Supporting Information

A Transmetalation Reaction Enables the Synthesis of [^{18}F]5-Fluorouracil from [^{18}F]Fluoride for Human PET Imaging

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Materials and Methods

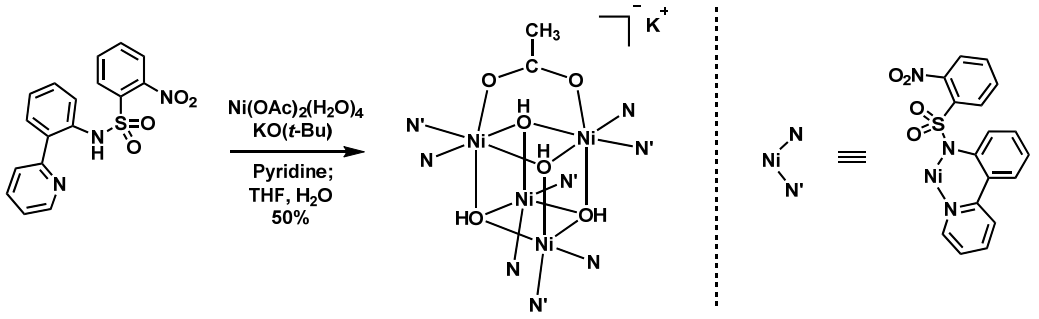
Thin layer chromatography (TLC) was performed using EMD TLC plates pre-coated with 250 μm thickness silica gel 60 F_{254} plates and visualized by fluorescence quenching under UV light. Flash chromatography was performed using silica gel (230 – 400 mesh) purchased from Silicycle Inc. NMR spectra were recorded on either a Varian Unity/Inova 600 spectrometer operating at 600 MHz for ^{1}H acquisitions, a Varian Unity/Inova 500 spectrometer operating at 500 MHz and 125 MHz for ^{1}H and ^{13}C acquisitions, respectively, or a Varian Mercury 400 spectrometer operating at 400 MHz, 100 MHz, and 375 MHz for ^{1}H, ^{13}C, and ^{19}F acquisitions, respectively. Chemical shifts for ^{1}H and ^{13}C acquisitions are reported in ppm with the solvent resonance as the internal standard (^{1}H: $CDCl_3$, δ 7.26; pyridine-d_5, δ 8.74), (^{13}C: $CDCl_3$, δ 77.16; pyridine-d_5, δ 135.91). Chemical shifts for ^{19}F acquisitions are reported in ppm with PhF as the external standard (^{19}F: $CDCl_3$, δ –113.15). Data are reported as follows: s = singlet, br = broad, d = doublet, t = triplet, q = quartet, m = multiplet; coupling constants in Hz; integration. All deuterated solvents were purchased from Cambridge Isotope Laboratories. High-resolution mass spectra were obtained using an Agilent ESI-TOF (6210) mass spectrometer or a Bruker q-TOF Maxis Impact mass spectrometer. FTIR spectra were obtained using a Bruker ALPHA Platinum ATR spectrometer. Concentration under reduced pressure was performed by rotary evaporation at 25–30 ºC at appropriate pressure. Purified compounds were further dried under high vacuum (0.2 Torr). Yields refer to purified and spectroscopically pure compounds unless otherwise indicated.

