Supporting information

Identification of sulfonylated indolo[1,2-*a*]quinolines as EGFR tyrosine kinase inhibitors

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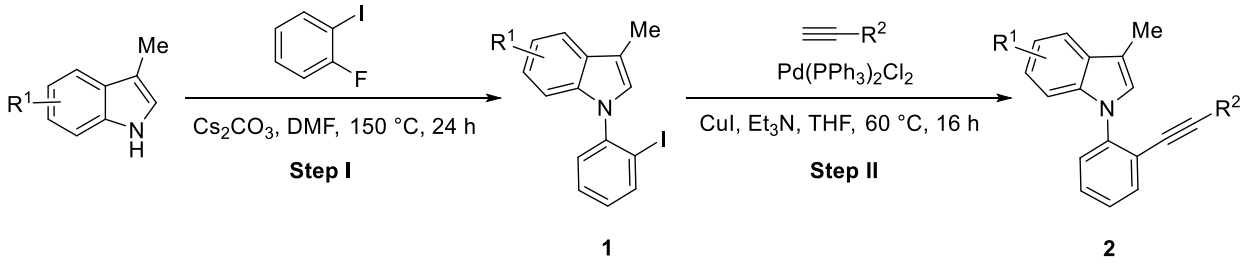
1. General Information

^{1}H (400 MHz), ^{13}C (101 MHz), and ^{19}F (376 MHz) NMR spectra were reported in ppm and recorded with a Bruker Ascend^{TM} spectrometer (400 MHz). Chemical shift (δ) was measured using tetramethylsilane (TMS) (δ = 0 ppm) and trichlorofluoromethane ($CFCl_3$) (δ = 0 ppm) as an internal reference. Melting points (uncorrected) were recorded in degree celsius (°C) using a Buchi M565 melting point apparatus. IR spectra were recorded on a Bruker ALPHA FTIR spectrometer equipped with a Platinum ATR module. The high-resolution mass spectra (HRMS) were recorded on a Bruker micro TOF spectrometer in the ESI mode. Unless otherwise noted, the commercial-grade chemicals were used without prior purification. Common solvents for purification (hexanes, dichloromethane, acetone and ethyl acetate) were distilled before used. Reactions were monitored by thin-layer chromatography and visualized by UV (254 and 320 nm). Column chromatography was performed with silica gel 60 (Merck, 70-230 mesh).

2. Experimental Procedure

2.1 Experimental Procedure 1: Synthesis of 3-Methyl-1-(2-(alkynyl)phenyl)indoles (2)

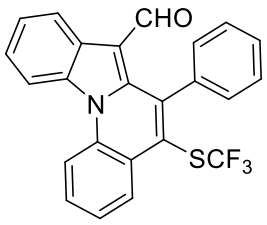
The 3-methyl-1-(2-(alkynyl)phenyl)indoles (**2**) utilized in this study were synthesized by following the previously published methods.[1-2]



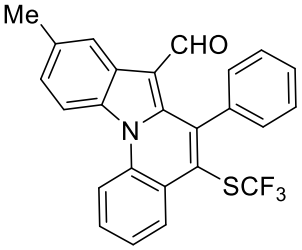
Step I: To a round-bottomed flask charged with 3-methyl-1*H*-indoles (5.0 mmol, 1.0 equiv), Cs_2CO_3 (20.0 mmol, 4.0 equiv), and 1-fluoro-2-iodobenzene (20.0 mmol, 4.0 equiv) was added DMF (20 mL). The reaction flask was sealed with a septum. The reaction mixture was heated at 150 °C for 24 h before it was extracted with EtOAc (3 × 20 mL). The combined organic layers were washed with brine (100 mL) and dried over anh. Na_2SO_4, and filtered. The solvent was removed in vacuo, and the crude residue was purified by column chromatography on silica gel using 100% hexanes as the eluent to afford 3-methyl-1-(2-iodophenyl)-1*H*-indole derivatives (**1**).

Step II: To a two-necked flask charged with **1** (3.0 mmol, 1.0 equiv), $PdCl_2(PPh_3)_2$ (0.15 mmol, 0.05 equiv), and CuI (0.03 mmol, 0.01 equiv) was added THF (15 mL) under an argon atmosphere. Then, Et_3N (6.0 mmol, 2.0 equiv) and arylacetylene (3.6 mmol, 1.2 equiv) were added to the reaction mixture. The resulting mixture was allowed to stir at 60 °C under an argon atmosphere overnight (16 h). The reaction mixture was extracted with EtOAc (3 × 20 mL). The combined organic layers were washed with brine (20 mL) and dried over anh. Na_2SO_4, and filtered. The solvent was removed in vacuo, and the crude residue was purified by column chromatography on silica gel using 100% hexanes as the eluent to obtain 3-alkyl-1-(2-(alkynyl)phenyl)indoles (**2**).

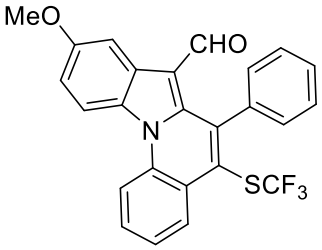
2.2 Experimental Procedure 2: Synthesis of 6-Trifluoromethylthio indolo[1,2-*a*]quinolines (IQSCF$_3$-I – IQSCF$_3$-VI)



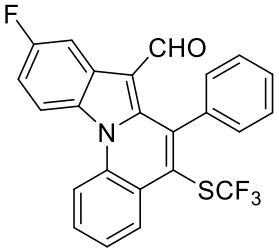
IQSCF$_3$-I



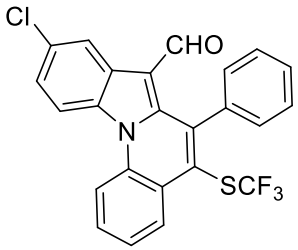
IQSCF$_3$-II



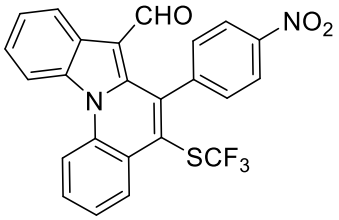
IQSCF$_3$-III



IQSCF$_3$-IV

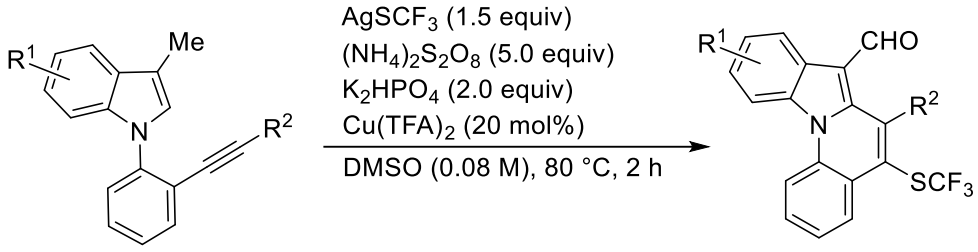


IQSCF$_3$-V



IQSCF$_3$-VI

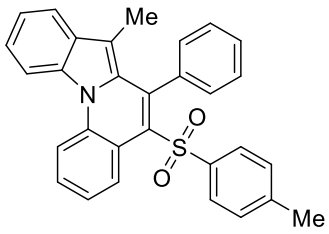
The 6-trifluoromethylthio indolo[1,2-*a*]quinolines (**IQSCF$_3$-I** – **IQSCF$_3$-VI**) used in the present work were prepared by following the previously reported protocol.[1]



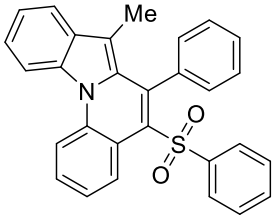
2 **IQSCF$_3$s**

To a round-bottomed flask charged with **2** (0.25 mmol), $AgSCF_3$ (1.5 equiv), $(NH_4)_2S_2O_8$ (5.0 equiv), K_2HPO_4 (2.0 equiv), and $Cu(TFA)_2$ (20 mol%) was added DMSO (3 mL, 0.08 M). The reaction flask was sealed with a septum and stirred at 80 °C. After 2 h, the resulting mixture was diluted with CH_2Cl_2 and filtered through a pad of aluminum oxide (type E), which was eluted with CH_2Cl_2. Then, the filtrate was extracted with CH_2Cl_2 (3× 10 mL). The organic layer was washed with brine (20 mL) and dried over anh. Na_2SO_4. The solvent was removed in vacuo, and the crude residue was purified by column chromatography on silica gel using CH_2Cl_2/hexane as the eluent to provide the corresponding products (**IQSCF$_3$s**).

