9, 142.0, 130.9, 130.5, 127.0, 126.6, 124.4, 123.4, 122.6, 120.9, 119.2, 112.7, 110.3, 108.3, 57.7, 31.5 (3C), 29.5, 21.6; HPLC:

99.5%, column: Symmetry C-18 75*4.6 mm, 3.5µm, mobile phase A: 0.1 % Triflouro acetic acid in water mobile phase B: CH_3CN (gradient) T/%B: 0/20, 2/20, 9/98, 14/98, 16/20, 20/20; flow rate: 1.0 mL/min; UV 270 nm retention time 6.47 min; MS (ES mass): *m*/*z* 318.0 (M+1).

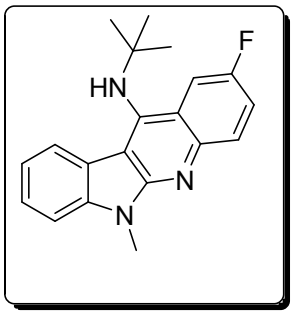
***N*-tert-butyl-2-chloro-6-methyl-6*H*-indolo[2,3-*b*]quinolin-11-amine (3k)**



3k was prepared *via* the reaction of **1k** with **2a** according to the general procedure as mentioned above.

Olive green solid; yield: 65%; mp: 118.5-119.5 °C; R_f (15% EtOAc-*n*-Hexane) 0.58; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.39 (d, *J* = 2.4 Hz, 1H), 8.33 (d, *J* = 8.0 Hz, 1H), 7.99 (d, *J* = 8.8 Hz, 1H), 7.61-7.53 (m, 2H), 7.41 (d, *J* = 8.0 Hz, 1H), 7.30 (t, *J* = 7.6 Hz, 1H), 4.07 (s, 1H), 3.96 (s, 3H), 1.37 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 153.6, 146.6, 142.1, 129.2, 128.7, 127.2, 124.7, 122.6, 121.3, 120.5, 119.3, 118.0, 113.2, 110.8, 108.6, 57.8, 31.5 (3C), 27.6; HPLC: 91.6%, column: Symmetry C-18 150*4.6 mm, 5µm, mobile phase A: 5mM Ammonium Formate in water mobile phase B: CH_3CN (gradient) T/%B: 0/70, 2/70, 12/98, 20/98, 22/70, 25/70; flow rate: 1.0 mL/min; UV 280 nm retention time 9.22 min; MS (ES mass): *m*/*z* 338.1 (M+1).

***N*-tert-butyl-2-fluoro-6-methyl-6*H*-indolo[2,3-*b*]quinolin-11-amine (3l)**

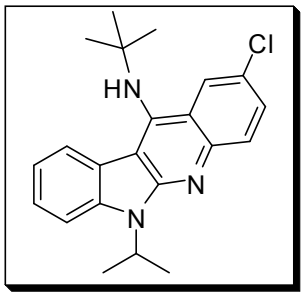


3l was prepared *via* the reaction of **1l** with **2a** according to the general procedure as mentioned above.

Orange solid; yield: 65%; mp: 142.6-143.9 °C; R_f (10% EtOAc/*n*-hexane) 0.63; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.37 (d, *J* = 8 Hz, 1H), 8.08-8.03 (m, 2H), 7.56 (t, *J* = 8 Hz, 1H), 7.48-7.40 (m,

2H), 7.30 (t, *J* = 7.6 Hz, 1H), 4.04 (bs, 1H), 3.97 (s, 3H), 1.38 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 158.8 (C-F *J* = 240 Hz), 153.2, 146.7 (C-F *J* = 5.5 Hz), 144.7, 142.2, 129.2 (C-F *J* = 8.6 Hz), 127.2, 124.1 (C-F *J* = 8.6 Hz), 122.7, 120.3, 119.4, 118.6 (C-F *J* = 25.5 Hz), 113.4, 109.1 (C-F *J* = 23.3 Hz), 108.5 57.8, 31.5 (3C), 27.6; ^{19}F NMR (376 MHz, $CDCl_3$) δ -120.1 (m); HPLC: 96.02%, column: X Bridge C-18 75*4.6 mm 3.5µ, mobile phase A: 0.1 % TFA in water mobile phase B: CH_3CN (gradient) T/B% : 0/20, 0.5/20, 2/95, 8/95, 20/95, 10/20, 12/20; flow rate: 1 mL/min; UV 270 nm, retention time 2.99 min; MS (ES mass): *m*/*z* 322.2 (M+1).

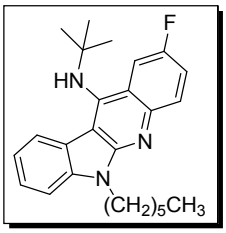
_N_-tert-butyl-2-chloro-6-isopropyl-6_H_-indolo[2,3-_b_]quinolin-11-amine (3m)



3m was prepared *via* the reaction of **1m** with **2a** according to the general procedure as mentioned above.