Dry pyridine used in nickel(II) aryl synthesis was distilled from CaH_2 under N_2 (1 atm). Anhydrous pyridine (99.8%, <0.003% water, Aldrich Sure/Seal™) used in the synthesis of **1** was purchased from Sigma-Aldrich. Anhydrous THF (≤0.05% water) was purchased from BDH. Anhydrous diethyl ether (≤0.03% water) was purchased from EMD. Dry, degassed dioxane and DMSO were obtained by N_2 sparging of anhydrous dioxane and DMSO purchased from Sigma-Aldrich. Potassium acetate was dried in a glass flask under high-vacuum with a flame. All other commercial chemicals were used as received. Nickel(II) acetate tetrahydrate (≥99.0%), benzothiophene-3-boronic acid (≥95%), 5-bromobenzo[c] [1,2,5]thiadiazole (95%), 5-bromo-2-methoxyphenylboronic acid, 3-bromobenzo[b]thiophene (95%), 2.0M isopropylmagnesium bromide solution in THF, and 4-bromo-2-fluoroanisole (**6a**) (97%) were purchased from Sigma-Aldrich. 2,4-Di-tert-butoxypyrimidine-5-boronic acid (98%) was purchased from Combi-Blocks. Palladium dichloride-bis(diphenylphosphino) ferrocene-dichloromethane complex, 5-fluorouracil, and 5-bromobenzo[c][1,2,5]thiadiazole (**6d**) were purchased from Oakwood. 6-Chloropyridine-3-boronic acid (96%) was purchased from Frontier Scientific. 5-Bromo-2-(2-methyl-2H-tetrazol-5-yl)pyridine (98%) was purchased from Astatech. 2-Nitro-N-(2-(pyridin-2-yl)phenyl)benzenesulfonamide was prepared from 2-bromopyridine in two steps.[1] The oxidant (1,1'-(phenyl-λ^3-iodanediyl)bis(4-methoxypyridinium)bis(trifluoromethanesulfonate)) was prepared from iodobenzenediacetate.[1] 3-Bromo-5-(pyridin-2-ylethynyl)benzonitrile[2] was a gift from Dr. Nickeisha Stephenson (Vasdev Laboratory, Harvard Medical School and Massachusetts General Hospital). 2,4-Dichloro-5-fluoropyrimidine was prepared from 5-fluorouracil.[3]

Experimental Data

Experimental Procedures and Compound Characterization

Synthesis of nickel(II) hydroxide cubane 1



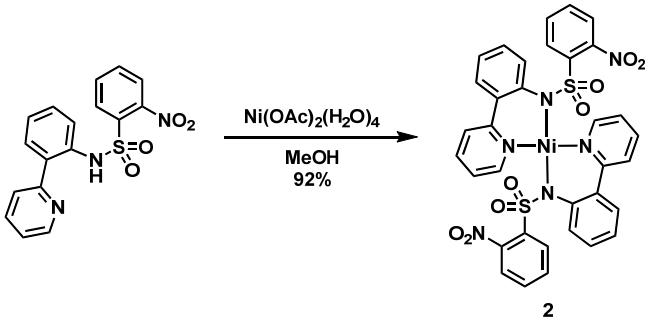
To a 1 L round-bottomed flask were added nickel(II) acetate tetrahydrate (2.80 g, 11.3 mmol, 1.00 equiv.) and a Teflon-coated stirbar. The flask was fitted with a septum, and the headspace was filled with nitrogen. Anhydrous pyridine (114 mL) was added, and a blue solution was observed after mixing. To this solution was added 2-nitro-N-(2-(pyridin-2-yl)phenyl)benzenesulfonamide[1] (4.00 g, 11.3 mmol, 1.00 equiv., as a solution in 166 mL anhydrous pyridine) by cannula over 3 minutes, and a green-blue solution was observed. To this solution was added potassium *tert*-butoxide (2.53 g, 22.5 mmol, 2.00 equiv., as a solution in 80 mL anhydrous pyridine) by cannula over 5 minutes. A yellow-green solution with colorless precipitate was observed, which was stirred at 23 ºC for 45 minutes, before being concentrated in vacuo (by rotary evaporation at 60 ºC until all liquid pyridine was removed, and then under high vacuum at 23 ºC) to give a mixture of green and orange solids. These solid residues were triturated with anhydrous THF (130 mL) in order to dissolve the green solid. The mixture was filtered through celite on a glass frit, which was then rinsed with anhydrous THF (2 × 20 mL). The THF filtrates were combined to give a dark green solution that was treated dropwise with H_2O (5.0 mL) over 30 minutes, with magnetic stirring, which caused a light green solid to precipitate. The solid was collected by filtration on a glass frit, rinsed with THF (2 × 20 mL), and dried in vacuo (0.2 Torr, 50 ºC, 40 minutes; then 0.2 Torr, 150 ºC, 2 hours) to afford 2.66 g of the title compound (as a solvate with 4 water molecules) as a green solid (50% yield).