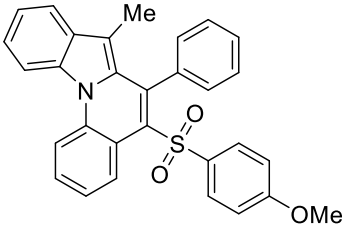
2.3 Experimental Procedure 3: Synthesis of 6-Arenesulfonyl indolo[1,2-*a*]quinolines (IQSO$_2$R-I – IQSO$_2$R-IX)



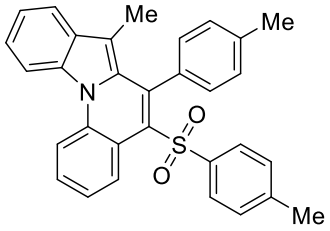
IQSO$_2$R-I



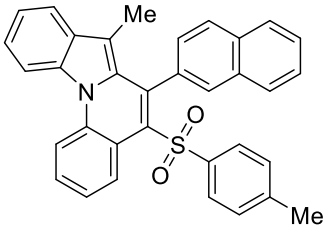
IQSO$_2$R-II



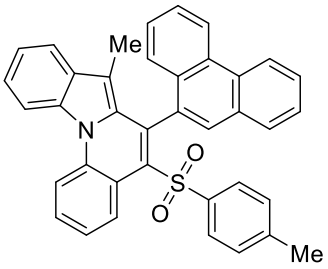
IQSO$_2$R-III



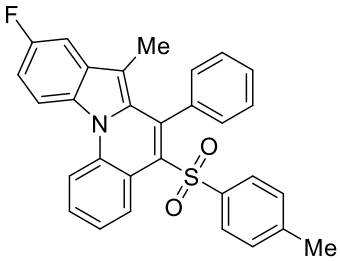
IQSO$_2$R-IV



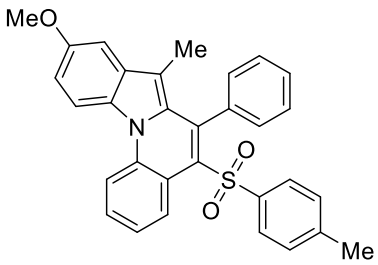
IQSO$_2$R-V



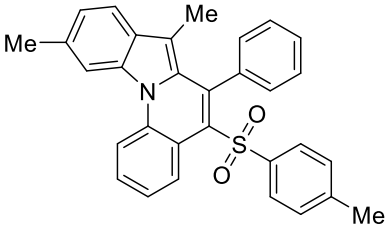
IQSO$_2$R-VI



IQSO$_2$R-VII

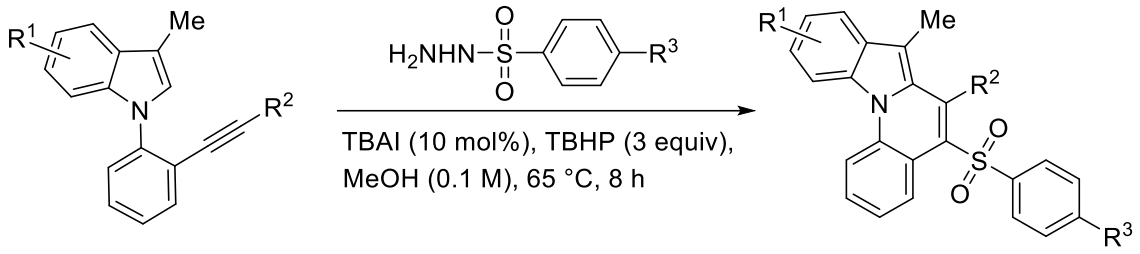


IQSO$_2$R-VIII



IQSO$_2$R-IX

The 6-arenesulfonyl indolo[1,2-*a*]quinolines (**IQSO$_2$R-I** – **IQSO$_2$R-IX**) used in the present work were prepared by following the previously reported protocol.[2]

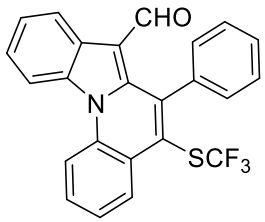


2 **IQSO$_2$Rs**

To a stirred solution of **2** (0.5 mmol) in MeOH (5 mL, 0.1 M) was added arenesulfonyl hydrazide (2.0 equiv). Then, TBAI (10 mol%), TBHP (3 equiv) were added. The mixture was allowed to stir at 65 °C for 8 h. After the reaction was completed, the solvent was evaporated under vacuo. Then, the residue was quenched with water (5 mL). The resulting mixture was then extracted with CH_2Cl_2 (3 ×15 mL). The combined organic layers were dried over anh. Na_2SO_4, filtered, and concentrated under reduced pressure to provide a crude compound. The residue was purified by silica gel chromatography (10% EtOAc/hexanes) to afford the corresponding products (**IQSO$_2$Rs**).

3. Characterization Data

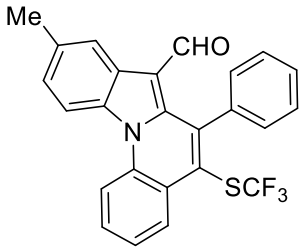
6-Phenyl-5-((trifluoromethyl)thio)indolo[1,2-*a*]quinoline-7-carbaldehyde (IQSCF$_3$-I)



IQSCF$_3$-I

Prepared according to experimental procedure 2. Isolated by column chromatography (SiO_2, 40% CH_2Cl_2/hexanes) as a yellow solid (82.4 mg, 78% yield). The NMR data for **IQSCF$_3$-I** were reported in the previously published literature.[1]

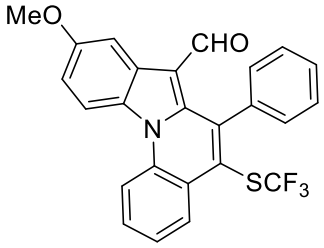
9-Methyl-6-phenyl-5-((trifluoromethyl)thio)indolo[1,2-*a*]quinoline-7-carbaldehyde (IQSCF$_3$-II)



IQSCF$_3$-II

Prepared according to experimental procedure 2. Isolated by column chromatography (SiO_2, 40% CH_2Cl_2/hexanes) as a yellow solid (72.9 mg, 67% yield). The NMR data for **IQSCF$_3$-II** were reported in the previously published literature.[1]

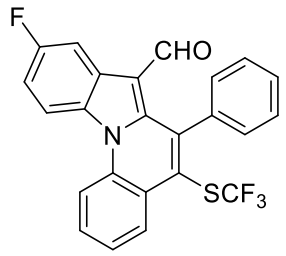
9-Methoxy-6-phenyl-5-((trifluoromethyl)thio)indolo[1,2-*a*]quinoline-7-carbaldehyde (IQSCF$_3$-III)



IQSCF$_3$-III

Prepared according to experimental procedure 2. Isolated by column chromatography (SiO_2, 40% CH_2Cl_2/hexanes) as a yellow solid (58.0 mg, 51% yield). The NMR data for **IQSCF$_3$-III** were reported in the previously published literature.[1]

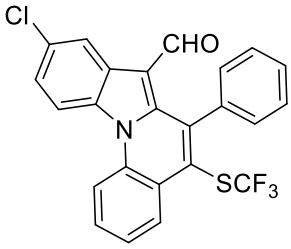
9-Fluoro-6-phenyl-5-((trifluoromethyl)thio)indolo[1,2-*a*]quinoline-7-carbaldehyde (IQSCF$_3$-IV)



IQSCF$_3$-IV

Prepared according to experimental procedure 2. Isolated by column chromatography (SiO_2, 40% CH_2Cl_2/hexanes) as a yellow solid (63.4 mg, 58% yield). The NMR data for **IQSCF$_3$-IV** were reported in the previously published literature.[1]