Light yellow semi-solid; yield: 62%; R_f (15% EtOAc-*n*-Hexane) 0.69; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.41-8.39 (m, 2H), 7.98 (d, *J* = 8.8 Hz, 1H), 7.63-7.57 (m, 2H), 7.54-7.50 (m, 1H), 7.30-7.28 (m, 1H), 5.70-5.62 (m, 1H), 4.09 (s, 1H), 1.77 (d, *J* = 7.2 Hz, 6H), 1.39 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 153.0, 146.4, 146.0, 140.5, 129.1, 129.0, 126.7, 126.6, 124.5, 124.1, 122.9, 121.1, 119.0, 113.1, 110.6, 57.8, 44.9, 31.5 (3C), 20.4 (2C); HPLC: 98.6%, column: X-BRIDGE C-18 150*4.6 mm, 5µ, mobile phase A: 5mM Ammonium Formate in water mobile phase B: CH_3CN (gradient) T/%B: 0/70, 2/70, 12/98, 20/98, 22/70, 25/70; flow rate: 1.0 mL/min; UV 290 nm retention time 12.52 min; MS (ES mass): *m*/*z* 366.0 (M+1).

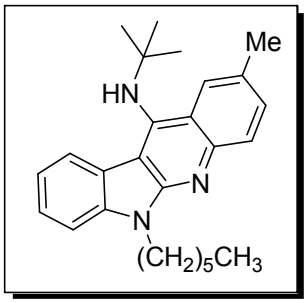
_N_-tert-butyl-2-fluoro-6-hexyl-6_H_-indolo[2,3-_b_]quinolin-11-amine (3n)



3n was prepared *via* the reaction of **1n** with **2a** according to the general procedure as mentioned above.

Yellow semi-solid; yield: 58%; R_f (15% EtOAc-*n*-Hexane) 0.36; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.37 (d, *J* = 7.6 Hz, 1H), 8.06-8.03 (m, 2H), 7.54 (t, *J* = 7.3 Hz, 1H), 7.47-7.41 (m, 2H), 7.30-7.26 (m, 1H), 4.49 (t, *J* = 7.4 Hz, 2H), 4.04 (bs, 1H), 1.98-1.90 (m, 2H), 1.49-1.42 (m, 2H), 1.38 (s, 9H), 1.35-1.27 (m, 4H), 0.88 (t, *J* = 7.2 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 158.8 (C-F *J* = 239.1 Hz), 152.9, 146.6, 146.5, 144.8, 141.7, 129.4 (C-F *J* = 8.4 Hz), 127.0, 124.0 (C-F *J* = 8.4 Hz), 122.8, 120.4, 119.2, 118.4 (C-F *J* = 25.6 Hz), 113.3, 109.0 (C-F *J* = 23.2 Hz), 108.8, 57.8, 41.4, 31.5 (3C), 28.4, 26.7, 22.5, 14.0; HPLC: 96.8%, column: X-BRIDGE C-18 150*4.6 mm, 5µ, mobile phase A: 5mM Ammonium Formate in water mobile phase B: CH_3CN (gradient) T/%B: 0/70, 2/70, 12/98, 20/98, 22/70, 25/70; flow rate: 1.0 mL/min; UV 280 nm retention time 14.12 min; MS (ES mass): *m*/*z* 392.1 (M+1).

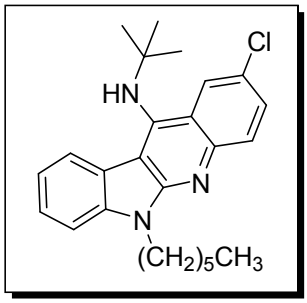
***N*-tert-butyl-6-hexyl-2-methyl-6*H*-indolo[2,3-*b*]quinolin-11-amine (3o)**



3o was prepared *via* the reaction of **1o** with **2a** according to the general procedure as mentioned above.

Brown semi-solid; yield: 66%; R_f (15% EtOAc-*n*-Hexane) 0.73; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.38 (d, *J* = 7.6 Hz, 1H), 8.17 (s, 1H), 7.97 (d, *J* = 8.4 Hz, 1H), 7.51-7.49 (m, 2H), 7.40 (d, *J* = 8.0 Hz, 1H), 7.26 (t, *J* = 7.6 Hz, 1H), 4.49 (t, *J* = 7.6 Hz, 2H), 4.08 (bs, 1H), 2.57 (s, 3H), 1.97-1.90 (m, 2H), 1.49-1.41 (m, 2H), 1.38 (s, 9H), 1.34-1.27 (m, 4H), 0.87 (t, *J* = 7.2 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 152.9, 146.7, 146.5, 141.5, 130.7, 130.4, 127.2, 126.5, 124.4, 123.5, 122.7, 121.0, 118.9, 112.7, 108.7, 57.7, 41.3, 31.6, 31.5 (3C), 28.4, 26.8, 22.6, 21.7, 14.1; HPLC: 93.7%, column: Symmetry C-18 75*4.6 mm, 3.5µm, mobile phase A: 0.1 % Triflouro acetic acid in water mobile phase B: CH_3CN (gradient) T/%B: 0/20, 1/20, 6/98, 10/98, 12/20, 15/20; flow rate: 1.0 mL/min; UV 275 nm retention time 5.84 min; MS (ES mass): *m*/*z* 388.1 (M+1).

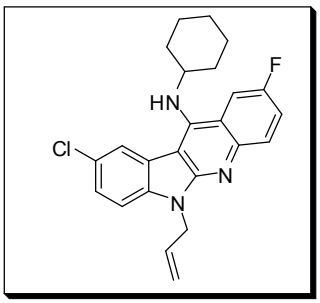
***N*-tert-butyl-2-chloro-6-hexyl -6*H*-indolo[2,3-*b*]quinolin-11-amine (3p)**



3p was prepared *via* the reaction of **1p** with **2a** according to the general procedure as mentioned above.