NMR Spectroscopy: ^{1}H NMR (600 MHz, pyridine-d_5, 23 ºC, δ): 46.1, 45.2, 44.8, 44.5, 43.0, 41.8, 40.9, 40.7, 39.6, 39.4, 39.2, 37.4, 36.1, 33.6, 33.1, 32.2, 30.4, 29.4, 24.4, 22.0–19.0, 20.3, 19.9, 19.6, 18.3, 17.9, 17.7, 17.5, 16.7, 16.0. 14.8, 14.7, 14.4, 14.1, 13.8, 13.5, 13.1, 12.6, 11.9, 11.4, 10.9, 10.7, 10.6, 10.2, 9.9, 8.3, 8.1, 7.5, 6.9, 6.6, 6.3, 6.2, 6.1, 6.0, 5.8, 5.4, 1.9, 1.0, 0.5, 0.2, –0.2, –0.4, –0.7, –1.0, –1.9, –2.0, –2.6, –3.6, –4.0, –4.6, –5.0, –5.5, –6.5. Due to gradual decomposition in solution, ^{13}C NMR analysis was not performed. Anal: calcd for $C_{70}H_{55}N_{12}S_4O_{22}KNi_4(H_2O)_4$: C, 44.47; H, 3.36, N, 8.89; found: C, 44.63; H, 3.07; N, 8.77. IR

(neat, ν, cm^{-1}): 1594 (w), 1575 (w), 1535 (s), 1489 (m), 1476 (w), 1428 (m), 1367 (m), 1275 (m), 1234 (m), 1147 (s), 1129 (s), 1117 (s), 1061 (m), 972 (s), 852 (w), 824 (m), 753 (s), 730 (s), 651 (m), 630 (w), 593 (s), 562 (s), 528 (m), 434 (m).

Crystals for X-ray analysis were obtained as follows. The mixture of green and orange solids obtained after pyridine evaporation (by the above procedure) (20.0 mg) was treated with anhydrous THF (2.0 mL). To this mixture was then added anhydrous diethyl ether (1.8 mL) dropwise. The resulting mixture was filtered through celite into a scintillation vial that was then capped with a septum. A needle was inserted into the septum, so that water from wet air could slowly diffuse into the solution. Green crystals were obtained. For crystallography data, see the X-ray Crystallography section.

Synthesis of nickel(II) complex 2



2

To a 1 dram scintillation vial were added nickel(II) acetate tetrahydrate (35.0 mg, 0.141 mmol, 1.00 equiv.), methanol (2.5 mL), and a Teflon stirbar. The mixture was sonicated to afford a homogeneous light green solution. To this solution was added 2-nitro-N-(2-(pyridin-2-yl)phenyl)benzenesulfonamide[1] (100. mg, 0.281 mmol, 2.00 equiv.), and the vial was sealed with a Teflon-lined cap. An orange precipitate formed. The mixture was stirred at 23 ºC for 3 days and 15 hours, and was then filtered on a glass frit. The collected solid was rinsed with methanol and dried in vacuo to afford 99.4 mg of the title compound (92% yield).

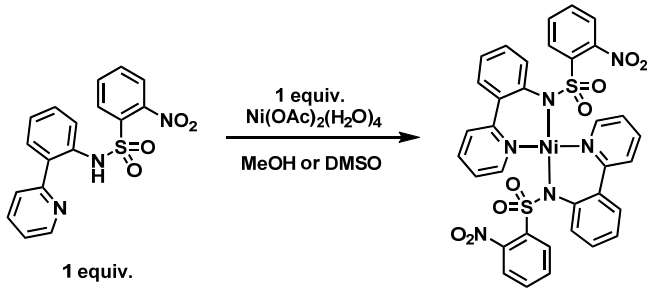
Complex **2** was not characterized by NMR, due to insolubility.