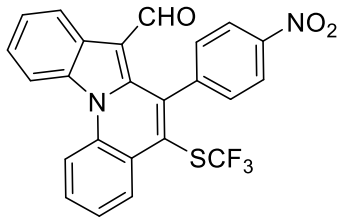
9-Chloro-6-phenyl-5-((trifluoromethyl)thio)indolo[1,2-*a*]quinoline-7-carbaldehyde (IQSCF$_3$-V)



IQSCF$_3$-V

Prepared according to experimental procedure 2. Isolated by column chromatography (SiO_2, 40% CH_2Cl_2/hexanes) as a yellow solid (78.5 mg, 69% yield). The NMR data for **IQSCF$_3$-V** were reported in the previously published literature.[1]

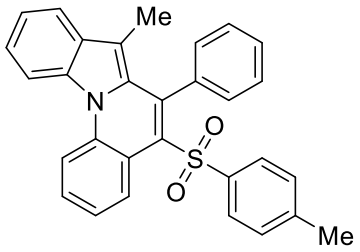
6-(4-Nitrophenyl)-5-((trifluoromethyl)thio)indolo[1,2-*a*]quinoline-7-carbaldehyde (IQSCF$_3$-VI)



IQSCF$_3$-VI

Prepared according to experimental procedure 2. Isolated by column chromatography (SiO_2, 50% CH_2Cl_2/hexanes) as a yellow solid (58.3 mg, 50% yield). The NMR data for **IQSCF$_3$-VI** were reported in the previously published literature.[1]

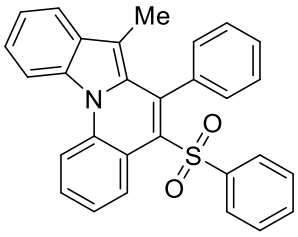
7-Methyl-6-phenyl-5-tosylindolo[1,2-*a*]quinoline (IQSO$_2$R-I)



IQSO$_2$R-I

Prepared according to experimental procedure 3. Isolated by column chromatography (SiO_2, 10% EtOAc/hexanes) as a yellow solid (161.6 mg, 70% yield). The NMR data obtained for **IQSO$_2$R-I** are in agreement with previously reported literature values.[2,3] ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.76 (d, J = 8.6 Hz, 1H), 8.45 (d, J = 8.4 Hz, 1H), 8.35 (d, J = 8.6 Hz, 1H), 7.74 (d, J = 8.0 Hz, 1H), 7.58−7.54 (m, 2H), 7.52−7.33 (m, 6H), 7.31−7.23 (m, 3H), 7.11 (d, J = 8.1 Hz, 2H), 2.31 (s, 3H), 1.55 (s, 3H); ^{13}C{^{1}H} NMR (101 MHz, $CDCl_3$): δ 143.3, 140.3, 140.1, 136.0, 135.7, 132.3, 131.5, 130.2, 130.0 (2C), 129.4 (2C), 128.9, 128.3, 128.0, 127.7 (2C), 127.6, 126.6 (2C), 124.5, 122.7, 121.9, 120.0, 119.1, 115.3, 114.5, 114.3, 21.5, 10.0 ppm.

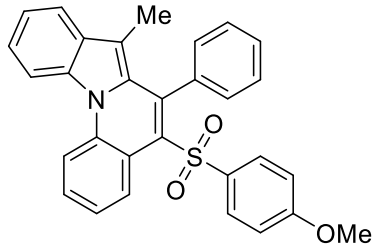
7-Methyl-6-phenyl-5-(phenylsulfonyl)indolo[1,2-*a*]quinoline (IQSO$_2$R-II)



IQSO$_2$R-II

Prepared according to experimental procedure 3. Isolated by column chromatography (SiO_2, 10% EtOAc/hexanes) as a yellow solid (147.7 mg, 66% yield). The NMR data obtained for **IQSO$_2$R-II** are in agreement with previously reported literature values.[2,3] ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.77 (d, J = 8.3 Hz, 1H), 8.49 (d, J = 8.4 Hz, 1H), 8.40 (d, J = 8.6 Hz, 1H), 7.77 (d, J = 8.1 Hz, 1H), 7.67 (d, J = 7.5 Hz, 2H), 7.58−7.50 (m, 2H), 7.46−7.28 (m, 10H), 1.58 (s, 3H); ^{13}C{^{1}H} NMR (101 MHz, $CDCl_3$): δ 143.3, 140.4, 136.0, 135.5, 132.5, 132.4, 131.6, 130.2, 130.1 (2C), 129.0, 128.8 (3C), 128.3, 127.8 (2C), 127.6, 126.5 (2C), 124.6, 122.8, 122.0, 120.0, 119.2, 115.4, 114.6, 114.4, 10.0 ppm.

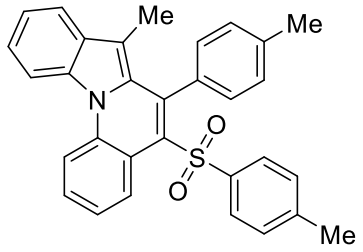
5-((4-Methoxyphenyl)sulfonyl)-7-methyl-6-phenylindolo[1,2-*a*]quinoline (IQSO$_2$R-III)



IQSO2R-III

Prepared according to experimental procedure 3. Isolated by column chromatography (SiO_2, 10% EtOAc/hexanes) as a yellow solid (143.3 mg, 60% yield). The NMR data obtained for **IQSO$_2$R-III** are in agreement with previously reported literature values.[2] ^{1}H NMR (400 MHz, $CDCl_3$): *δ* 8.81 (dd, *J* = 8.4, 1.2 Hz, 1H), 8.48 (d, *J* = 8.3 Hz, 1H), 8.39 (d, *J* = 8.6 Hz, 1H), 7.76 (d, *J* = 8.0 Hz, 1H), 7.61−7.49 (m, 4H), 7.45−7.36 (m, 4H), 7.31−7.27 (m, 3H), 6.79 (d, *J* = 9.0 Hz, 2H), 3.80 (s, 3H), 1.56 (s, 3H); ^{13}C{^{1}H} NMR (101 MHz, $CDCl_3$): *δ* 162.8, 139.7, 136.1, 135.8, 134.9, 132.4, 131.6, 130.3, 130.1 (2C), 129.0 (3C), 128.6, 128.3, 127.8 (2C), 127.7, 124.5, 122.7, 121.9, 120.0, 119.2, 115.4, 114.6, 114.2, 114.0 (2C), 55.6, 10.0 ppm.

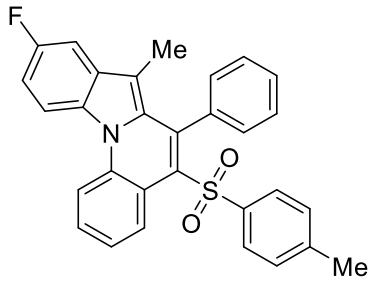
7-Methyl-6-(*p*-tolyl)-5-tosylindolo[1,2-*a*]quinoline (IQSO$_2$R-IV)



IQSO$_2$R-IV

Prepared according to experimental procedure 3. Isolated by column chromatography (SiO_2, 10% EtOAc/hexanes) as a yellow solid (130.8 mg, 55% yield). The NMR data obtained for **IQSO$_2$R-IV** are in agreement with previously reported literature values.[2] ^{1}H NMR (400 MHz, $CDCl_3$): *δ* 8.74 (d, *J* = 8.3 Hz, 1H), 8.46 (d, *J* = 8.4 Hz, 1H), 8.37 (d, *J* = 8.6 Hz, 1H), 7.76 (d, *J* = 8.0 Hz, 1H), 7.57−7.48 (m, 4H), 7.37 (t, *J* = 7.5 Hz, 1H), 7.28−7.24 (m, 1H), 7.21−7.16 (m, 4H), 7.12 (d, *J* = 8.1 Hz, 2H), 2.45 (s, 3H), 2.33 (s, 3H), 1.61 (s, 3H); ^{13}C{^{1}H} NMR (101 MHz, $CDCl_3$): *δ* 143.2, 140.4, 140.3, 138.1, 136.0, 132.7, 132.4, 131.6, 130.5, 129.9 (2C), 129.3 (2C), 128.9, 128.5 (2C), 128.2, 127.7, 126.7 (2C), 124.5, 122.7, 121.9, 120.0, 119.2, 115.3, 114.6, 114.3, 21.5 (2C), 10.2 ppm.