Yellow solid; yield: 63%; mp: 77-78.2 °C; R_f (15% EtOAc-*n*-Hexane) 0.62; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.40 (d, *J* = 2.4 Hz, 1H), 8.35 (d, *J* = 7.6 Hz, 1H), 8.00 (d, *J* = 8.8 Hz, 1H), 7.60 (dd, *J* = 9.2, 2.4 Hz, 1H), 7.58-7.53 (m, 2H), 7.44 (d, *J* = 8.0 Hz, 1H), 7.31-7.27 (m, 1H), 4.48 (t, *J* = 7.2 Hz, 2H), 4.05 (s, 1H), 1.98-1.90 (m, 2H), 1.48-1.41 (m, 2H), 1.39 (s, 9H), 1.35-1.28 (m, 3H), 0.89 (t, *J* = 7.2 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 153.3, 146.4, 146.3, 141.6, 140.3, 129.1, 128.9, 127.0,126.6, 124.6, 124.3, 122.7, 120.5, 119.3, 113.1, 108.9, 57.7, 41.4, 31.5 (3C), 28.4, 26.7, 22.5, 14.0; HPLC: 99.85%, column: X-BRIDGE C-18 150*4.6 mm, 5µ, mobile phase A: 5mM Ammonium Formate in water mobile phase B: CH_3CN (gradient) T/%B: 0/70, 2/70, 12/98, 20/98, 22/70, 25/70; flow rate: 1.0 mL/min; UV 290 nm retention time 15.78 min; MS (ES mass): *m*/*z* 408.0

6-allyl-9-chloro-*N*-cyclohexyl-2-fluoro-6*H*-indolo[2,3-*b*]quinolin-11-amine (3q)

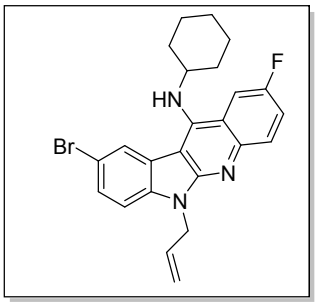


3q was prepared *via* the reaction of **1e** with **2b** according to the general procedure as mentioned above.

Yellow solid; yield: 68%; mp: 135.2-137.1 °C; R_f (10% EtOAc/*n*-hexane) 0.42; ^{1}H NMR (400 MHz, $CDCl_3$) δ: ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.02 (dd, *J* = 9.2, 5.6 Hz, 1H), 7.95 (d, *J* = 1.6 Hz, 1H), 7.72 (dd, *J* = 10.8, 2.8 Hz, 1H), 7.48-7.42 (m, 2H), 7.30 (d, *J* = 8.4 Hz, 1H), 6.09-5.99

(m, 1H), 5.21 (d, *J* = 9.6 Hz, 1H), 5.16-5.10 (m, 3H), 4.39-4.36 (m, 1H), 3.78-3.76 (m, 1H), 2.14-2.11 (m, 2H), 1.82-1.79 (m, 2H), 1.68-1.65 (m, 1H), 1.49-1.28 (m, 5H); ^{13}C NMR (100 MHz, $CDCl_3$)δ: 159.1 (C-F *J* = 240.1 Hz), 153.1, 147.8 (C-F *J* = 4.9 Hz), 144.9, 139.3, 132.5, 130.3 (C-F *J* = 8.6 Hz), 130.2, 126.1, 125.1, 122.0, 121.6, 118.9 (C-F *J* = 25.3 Hz), 116.9, 109.9, 106.2 (C-F *J* = 23.1 Hz), 105.5 , 57.3, 43.6, 35.4(2C) , 25.5, 25.3 (2C); ^{19}F NMR (376 MHz, $CDCl_3$) δ -119.0 (m); HPLC: 97.89%, column: X Bridge C-18 75*4.6 mm 3.5µ, mobile phase A: 0.1 % TFA in water mobile phase B: CH_3CN (gradient) T/B% : 0/20, 3/20, 8/40, 15/95, 20/95, 25/20,30/20; flow rate: 1 mL/min; UV 290 nm, retention time 3.30 min; MS (ES mass): *m*/*z* 408.2 (M+1).

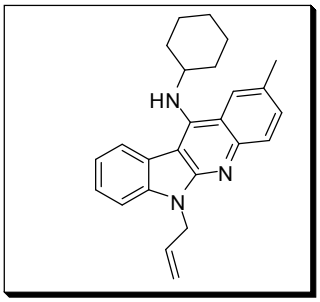
6-allyl-9-bromo-*N*-cyclohexyl-2-fluoro-6*H*-indolo[2,3-*b*]quinolin-11-amine (3r)



3r was prepared *via* the reaction of **1a** with **2b** according to the general procedure as mentioned above.