Anal: calcd for $C_{34}H_{24}N_6NiO_8S_2$: C, 53.21; H, 3.15, N, 10.95; found: C, 53.25; H, 3.19; N, 10.83.

Crystals of **2** for X-ray analysis were obtained by slow formation from a soluble ligated nickel(II) complex, prepared as follows. To a 1-dram scintillation vial were added nickel(II) acetate tetrahydrate (14.0 mg, 56.3 μmol, 1.00 equiv.), a Teflon stirbar, and dry pyridine (0.8 mL) to afford, after mixing, a blue solution that was then treated with potassium *tert*-butoxide (0.200 mL of a solution prepared from 34.4 mg KO*t*-Bu and 1.05 mL of dry pyridine, 1 equiv.). After stirring at 23 ºC for 9 minutes, 2-nitro-N-(2-(pyridin-2-yl)phenyl)benzenesulfonamide (20.0 mg, 56.3 μmol, 1.00 equiv., as a solution in 0.60 mL dry pyridine) was added, followed by additional potassium *tert*-butoxide (0.200 mL of the pyridine solution, 1 equiv.). The vial, sealed with a Teflon-lined cap, was heated at 90 ºC for 5 minutes. After cooling to 23 ºC, the solvent was

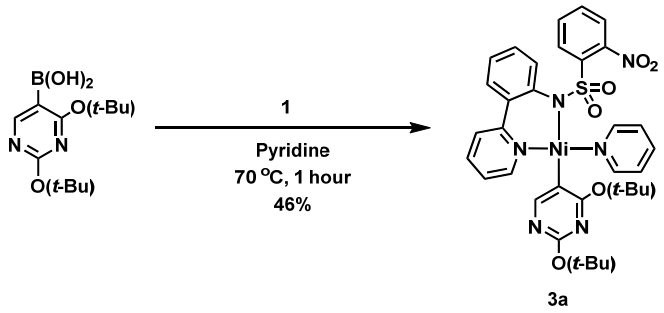
removed in vacuo. The residue was treated with anhydrous THF to give a mixture that was filtered through celite, and the filtrate was concentrated in vacuo. A portion of the resulting solid was dissolved in CD_2Cl_2 (in an NMR tube with plastic slip-on cap) to give a green solution that, over time, deposited orange crystals suitable for X-ray analysis. For crystallography data, see the X-ray Crystallography section.

Formation of nickel(II) complex 2 in MeOH and DMSO



To a 20 mL scintillation vial was added nickel(II) acetate tetrahydrate (31.1 mg, 0.125 mmol, 1.00 equiv.), a Teflon stirbar, and 5.00 mL of solvent (MeOH or DMSO), to afford a homogeneous green solution upon mixing. To the magnetically-stirred solution at 23 ºC was added 2-nitro-N-(2-(pyridin-2-yl)phenyl)benzenesulfonamide[1] (44.4 mg, 0.125 mmol, 1.00 equiv.). An orange precipitate formed (after 10 seconds in MeOH solvent, and after 5 minutes in DMSO solvent).

Synthesis of nickel(II) aryl complex 3a



To a 250 mL 2-necked round-bottomed flask were added nickel cube **1** (185 mg, 98.1 μmol, 0.250 equiv.), 2,4-di-tert-butoxypyrimidine-5-boronic acid (105 mg, 0.392 mmol, 1.00 equiv.), and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a septum, and its atmosphere was evacuated and backfilled with nitrogen three times. Dry pyridine (15.5 mL) was added by syringe, and the septum was replaced with a glass stopper. The mixture was heated with stirring at 70 ºC for 1 hour. Once cooled to 23 ºC, hexanes (155 mL) was added with stirring. The mixture was filtered through celite on a glass frit, which was then rinsed with hexanes (20 mL). The hexanes filtrate was discarded, and the collected residue was dissolved in