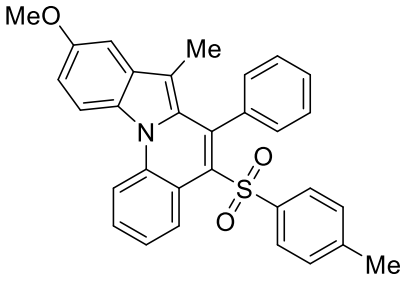
9-Fluoro-7-methyl-6-phenyl-5-tosylindolo[1,2-*a*]quinoline (IQSO$_2$R-V)



IQSO$_2$R-V

Prepared according to experimental procedure 3. Isolated by column chromatography (SiO_2, 10% EtOAc/hexanes) as a light-yellow crystal (167.8 mg, 70% yield); m.p. 225–227 °C (EtOAc/hexanes). ^{1}H NMR (400 MHz, $CDCl_3$): *δ* 8.80 (dd, *J* = 8.4 Hz, 1H), 8.41 (d, *J* = 8.4 Hz, 1H), 8.33 (dd, *J* = 9.3, 4.3 Hz, 1H), 7.59−7.55 (m, 3H), 7.48−7.35 (m, 4H), 7.32−7.22 (m, 4H), 7.13 (d, *J* = 8.1 Hz, 2H), 2.34 (s, 3H), 1.51 (s, 3H) ppm; ^{13}C{^{1}H} NMR (101 MHz, $CDCl_3$): *δ* 158.4 (d, *J* = 239.4 Hz, CF), 143.4 (2C), 140.3, 139.5, 135.8, 135.5, 132.5 (d, *J* = 9.3 Hz, C), 131.6, 130.1 (2C), 129.4 (2C), 129.2, 128.9 (d, *J* = 16.8 Hz, C), 128.4, 127.9, 127.8 (2C), 126.7 (2C), 122.9, 119.0, 115.7 (d, *J* = 9.1 Hz, CH), 115.1, 113.7 (d, *J* = 5.2 Hz, C), 113.1 (d, *J* = 26.3 Hz, CH), 104.4 (d, *J* = 22.9 Hz, CH), 21.5, 10.0 ppm; ^{19}F NMR (376 MHz, $CDCl_3$): *δ* -120.2 (s, 1F) ppm. IR (ATR): 3053, 2915, 2859, 1616, 1372, 1219, 1450, 1302, 1137, 1084, 830,749, 672, 654, 556, 527 cm^{-1}; HRMS (ESI-TOF) m/z: $[M + Na]^+$ calcd for $C_{30}H_{22}FNO_2SNa$, 502.1253; found, 502.1250.

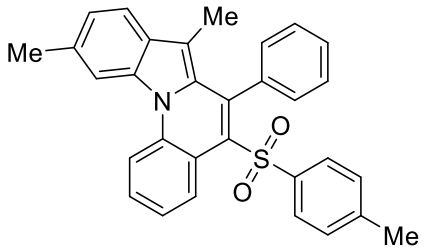
9-Methoxy-7-methyl-6-phenyl-5-tosylindolo[1,2-*a*]quinoline (IQSO$_2$R-VI)



IQSO$_2$R-VI

Prepared according to experimental procedure 3. Isolated by column chromatography (SiO_2, 10% EtOAc/hexanes) as a yellow solid (164.7 mg, 67% yield); m.p. 205–206 °C (EtOAc/hexanes). ^{1}H NMR (400 MHz, $CDCl_3$): *δ* 8.77 (dd, *J* = 8.4 Hz, 1H), 8.41 (d, *J* = 8.2 Hz, 1H), 8.29 (d, *J* = 9.2 Hz, 1H), 7.59−7.52 (m, 3H), 7.48−7.38 (m, 3H), 7.33−7.30 (m, 2H), 7.28−7.24 (m, 1H), 7.17−7.12 (m, 3H), 7.08 (d, *J* = 2.5 Hz, 1H), 3.91 (s, 3H), 2.34 (s, 3H), 1.53 (s, 3H); ^{13}C{^{1}H} NMR (101 MHz, $CDCl_3$): *δ* 155.3, 143.3, 140.5, 139.7, 135.8, 132.5, 130.8, 130.1 (2C), 129.4 (3C), 129.0, 128.3, 127.72 (3C), 126.69 (2C), 126.7 (2C), 122.5, 118.9, 115.6, 115.5, 115.1, 113.5, 100.0, 55.6, 21.5, 10.1 ppm; IR (ATR): 2911, 2829, 1615, 1487, 1448, 1370, 1303, 1234, 1144, 1087, 819, 701, 656, 553, 527 cm^{-1}; HRMS (ESI-TOF) m/z: $[M + Na]^+$ calcd for $C_{31}H_{25}NO_3SNa$, 514.1453; found, 514.1456.

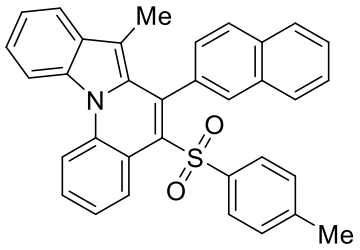
7,10-Dimethyl-6-phenyl-5-tosylindolo[1,2-*a*]quinoline (IQSO$_2$R-VII)



IQSO$_2$R-VII

Prepared according to experimental procedure 3. Isolated by column chromatography (SiO_2, 10% EtOAc/hexanes) as a yellow solid (180.7 mg, 76% yield); m.p. 156–158 °C (EtOAc/hexanes). ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.73 (dd, J = 8.4, 1.2 Hz, 1H), 8.46 (d, J = 8.4 Hz, 1H), 8.18 (s, 1H), 7.65 (d, J = 8.2 Hz, 1H), 7.58−7.52 (m, 3H), 7.41−7.37 (m, 3H), 7.31−7.28 (m, 2H), 7.24−7.21 (m, 2H), 7.13 (d, J = 8.1 Hz, 2H), 2.64 (s, 3H), 2.34 (s, 3H), 1.55 (s, 3H); ^{13}C{^{1}H} NMR (101 MHz, $CDCl_3$): δ 143.2, 140.4, 140.3, 136.0, 135.8, 134.8, 132.8, 130.0 (2C), 129.8, 129.6, 129.3 (2C), 128.7, 128.2, 127.7 (2C), 127.5, 127.3, 126.6 (2C), 123.9, 122.6, 119.6, 119.3, 115.3, 114.4, 114.2, 22.5, 21.4, 10.0 ppm; IR (ATR): 3053, 2917, 2859, 1598, 1445, 1365, 1304, 1145, 1084, 783, 698, 655, 594, 561, 530 cm^{-1}; HRMS (ESI-TOF) m/z: $[M + H]^+$ calcd for $C_{31}H_{26}NO_2S$, 476.1684; found, 476.1675.

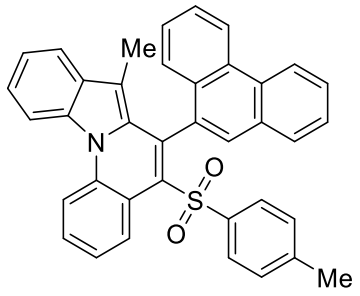
7-Methyl-6-(naphthalen-2-yl)-5-tosylindolo[1,2-*a*]quinoline (IQSO$_2$R-VIII)



IQSO$_2$R-VIII

Prepared according to experimental procedure 3. Isolated by column chromatography (SiO_2, 10% EtOAc/hexanes) as a yellow solid (153.5 mg, 60% yield). The NMR data obtained for **IQSO$_2$R-VIII** are in agreement with previously reported literature values.[2] ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.87 (dd, J = 8.3, 1.0 Hz, 1H), 8.51 (d, J = 8.4 Hz, 1H), 8.41 (d, J = 8.6 Hz, 1H), 7.92 (d, J = 7.8 Hz, 1H), 7.84 (d, J = 8.4 Hz, 1H), 7.74−7.72 (m, 2H), 7.64 (s, 1H), 7.61−7.50 (m, 4H), 7.45−7.31 (m, 5H), 6.96 (d, J = 8.2 Hz, 2H), 2.27 (s, 3H), 1.48 (s, 3H); ^{13}C{^{1}H} NMR (101 MHz, $CDCl_3$): δ 143.3, 140.3, 139.6, 136.0, 133.0, 132.9, 132.6, 132.4, 131.6, 130.4, 129.3, 129.2, 129.15 (2C), 129.06, 128.4, 128.2, 127.83, 127.81, 127.2, 126.7 (2C), 126.5, 126.2, 124.5, 122.8, 122.0, 120.0, 119.4, 115.4, 114.6, 114.1, 21.4, 10.4 ppm.