Yellow solid; yield: 68%; mp: 160-161.2 °C; R_f (10% EtOAc/*n*-hexane) 0.40; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.12 (d, *J* = 1.2 Hz, 1H), 8.04 (dd, *J* = 9.2, 5.6 Hz, 1H), 7.73 (dd, *J* = 10.4, 2.4 Hz, 1H), 7.60 (dd, *J* = 8.4, 1.6 Hz, 1H), 7.50-7.42 (td, *J* = 9.2, 2.4 Hz, 1H), 7.27 (s, 1H), 6.10-6.01 (m, 1H), 5.22 (d, *J* = 10.4 Hz, 1H), 5.20-5.12 (m, 3H), 4.44-4.35 (d, *J* = 10.4 Hz, 1H), 3.80-3.77 (m, 1H), 2.113 (d, *J* = 11.2 Hz, 2H), 1.84-1.81 (m, 2H), 1.69 (d, *J* = 9.6 Hz, 1H), 1.49-1.27 (m, 6H); ^{13}C NMR (100 MHz, $CDCl_3$)δ: 159.1 (C-F *J* = 240.3 Hz), 153.0, 147.8 (C-F *J* = 4.8 Hz), 144.9, 139.6, 132.5, 130.3 (C-F *J* = 8.6 Hz), 130.2, 128.8, 125.0, 122.2, 119.2 (C-F *J* = 8.2 Hz), 119.0 (C-F *J* = 25.3 Hz), 116.9, 112.4, 110.4, 106.2 (C-F *J* = 23.1 Hz), 105.4, 57.3, 43.6, 35.4 (2C), 25.5, 25.3 (2C); ^{19}F NMR (376 MHz, $CDCl_3$) δ -118.9 (m); HPLC: 99.34%, column: X Bridge C-18 75*4.6 mm 3.5µ, mobile phase A: 0.1 % TFA in water mobile phase B: CH_3CN (gradient) T/B% : 0/20, 3/20, 8/40, 15/95, 20/95, 25/20,30/20; flow rate: 1 mL/min; UV 290 nm, retention time 3.37 min; MS (ES mass): *m*/*z* 452.2 (M+1).

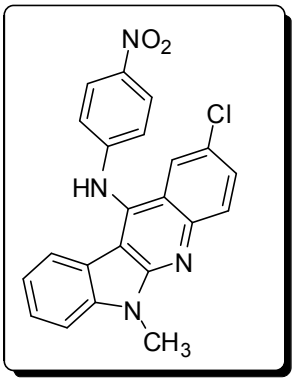
6-allyl-N-cyclohexyl-2-methyl-6H-indolo[2,3-b]quinolin-11-amine (3s)



3s was prepared *via* the reaction of **1i** with **2b** according to the general procedure as mentioned above.

Off white solid; yield: 47%; mp: 139-141 °C; R_f (10% EtOAc/*n*-hexane) 0.33; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.03 (d, *J* = 7.6 Hz, 1H), 7.96 (d, *J* = 8.8 Hz, 1H), 7.88 (s, 1H), 7.51-7.46 (m, 2H), 7.38 (d, *J* = 7.9 Hz, 1H), 7.29 (d, *J* = 7.2 Hz, 1H), 6.12-6.03 (m, 1H), 5.21-5.14 (m, 4H), 4.55 (bs, 1H), 3.86-3.81 (m, 1H), 2.59 (s, 3H), 2.13 (d, *J* = 10.8 Hz, 2H), 1.80-1.77 (m, 2H), 1.66-1.64 (m, 3H), 1.47-1.37 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$)δ: 147.6, 146.2, 140.9, 133.0, 131.0, 130.9, 127.9, 125.9, 122.1, 121.2, 120.9, 119.6, 119.2, 116.7, 109.9, 109.1, 106.1, 57.2, 43.6, 35.4, 25.7, 25.4, 21.8; HPLC: 99.11%, column: Symmetry C-18 75*4.6 mm 3.5µm, mobile phase A: 0.1 % TFA in water mobile phase B: CH_3CN (gradient) T/B% : 0/20, 0.5/20, 2/95, 10/95, 10.5/20, 12/20; flow rate: 0.1 mL/min; UV 210 nm, retention time 3.19 min; MS (ES mass): *m*/*z* 370.2 (M+1).

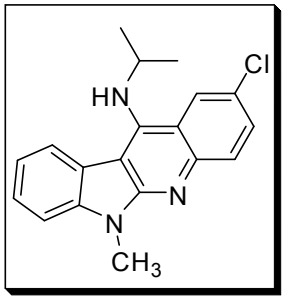
4-chloro-6-methyl-*N*-(4-nitrophenyl)-6*H*-indolo[2,3-*b*]quinolin-11-amine (3t)



3t was prepared *via* the reaction of **1k** with **2c** according to the general procedure as mentioned above.

White solid; yield: 26%; mp: 148-150 °C; R_f (15% EtOAc-*n*-Hexane) 0.42; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.10 (d, *J* = 8.4 Hz, 1H), 7.97 (s, 1H), 7.88-7.85 (m, 1H), 7.83 (d, *J* = 1.6 Hz, 1H), 7.38-7.35 (m, 1H), 7.33 (d, *J* = 8.4 Hz, 2H), 7.28 (d, *J* = 1.6 Hz, 1H), 7.26-7.23 (m, 1H), 7.12 (dd, *J* = 8.4, 1.9 Hz, 1H), 6.67-6.63 (m, 1H), 4.37 (bs, 1H), 3.87 (s, 3H); HPLC: 95.1%, column: X-Bridge C-18 150*4.6 mm, 5µm, mobile phase A: 5mM Ammonium Formate in water mobile phase B: CH_3CN (gradient) T/%B: 0/70, 2/70, 12/98, 20/98, 22/70, 25/70; flow rate: 1.0 mL/min; UV 254.4 nm retention time 4.45 min; MS (ES mass): *m*/*z* 403.2 (M+1).