DCM/pyridine 95:5 (v/v), passed through the frit, and concentrated in vacuo to give a residue. The residue was quickly purified by chromatography on silica gel/K_2CO_3 9:1 (w/w), eluting with DCM/pyridine 95:5 (v/v). The yellow/orange band was collected and concentrated in vacuo to remove DCM, to about 0.5 mL. Hexanes (0.8 mL) was added slowly, dropwise with mixing until turbid, and the sides of the vessel were scratched with a metal spatula to induce crystallization. More hexanes (9.2 mL) was added dropwise with mixing, and the resulting solid was triturated with a metal spatula, centrifuged, and the supernatant was decanted. The remaining solid was triturated with hexanes (10 mL), the mixture was centrifuged, and the supernatant was decanted. The solid was dried in vacuo (0.2 Torr, 23 ºC, 17 hours) to afford 139 mg of the title compound (as a solvate with 0.75 pyridine molecules) as a yellow solid (46% yield).

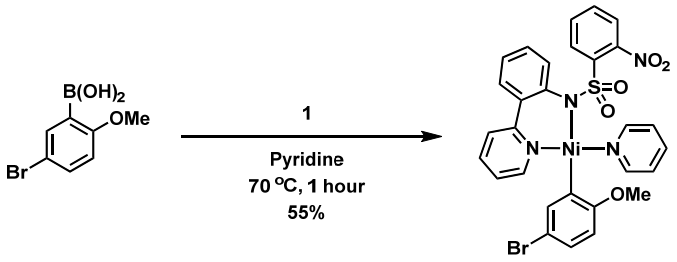
NMR Spectroscopy: ^{1}H NMR (600 MHz, pyridine-d_5, 23 ºC, δ): 8.86 (br s, 1H), 8.64 (br s, 1H), 7.85 (d, J = 8.0 Hz, 1H), 7.81 (d, J = 7.6 Hz, 1H), 7.66–7.61 (m, 1H), 7.47 (dd, J = 7.6, 7.6 Hz, 1H), 7.43 (d, J = 7.4 Hz, 1H), 7.37–7.28 (m, 2H), 7.15–7.10 (m, 1H), 6.98–6.87 (br m, 1H), 6.59 (br s, 1H) 1.53 (s, 9H), 1.48 (br s, 9H). ^{13}C NMR (125 MHz, pyridine-d_5, 23 ºC, δ): 175.0 (br), 164.1, 160.9 (br), 156.3, 152.4 (br), 148.1, 141.8, 138.4, 137.4, 136.9, 132.0, 131.3, 131.2, 130.9, 129.5, 129.2, 125.1, 123.62, 123.59, 123.0, 79.9 (br), 78.7, 29.1, 29.0. Anal: calcd for $C_{34}H_{36}N_6NiO_6S(C_5H_5N)_{0.75}$: C, 58.52; H, 5.17, N, 12.20; found: C, 58.74; H, 5.22; N, 12.05. HRMS (ESI-TOF) (m/z): calcd for $C_{29}H_{32}N_5NiO_6S$ $[M - \text{pyridine} + H]^+$, 636.1427; found, 636.1438.