7-Methyl-6-(phenanthren-9-yl)-5-tosylindolo[1,2-*a*]quinoline ($IQSO_2R$-IX)



$IQSO_2R$-IX

Prepared according to experimental procedure 3. Isolated by column chromatography (SiO_2, 10% EtOAc/hexanes) as a yellow solid (154.5 mg, 55% yield); m.p. 235–237 °C (EtOAc/hexanes). ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.99 (dd, J = 8.3, 1.2 Hz, 1H), 8.71 (d, J = 8.3 Hz, 1H), 8.65 (d, J = 8.3 Hz, 1H), 8.58 (d, J = 8.2 Hz, 1H), 8.45 (d, J = 8.6 Hz, 1H), 7.86 (d, J = 7.3 Hz, 1H), 7.76−7.72 (m, 2H), 7.68−7.56 (m, 4H), 7.53−7.45 (m, 2H), 7.41−7.37 (m, 1H), 7.35−7.30 (m, 4H), 6.68 (d, J = 8.1 Hz, 2H), 2.13 (s, 3H), 1.21 (s, 3H); ^{13}C{^{1}H} NMR (101 MHz, $CDCl_3$): δ 143.1, 139.2 , 136.6, 136.4, 132.4, 132.1, 131.9, 131.6, 131.5, 131.0, 130.8, 130.0, 129.9, 129.31, 129.29, 129.1, 128.6 (2C), 127.9, 127.4, 127.2 (2C), 126.9, 126.7 (2C), 126.6, 124.4, 122.9, 122.7, 122.6, 121.9, 120.0, 119.3, 115.5, 114.7, 113.6, 21.3, 9.8 ppm; IR (ATR): 3054, 2908, 2860, 1438, 1448, 1375, 1263, 1305, 1142, 729, 663, 585, 526 cm^{-1}; HRMS (ESI-TOF) m/z: $[M + Na]^+$ calcd for $C_{38}H_{27}NO_2SNa$, 584.1660; found, 584.1663.

4. EGFR-TK Inhibition Assay

The EGFR tyrosine kinase activity in the series of **$IQSCF_3$s** and **$IQSO_2R$s** were screened using ADP-Glo™kinase Assay.[4] In brief, EGFR enzyme (1.25 ng/µl) and inhibitors were added to a well plate, followed by a mixture of 25 µM ATP and 12.5 µM poly (Glu-Tyr), and incubated for 1 h at room temperature. The ADP Glo reagent was added and incubated for 40 min. The kinase detection reagent was incubated for 30 min at room temperature. The ATP was detected by measuring the luminescence using a Synergy Neo2 Multi-Mode Microplate Reader (BioTek Instruments, VT, USA). The relative inhibition (%) was calculated compared to the control with no inhibitor, as shown in the equation.

$$\% \text{ relative inhibition} = \frac{[(\text{positive} - \text{negative}) \times (\text{sample} - \text{negative})]}{(\text{positive} - \text{negative})} \times 100$$

5. Cytotoxicity in Cancer Cell line

A549 Lung adenocarcinoma, SCC head and neck squamous cells, LS174T carcinoma colorectal cancer cells, and Vero cell lines were obtained from American Type Culture Collection (Manassas, VA, USA). The A549 cells were maintained by a Kaighn's Modification of Ham's F-12 Medium (F-12k Medium) included with 10% FBS and 100 U/mL penicillin (Life Technologies, Carlsbad, CA, USA). The SCC cells were maintained by a Dulbecco's Modified Eagle Medium/Ham's F-12 (DMEMF-12) with 10% FBS and 100 U/mL penicillin. The LS174T cells were maintained by an RPMI-1640 containing 10% FBS and 100 U/mL penicillin. The Vero cells were maintained by an Eagle's Minimum Essential Medium (EMEM) with 10% FBS and 100 U/mL penicillin. All cells were kept at 37 ^{o}C under humidified 95% O_2 with 5% CO_2 atmosphere.

Cell viability of A549, SCC, LS174T and Vero cells was assessed using MTT assays. Briefly, A549 cells (5,000 cells/well), SCC cells (10,000 cells/well), LS174T cells (7,000 cells/well) and Vero cells (4,000 cells/well) were seeded and incubated at 37 ^{o}C for 24-hrs in 96 well plates. After 24-hrs incubation, the cells were treated with different concentrations of second-generation EGFR-TKIs drug (afatinib), **IQSCF$_3$s** and **IQSO$_2$Rs** at 37 ^{o}C for 72-hrs incubation. The MTT solution (5 mg/ml) was added to the A549, SCC, LS174T and Vero cells and kept for 2-hrs incubation at 37 ^{o}C. The reaction was stopped by DMSO (100 μL/well). Colorimetric quantification was measured with absorbance at 570 nm using a Synergy Neo2 Multi-Mode Microplate Reader (BioTek Instruments, VT, USA).

6. Physicochemical Property Prediction

Physicochemical features, including the number of hydrogen bond donors, hydrogen bond acceptors, and drug-likeness, play a crucial role in the drug discovery and development process. In this study, we calculated these properties for four compounds (**IQSO$_2$R-I, IQSO$_2$R-V, IQSO$_2$R-VI,** and **IQSO$_2$R-VII**) using the web-based application SwissADME (www.swissadme.ch/).

7. Molecular Dynamic Simulation

The crystal structure of erlotinib in complex with the wild-type EGFR-TK was obtained from the Protein Data Bank (PDB ID: 1M17). The **IQSO$_2$R-I**/EGFR-TK complex was derived molecular docking using GOLD program[5] according to our study on this system.[6] The best docking pose (highest GOLD fitness score) of **IQSO$_2$R-I** binding to EGFR-TK was considered as the starting structure for all-atom molecular dynamics simulation with three independent runs using different initial velocities. The system was simulated under periodic boundary conditions using the isothermal-isobaric (NPT) ensemble, with a temperature of 310 K and a pressure of 1 atm as per previous studies.[7,8] The AMBER ff14SB force field and the generalized AMBER force field version 2 (GAFF2) were employed to handle the bonded and non-bonded interaction parameters for the protein and ligand, respectively. The system was solvated in TIP3P water model. The chloride ions were randomly introduced to neutralize the overall charge of the system. The hydrogen atoms and water molecules were subjected to energy minimization using 500 steps of steepest descent followed by 1500 steps of conjugated gradient methods, while the remaining molecules were kept fixed. The protein-ligand complex (constrained solvents) and the entire complex system were then subjected to further minimization,