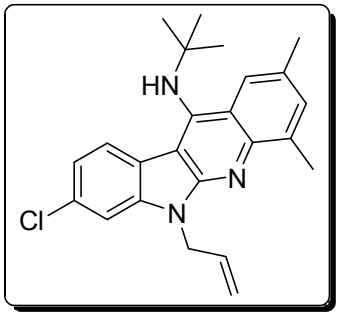
2-Chloro-*N*-isopropyl-6-methyl-6*H*-indolo[2,3-*b*]quinolin-11-amine (**3u**)



3u was prepared *via* the reaction of **1k** with **2d** according to the general procedure as mentioned above.

Pale yellow low melting solid; yield: 57%; R_f (15% EtOAc-*n*-Hexane) 0.59; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.38 (d, *J* = 2.3 Hz, 1H), 8.32 (d, *J* = 7.9 Hz, 1H), 7.98 (d, *J* = 8.7 Hz, 1H), 7.60-7.53 (m, 2H), 7.40 (d, *J* = 7.9 Hz, 1H), 7.30 (t, *J* = 7.5 Hz, 1H), 4.64-4.33 (m, 1H), 4.05 (s, 1H), 3.96 (s, 3H), 1.38 (d, *J* = 5.2 Hz, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 153.5, 146.4, 142.0, 129.1, 128.6, 127.1, 124.6, 122.5, 121.2, 120.4, 119.2, 118.0, 113.1, 110.7, 108.5, 44.9, 27.6, 20.5 (2C); HPLC: 95.7%, column: Symmetry C-18 150*4.6 mm, 5µm, mobile phase A: 5mM Ammonium Formate in water mobile phase B: CH_3CN (gradient) T/%B: 0/70, 2/70, 12/98, 20/98, 22/70, 25/70; flow rate: 1.0 mL/min; UV 280 nm retention time 9.20 min; MS (ES mass): *m*/*z* 324.5 (M+1).

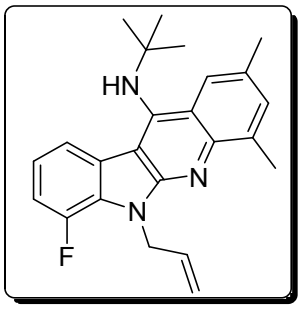
6-Allyl-*N*-tert-butyl-8-Chloro-2,4-dimethyl-6*H*-indolo[2,3-*b*]quinolin-11-amine (4a)



4a was prepared *via* the reaction of **1q** with **2a** according to the general procedure as mentioned above.

brown semi-solid; yield: 60%; mp: R_f (10% EtOAc/*n*-hexane) 0.72; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.38 (d, *J* = 1.6 Hz, 1H), 7.96 (s, 1H), 7.42 (m, 2H), 7.30-7.27 (d, *J* = 8.4 Hz, 1H), 6.10-6.00 (m, 1H), 5.23 (d, *J* = 1.2 Hz, 1H), 5.21-5.18 (m, 1H), 5.14 (d, *J* = 5.6 Hz, 2H), 3.93 (s, 1H), 2.82 (s, 3H), 2.53 (s, 3H), 1.36 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 151.8, 147.2, 145.6, 139.7, 134.9, 132.7, 131.3, 130.3, 126.2, 124.3, 123.4, 122.7, 122.5, 121.7, 117.0, 111.8, 109.8, 57.8, 43.7 31.3 (3C), 21.6, 18.4; HPLC: 94.65%, column: X Bridge C-18 75*4.6 mm 3.5µ, mobile phase A: 0.1 % TFA in water mobile phase B: CH_3CN (gradient) T/B% : 0/20, 3/20, 8/40, 15/95, 20/95, 25/20,30/20; flow rate: 1 mL/min; UV 285 nm, retention time 5.66 min; MS (ES mass): *m*/*z* 392.2 (M+1).

6-allyl-*N*-tert-butyl-7-fluoro-2,4-dimethyl-6*H*-indolo[2,3-*b*]quinolin-11-amine (4b)

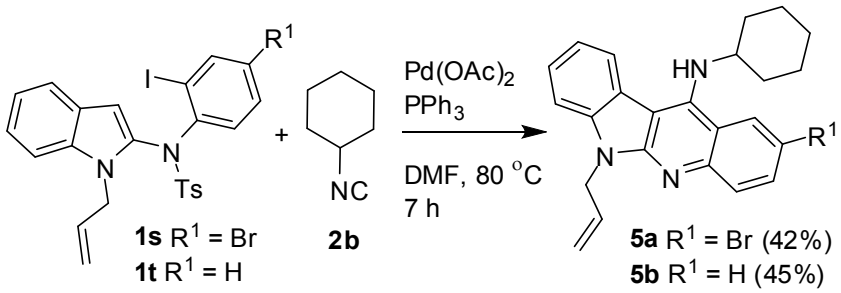


4b was prepared *via* the reaction of **1r** with **2a** according to the general procedure as mentioned above.