Quantification of solvate pyridine stoichiometry was not trivial. In NMR solvents other than pyridine-d_5, complex **3a** condenses with itself to form unidentified oligomeric species plus unligated pyridine. In pyridine-d_5 solvent, the residual pyridine solvent peaks are too intense compared to pyridine-H_5 from the analytical sample to permit unambiguous quantification. Therefore the following degradation experiment was performed to completely liberate the ligands bound to nickel, so that the relative amount of pyridine present could be directly observed and quantified. To a 4 mL 1-dram vial was added nickel complex **3a** (2.2 mg), a Teflon-coated stirbar, and a large excess of KCN in CD_3OD (0.420 mL of a 0.292M solution of KCN in CD_3OD). The vial was sealed with a Teflon-lined cap, and heated at 60 ºC for 1 hour to afford a light yellow homogeneous solution. Once cooled to 23 ºC, the solution was analyzed by ^{1}H NMR spectroscopy, and the ratio of pyridine to the potassium salt of the pyridylsulfonamide ligand was determined by integration to be 1.75 : 1.00. Since **3a** already contains 1 pyridine covalently bound to nickel, the balance of 0.75 pyridine molecules is attributed as solvate pyridine molecules contained within the lattice of the solid. This assignment is consistent with the results of CHN elemental analysis (*vide supra*).

Crystals of **3a** for X-ray analysis were obtained as follows. To a 1-dram (4 mL) scintillation vial were added nickel(II) aryl complex **3a** (7.8 mg) and pyridine (0.410 mL), to give a yellow solution. This vial was placed in a 20 mL scintillation vial containing hexanes (5 mL), and the larger vial was sealed, so that crystallization by vapor diffusion would occur. After 2 days and 16 hours at 23 ºC, yellow crystals had formed. For crystallography data, see the X-ray

Crystallography section.

Synthesis of nickel(II) aryl complex 3b



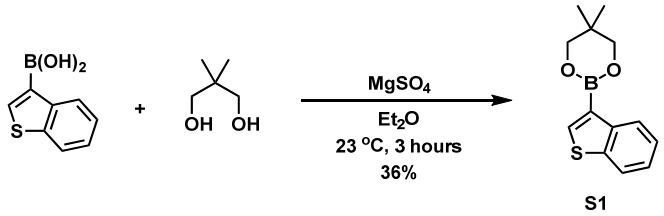
3b

To a 250 mL 2-necked round-bottomed flask were added nickel cube **1** (142 mg, 75.0 μmol, 0.250 equiv.), 5-bromo-2-methoxyphenylboronic acid (69.0 mg, 0.300 mmol, 1.00 equiv.), and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a septum, and its atmosphere was evacuated and backfilled with nitrogen three times. Dry pyridine (12 mL) was added by syringe, and the septum was replaced with a glass stopper and Teflon sleeve. The mixture was heated with stirring at 70 ºC for 1 hour. Once cooled to 23 ºC, hexanes (120 mL) was added with stirring. The mixture was filtered through celite on a glass frit, which was then rinsed with hexanes (20 mL). The hexanes filtrate was discarded, and the collected residue was dissolved in DCM/pyridine 95:5 (v/v), passed through the frit, and concentrated in vacuo to a residue that was purified by chromatography on silica gel/K_2CO_3 9:1 (w/w) (4.2 g, diameter = 1 cm, length = 12 cm), eluting with DCM/pyridine 95:5 (v/v). The yellow-orange band was collected and concentrated in vacuo to remove DCM, to about 1 mL. Hexanes (10 mL) was added dropwise with mixing. The resulting precipitate was triturated, the mixture was centrifuged, and the supernatant was decanted. The precipitate was dried in vacuo, and then dissolved in DCM (1.8 mL). To the resulting solution was added pentane (9.0 mL), dropwise. The mixture was triturated, centrifuged, and the supernatant was decanted. The solid was dried in vacuo (0.2 Torr, 23 ºC, 8 hours) to afford 119 mg of the title compound (as a solvate with 0.58 dichloromethane molecules) as an orange solid (55% yield).