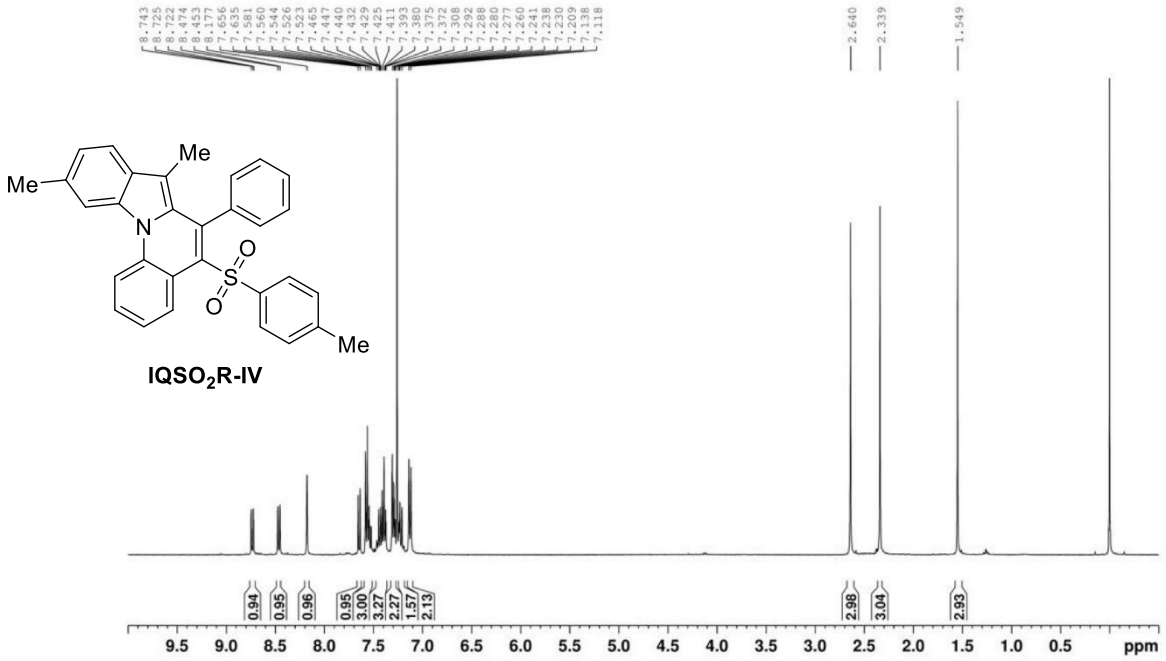
following the same procedure. Electrostatic interactions were handled using the particle mesh Ewald summation approach, and hydrogen atoms were constrained using the SHAKE algorithm. The temperature gradually increased from 10 to 310 K using a Langevin thermostat with a collision frequency of 2 ps^{-1}, while the pressure was controlled using the Berendsen barostat. MD simulations were conducted for 500 ns with a time step increment of 2 fs. The MD outputs were analyzed using the cpptraj module, and the per-residue decomposition energy ($\Delta G_{bind,residue}$) was computed using MM/PBSA.py in AMBER20. The interactions of the potent EGFR inhibitor complexed with EGFR-TK were visualized using LigandScout 4.4.9 software.[9]

8. References

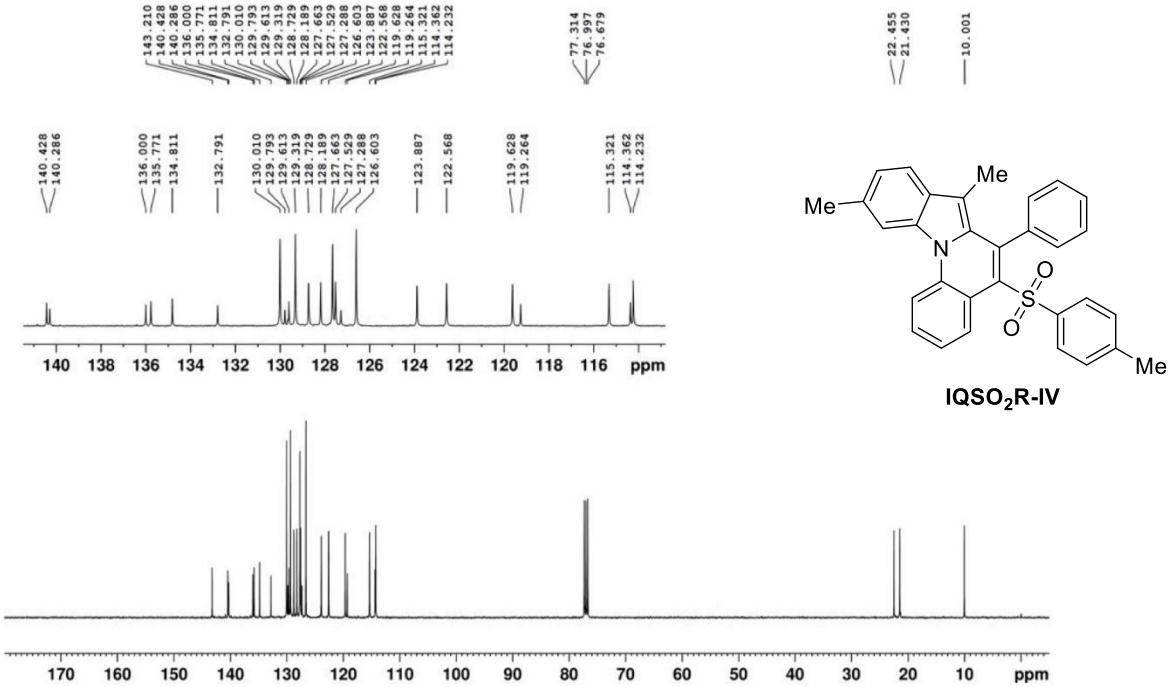
1. T. Uppalabat, N. Hassa, N. Sawektreeratana, P. Leowanawat, P. Janthakit, P. Nalaoh, V. Promarak, D. Soorukram, V. Reutrakul and C. Kuhakarn, Cascade Oxidative Trifluoromethylthiolation and Cyclization of 3-Alkyl-1-(2-(alkynyl)phenyl)indoles. *J. Org. Chem.*, 2023, **88**, 5403.
2. K. Sun, X. L. Chen, Y. L. Zhang, K. Li, X. Q. Huang, Y. Y. Peng, L. B. Qu and B. Yu, Metal-Free Sulfonyl Radical-Initiated Cascade Cyclization to Access Sulfonated Indolo[1,2-*a*]quinolines. *Chem. Commun.* **2019**, *55*, 12615.
3. Zhang, J. Yuan, G. Huang, C. Ma, J. Yang, L. Yang, Y. Xiao and L. Qu, Visible-Light-Induced Intramolecular Tandem Cyclization of Unactivated Indoloalkynes for the Synthesis of Sulfonylated and Selenylated Indolo[1,2-*a*]quinolines. *J. Org. Chem.* **2023**, *88*, 11712.
4. K. Hengphasatporn, T. Aiebchun, P. Mahalapbutr, A. Auepattanapong, O. Khaikate, K. Choowongkomon, C. Kuhakarn, J. Meesin, Y. Shigeta and T. Rungrotmongkol, Sulfonylated Indeno[1,2-*c*]quinoline Derivatives as Potent EGFR Tyrosine Kinase Inhibitors. *ACS Omega.* **2023**, *8*, 19645.
5. M. L. Verdonk, J. C. Cole, M. J. Hartshorn, C. W. Murray and R. D. Taylor, Improved protein–ligand docking using GOLD. *Proteins* **2003**, *52*, 609.
6. T. Aiebchun, P. Mahalapbutr, A. Auepattanapong, O. Khaikate, S. Seetaha, L. Tabtimmai, C. Kuhakarn, K. Choowongkomon and T. Rungrotmongkol, Identification of Vinyl Sulfone Derivatives as EGFR Tyrosine Kinase Inhibitor: In vitro and In silico Studies. *Molecules* **2021**, *26*, 2211.
7. D. Todsaporn, A. Zubenko, V. Kartsev, T. Aiebchun, P. Mahalapbutr, A. Petrou, A. Geronikaki, L. Divaeva, V. Chekrisheva, I. Yildiz, K. Choowongkomon and T. Rungrotmongkol, Discovery of Novel EGFR Inhibitor Targeting Wild-type and Mutant Forms of EGFR: In silico and In vitro Study. *Molecules* **2023**, *28*, 3014.
8. Todsaporn, D.; Mahalapbutr, P.; Poo-Arporn, R. P.; Choowongkomon, K.; Rungrotmongkol, T. Structural dynamics and kinase inhibitory activity of three generations of tyrosine kinase inhibitors against wild-type, L858R/T790M, and L858R/T790M/C797S forms of EGFR. *Comput. Biol. Med.* **2022**, *147*, 105787.
9. G. Wolber and T. Langer, LigandScout: 3-D Pharmacophores Derived from Protein-Bound Ligands and Their Use as Virtual Screening Filters**,** *J. Chem. Inf. Model.* 2005, **45**, 160.