Orange solid; yield: 58%; mp: 146.5-147.5 °C; R_f (10% EtOAc/*n*-hexane) 0.68; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.17 (d, *J* = 1.2 Hz, 1H), 7.99 (s, 1H), 7.40 (s, 1H), 7.22-7.12 (m, 2H), 6.19-6.09 (m, 1H), 5.32 (dd, *J* = 5.6, 1.2 Hz, 2H), 5.18 (d, , *J* = 17.2 Hz, 1H), 5.13 (dd, , *J* = 10, 1.2 Hz, 1H) 4.03 (s, 1H), 2.82 (s, 3H), 2.52 (s, 3H), 1.34 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ:

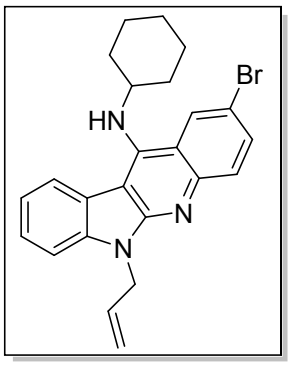
151.7, 150.2, 145.6, 134.9, 133.8, 131.2 (C-F J = 19.8 Hz), 130.8, 130.2, 128.6, 124.6, 123.4, 121.9, 119.2 (C-F J = 6.4 Hz), 118.6 (C-F J = 3.2 Hz), 116.5 , 113.1 (C-F J = 18.6 Hz), 109.9 57.7, 45.4 31.3 (3C), 21.6, 18.4; HPLC: 99.42%, column: X Bridge C-18 75*4.6 mm 3.5µ, mobile phase A: 0.1 % TFA in water mobile phase B: CH_3CN (gradient) T/B% : 0/20, 3/20, 8/40, 15/95, 20/95, 25/20,30/20; flow rate: 1 mL/min; UV 275 nm, retention time 5.99 min; MS (ES mass): m/z 376.2 (M+1).

General Procedure for the preparation of indolo[2,3-*b*]quinolin-11-amines (5)



To mixture of *N*-(4-substituted-2-iodophenyl)-*N*-(1-alkyl-1*H*-indol-2-yl)4-methylbenzene-sulfonamide (**1**) (0.3 mmol), $Pd_2(dba)_3$ (5 mol%), PPh_3 (11 mol%) and Cs_2CO_3 in anhydrous DMF (3 mL) was added slowly isocyanide (0.9 mmol) for 10 minutes then stirred at 60 °C under nitrogen atmosphere for 7 h. The progress of the reaction was monitored by TLC. Upon completion, the mixture was cooled to room temperature, and filtered to remove the solid materials. The filtrate was extracted with ethyl acetate (3 x 15 mL). The organic layers were collected, combined, dried over anhydrous Na_2SO_4, filtered and concentrated under a reduced pressure. The residue was purified by column chromatography over silica gel using ethyl acetate–hexane to give the desired product **3**.

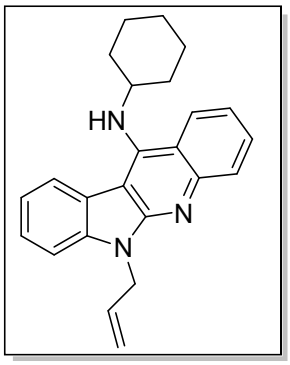
6-Allyl-2-bromo-*N*-cyclohexyl-6*H*-indolo[2,3-*b*]quinolin-11-amine (5a)



5a was prepared *via* the reaction of **1s** with **2b** according to the general procedure as mentioned above.

Yellow semi solid; yield: 42%; R_f (10% EtOAc/*n*-hexane) 0.56; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.25 (d, *J* = 2.4 Hz, 1H), 8.02 (d, *J* = 7.6 Hz, 1H), 7.92 (d, *J* = 8.8 Hz, 1H), 7.69 (dd, *J* = 9.2, 2.4 Hz, 1H), 7.52-7.48 (m, 1H), 7.40 (d, *J* = 8.0 Hz, 1H), 7.32 (t, *J* = 7.6 Hz, 1H), 6.11-6.02 (m, 1H), 5.21-5.12 (m, 4H), 4.52 (bs, 1H), 3.82 (bs, 1H), 2.36-2.29 (m, 2H), 2.15-2.12 (m, 2H), 2.05-1.99 (m, 4H), 1.81-1.78 (m, 2H); MS (ES mass): *m*/*z* 436.0 (M+2).

6-Allyl-*N*-cyclohexyl-6*H*-indolo[2,3-*b*]quinolin-11-amine (5b)



5b was prepared *via* the reaction of **1t** with **2b** according to the general procedure as mentioned above.

Yellow solid; yield: 45%; mp: 132-134 °C; R_f (10% EtOAc/*n*-hexane) 0.35; ^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.15-8.12 (m, 1H), 8.05-8.02 (m, 2H), 7.69-7.62 (m, 1H), 7.48-7.46 (m, 1H), 7.40-7.38 (m, 2H), 7.32-7.29 (m, 1H), 6.13-6.03 (m, 1H), 5.21-5.15 (m, 4H), 4.62 (s, 1H), 3.89-3.84 (m, 1H), 2.16-2.12 (m, 2H), 1.80-1.77 (m, 2H), 1.66-1.61 (m, 1H), 1.43-1.40 (m, 2H), 1.37-1.28 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 148.2, 140.8, 132.9, 128.8, 128.1, 125.9 (2C), 122.5 (2C), 121.9, 121.6, 120.9, 119.8, 119.1, 116.7, 109.9, 109.2, 57.4, 43.7, 35.4 (2C), 25.6, 25.3 (2C); HPLC: 99.84%, column: Symmetry C-18 75*4.6 mm 3.5µm, mobile phase A: 0.1 % TFA in water mobile phase B: CH_3CN (gradient) T/%B : 0/20, 0.5/20, 2/95, 8/95, 10/20, 12/20; flow rate: 1.0 mL/min; UV 275 nm, retention time 4.52 min; MS (ES mass): *m*/*z* 356.2 (M+1).

Biology

Sulphorhodamine B (SRB) Assay:

The principle: The anti-proliferative activity and cancer cell selectivity of the synthesized compounds on cancer cells was evaluated using the SRB (Sulforhodamine B) cell proliferation assay. This assay was chosen because of its sensitivity, large dynamic range and the ability to measure cell proliferation over three days with normalization to initial cell number as well as to vehicle-treated cells. Further, this assay is the standardized assay of choice for screening of anticancer compounds at the National Cancer Institute (NIH). The SRB assay provides a colorimetric readout which can be spectrophotometrically measured and does not involve antibodies or toxic reagents. The assay is based on detection of total protein content of cells, which increases or decreases in proportion with cell number.

The methodology: Cancer cells (around 5000 in number) were seeded in 96-well plates and incubated overnight. The optimum cell number to be seeded was determined by a growth curve analysis for the cell line. Compounds (dissolved in 100% DMSO to a stock concentration of 200mM) were added to the adhered cells at a final concentration of 10μM. After 72h of treatment, the cells were washed with phosphate-buffered saline and ice-cold 10% trichloroacetic acid was added to the cells to precipitate the proteins. It was incubated for 1h at 4 ^{0}C. The cells were then washed with water and air-dried. Cellular proteins were then stained using 0.4% SRB solution in 1% acetic acid for 30 min at room temperature. The unbound dye was washed away by destaining with 1% acetic acid and bound dye was solubilized with 10mM Tris solution (pH 10.5). Absorbance of solubilized dye was measured at a wavelength of 590 nm. Percentage growth was determined by the formula

[(At-A0/Ac-A0)] X 100

where At=absorbance after 72h of test compound treatment,

A0=Absorbance at time 0,

Ac=Absorbance after 72h without treatment.

The known cytotoxic agent, gemcitabine was used as a positive control in the assay.

Zebrafish embryo studies:

Materials and Methods:

Husbandry:

Zebrafish obtained from a local vendor were maintained in in-house built recirculatory system under 14-10hrs light dark cycle and 28˚C temperature as described in (Banote et al., 2013). Breeding was carried out using females and males in ratio of 2:3 and the embryos obtained were collected in petridishes and maintained at 28^0C. (Westerfield et al., 2000, Nakhi et al.,2013).

Hepatotoxicity assay:

The Transparency of Zebrafish larvae and similarity in toxicity profiles with human allow them for various toxicity assessment studies of different test compounds like hepatotoxicity assay. In this assay 4dpf embryos were exposed to various concentration of test compound prepared from stock solutions as described above. The embryos were distributed in 24 well plate along with 250µl of 0.1% DMSO with 6 embryos in each well. Each well is added with the respective working stock solutions to obtain the final concentration of 1, 3, 10 and 30µM concentration of the drug. The plate was incubated at 28˚C until 7dpf. Embryos were washed with E3 medium on 7dpf and anesthetized using tricaine. The images of embryos treated with different compounds of various concentrations are analyzed using Image J software for their liver size, liver degeneration and yolk sac retention and percentages were calculated.

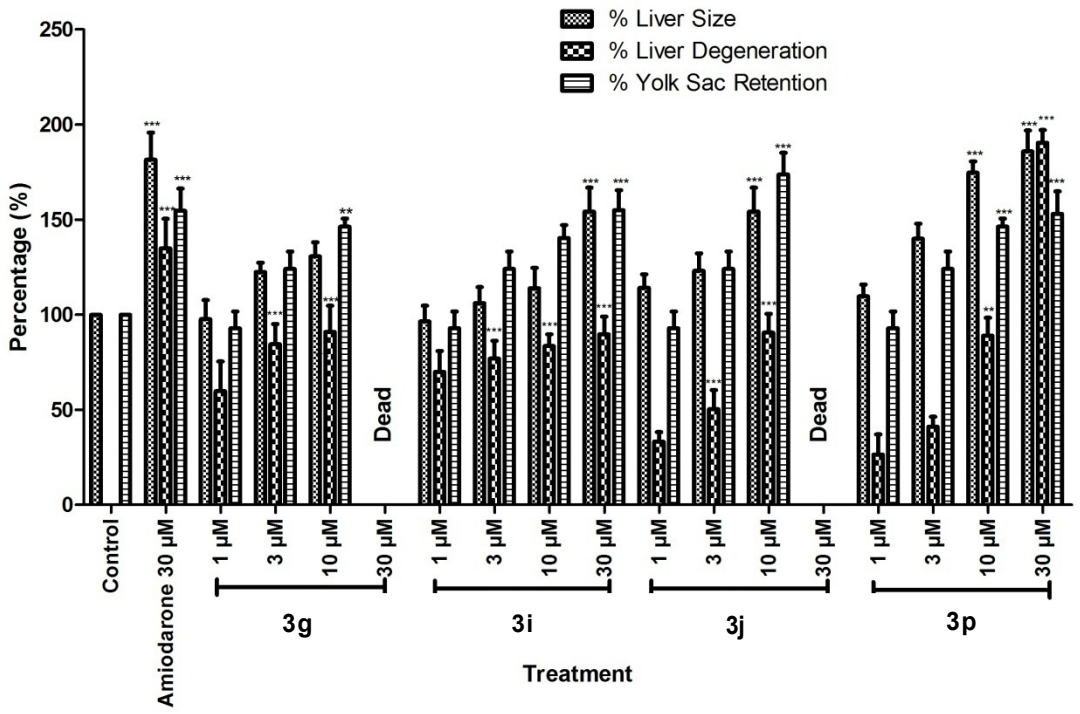
Teratogenicity assay:

Zebrafish appeal of rapid organogenesis and transparency of developing embryos made it as a promising model for teratogenicity assay. In this assay, 1dpf embryos at same developmental stage were sorted out and dechorionated using protease K. Test compounds stock solutions were prepared by dissolving in 100% DMSO. By serial dilution from stock solutions various concentrations were prepared and the final concentration of DMSO becomes 0.1%. The embryos were distributed in 24 well plate (3/well) and concentrations of test compounds starting from 1µM to 30µM compound was added to each well accordingly where n=6. The plate was incubated at 28˚C until 5dpf. The embryos were washed with PBS and anesthetized using tricaine (0.008%). Morphological scoring was done based on the procedure previously described (Panzica-Kelly et al, 2010).

Results:

Hepatotoxicity assay:

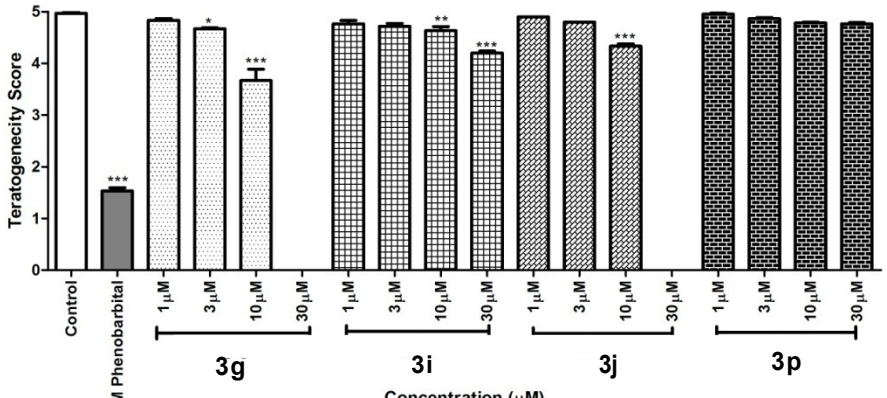
Compound **3g** did not show toxicity at 1µM whereas the toxicity increased significantly with increase in the concentration e.g. at 30µM embryos were found dead. The compound **3i** at 10µM showed minimal toxicity and it was significant at 30µM. The compound **3j** did not show any toxicity at 1µM & 3µM whereas toxicity was increased significantly at 10µM and embryos were dead at 30µM. Compound **3p** showed moderate toxicity at 10µM and severe toxicity at 30µM.



Graph S-1: All the statistical analysis was done using GraphPad Prism® software. The following graph represents the qualitative data of % liver size, % liver degeneration & % yolk sac retention of three compounds at different concentrations when compared to positive control Amiodarone.

Teratogenicity assay:

Compounds **3g**, **3i**, **3j** and **3p** were tested for Teratogenicity. Compound **3p** was found to be non toxic in all the tested concentrations whereas, compound **3i** was found to be toxic at 30µM but found to be safe 1, 3 and 10µM. In case of compound **3g** & **3j** toxicity was observed at 10 µM and embryos were dead at 30µM.



Graph S-2: Each embryo was scored based on their level of toxicity from 5 being non toxic and 0.5 being highly toxic. Statistical analysis for scoring was done using GraphPad Prism® software using two-way ANOVA. The following graph represents the teratogenicity scoring given compared to the positive control Phenobarbital.

Table S-1: Results of zebrafish embryo toxicity study with toxicological indices and major organs/systems affected in positive control and at MTC in test compounds. (- no effect, x- slightly toxic, xx-moderately toxic, xxx-severely toxic).

	3g	**3i**	**3j**	**3p**	**Phenobarbital**
Test Concentrations (µM)	1, 3, 10, 30	1, 3, 10, 30	1, 3, 10, 30	1, 3, 10, 30	3000
Statistically Significant Toxic Concentration (µM)	-	30	-	30	Positive Control
No Observed Adverse Effect Level (NOAEL) (µM)	1	3	3	30	
Parameters of toxicity at MTC					
Body Shape	-	xx	-	-	xxx
Somites	-	xx	-	-	xxx

Notochord	-	xxx	-	-	xxx
Tail	xx	xx	-	-	xxx
Fins	xx	xxx	-	-	xxx
Brain	-	xxx	-	-	xxx
Upper jaw	xx	xxx	-	-	xxx
Heart	xxx	xx	xx	-	xx
Intestine	xx	xx	xx	-	xxx
Lower jaw	xx	xxx	-	-	xx
Liver	xx	xxx	xx	-	xxx
Swim Bladder	-	xx	xx	-	xxx

References:

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