9. NMR Spectra

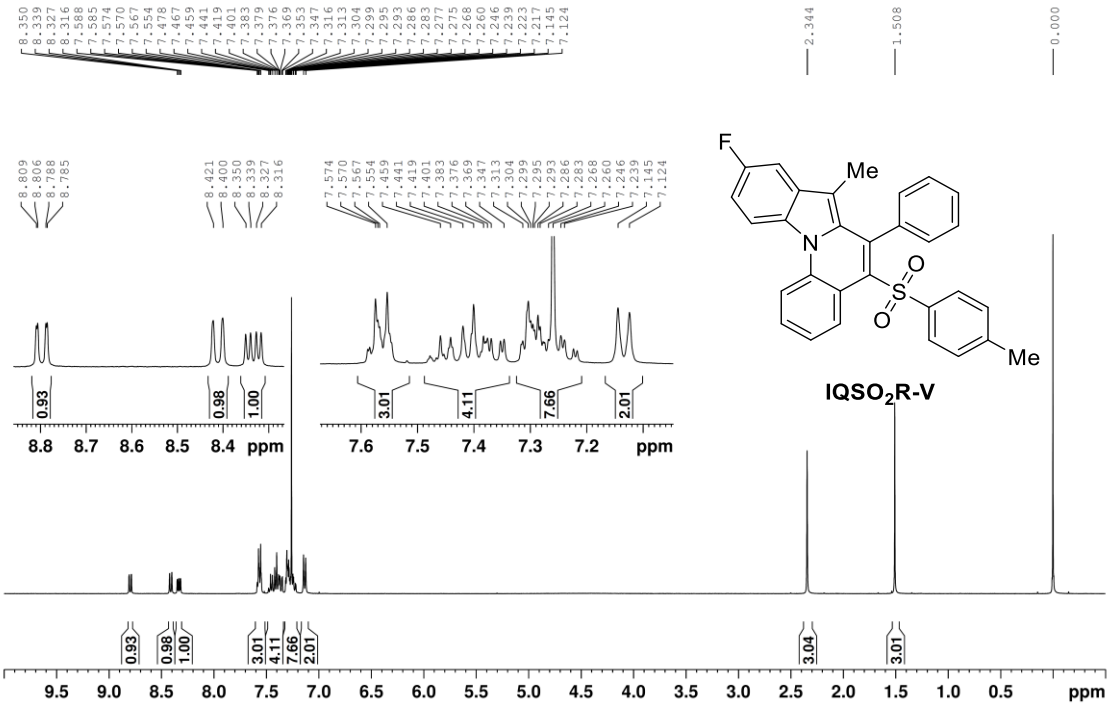
^{1}H NMR (400 MHz) of IQSO$_2$R-IV



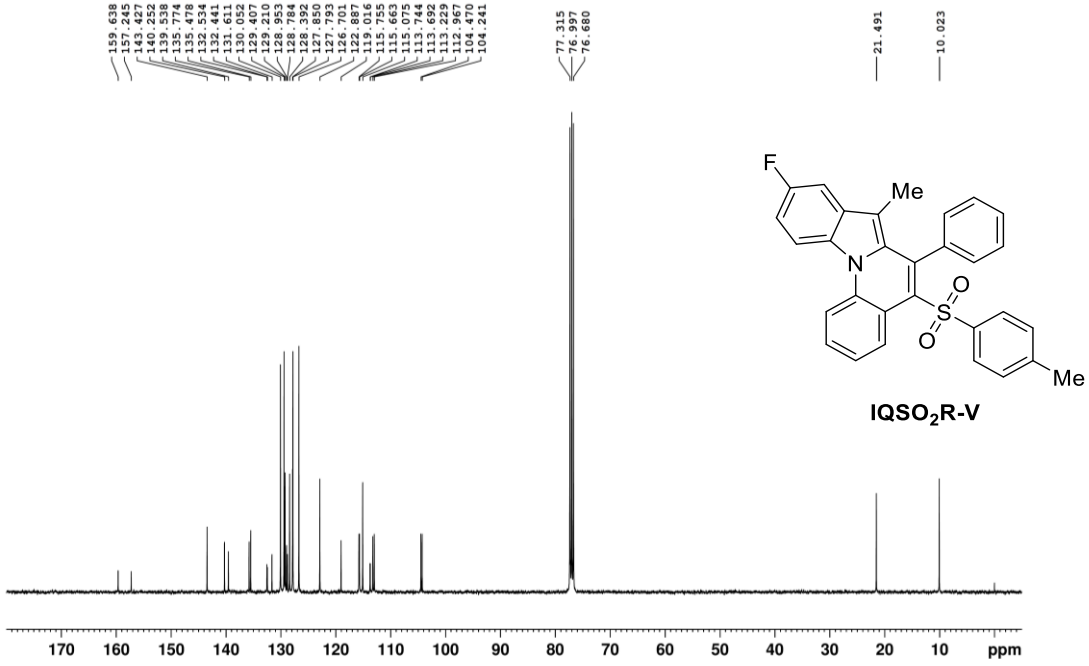
^{13}C NMR (101 MHz) of IQSO$_2$R-IV



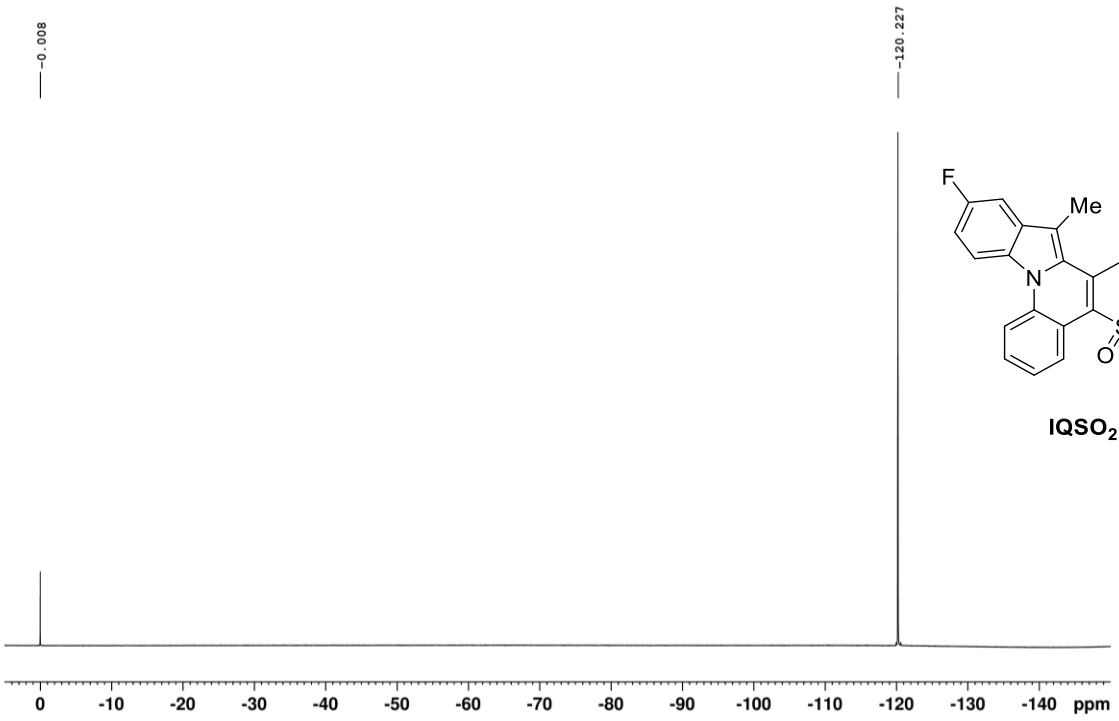
^{1}H NMR (400 MHz) IQSO$_2$R-V



^{13}C NMR (101 MHz) of IQSO$_2$R-V



^{19}F NMR (376 MHz) of IQSO$_2$R-V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Me
N
O
S
O
Me
0
-10
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-50
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-70
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-110
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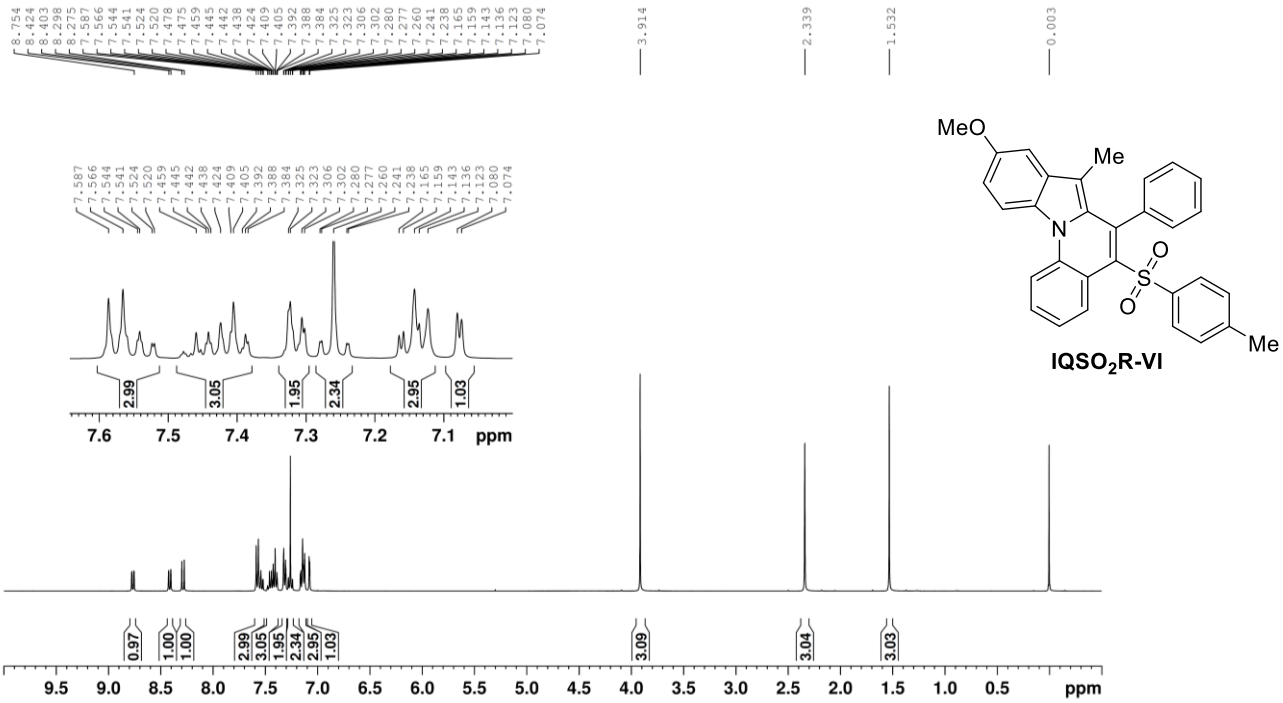
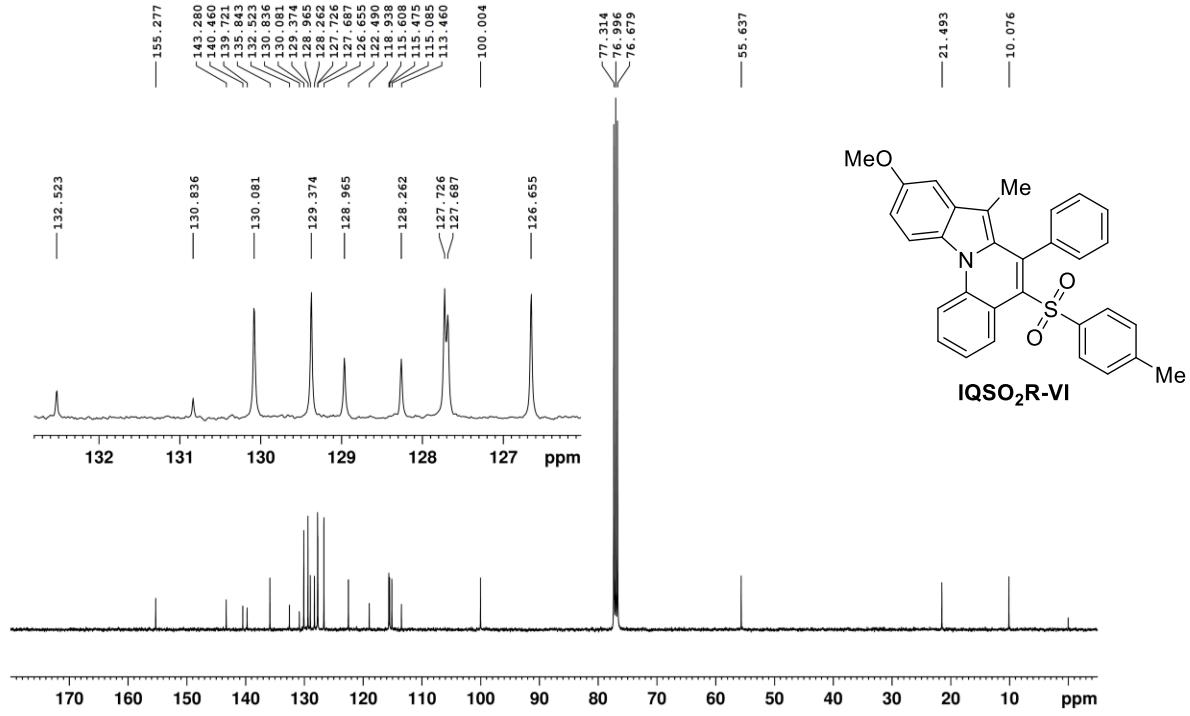
-130
-140
ppm

IQSO$_2$R-V

^{1}H NMR (400 MHz) of IQSO$_2$R-VI



^{13}C NMR (101 MHz) of IQSO$_2$R-VI



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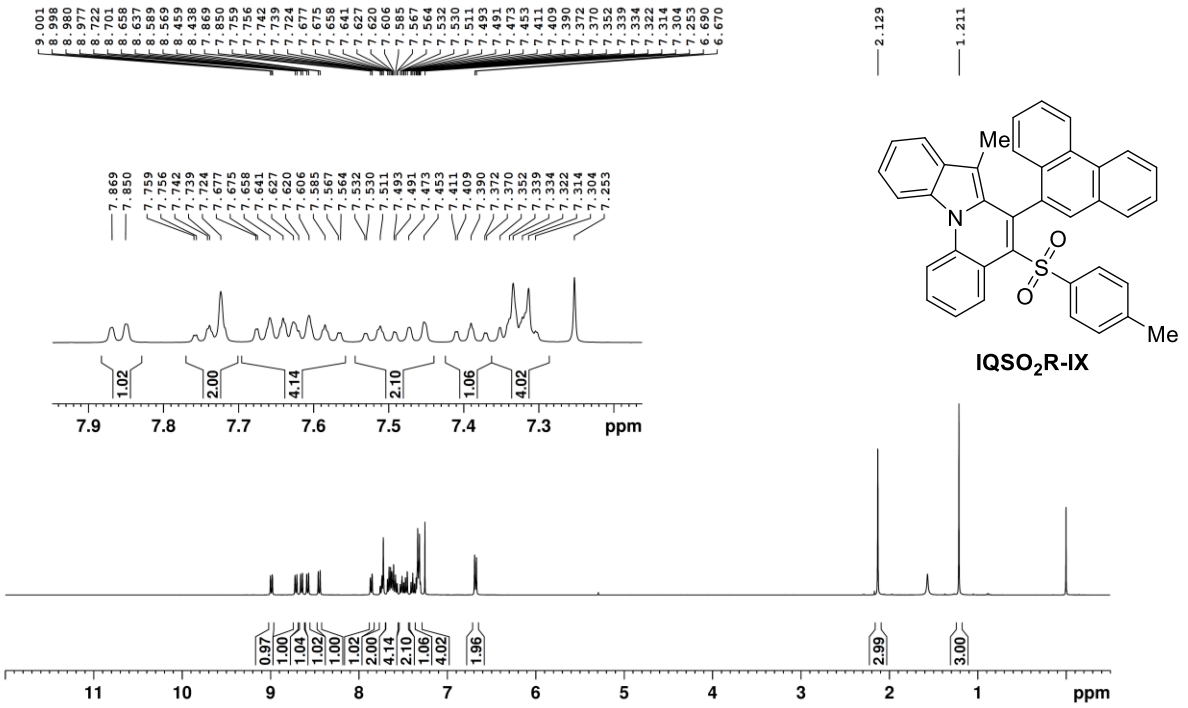

^{13}C NMR (101 MHz) of IQSO$_2$R-IX