NMR Spectroscopy [mixture of 2 rotamers]: ^{1}H NMR (600 MHz, $CDCl_3$, 23 ºC, δ): 9.09 (br s, 2H), 8.29 (br s, 1H), 7.68 (br s, 1H), 7.58–7.53 (m, 2H), 7.53–7.46 (m, 2H), 7.37 (ddd, *J* = 7.6, 7.6, 1.2 Hz, 1H), 7.28 (dd, *J* = 7.6, 7.6 Hz, 1H), 7.20–7.04 (m, 5H), 6.99 (br s, 1H), 6.98 (d, *J* = 7.6 Hz, 1H), 6.78 (dd, *J* = 8.4, 2.4 Hz, 1H), 6.61 (br s, 1H), 6.15 (br s, minor rotamer, 1H), 5.94 (br s, major rotamer, 1H), 4.09 (br s, minor rotamer, 3H), 3.42 (br s, major rotamer, 3H). ^{13}C NMR (125 MHz, $CDCl_3$, 23 ºC, δ): 162.3 (br), 156.1 (br), 151.4, 147.0 (br), 140.7 (br), 137.1, 136.7, 136.2 (br), 131.5, 130.3, 130.2, 129.8 (br), 128.8 (br), 127.9 (br), 126.4, 124.2, 124.1, 122.7, 122.3 (br), 121.8 (br), 112.3 (br), 109.1 (br), 55.6. Anal: calcd for $C_{29}H_{23}BrN_4NiO_5S(CH_2Cl_2)_{0.58}$: C, 48.84; H, 3.35, N, 7.70; found: C, 48.49; H, 3.25; N, 7.55. HRMS (ESI-TOF) (m/z): calcd for $C_{24}H_{19}{}^{79}BrN_3NiO_5S$ $[M - \text{pyridine} + H]^+$, 597.9583; found,

597.9566.

2-(Benzo[b]thiophen-3-yl)-5,5-dimethyl-1,3,2-dioxaborinane (S1)

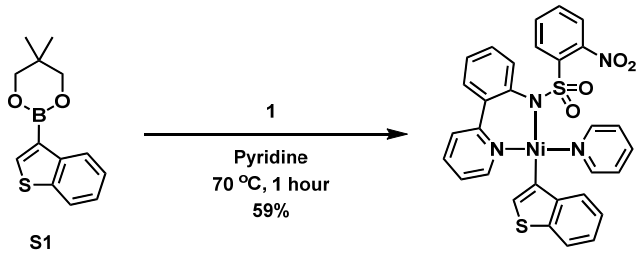


S1

To a 20 mL scintillation vial were added benzothiophene-3-boronic acid (534 mg, 3.00 mmol, 1.00 equiv.), 2,2-dimethyl-1,3-propanediol (328 mg, 3.15 mmol, 1.05 equiv.), a Teflon-coated stirbar, 10 mL of diethyl ether, and anhydrous magnesium sulfate (1.1 g). The vial was sealed with a Teflon-lined screw cap, shaken to mix, and stirred at 23 ºC for 3 hours. Then the mixture was filtered through filter paper, which was rinsed with ether, and the filtrate was concentrated in vacuo to afford a residue that was purified by chromatography on silica gel (diameter = 3 cm, length = 11 cm), eluting with hexanes/ethyl acetate 9:1 (v/v). A solid was obtained, which was triturated with 3 mL of hexanes and dried in vacuo to afford 263 mg of the title compound as a light-pink solid (36% yield).

NMR Spectroscopy: ^{1}H NMR (600 MHz, $CDCl_3$, 23 ºC, δ): 8.43 (d, J = 8.1 Hz, 1H), 8.04 (s, 1H), 7.90 (d, J = 8.0 Hz, 1H), 7.40 (dd, J = 8.2, 8.1 Hz, 1H), 7.34 (dd, J = 8.1, 8.1 Hz, 1H), 3.84 (s, 4H), 1.07 (s, 6H). ^{13}C NMR (100 MHz, $CDCl_3$, 23 ºC, δ): 142.9, 141.1, 137.9, 125.6, 124.2, 124.0, 122.3, 72.4, 32.1, 22.1. Anal: calcd for $C_{13}H_{15}BO_2S$: C, 63.44; H, 6.14; found: C, 63.11; H, 6.05. HRMS-FIA (ESI-TOF) (m/z): calcd for $C_{13}H_{16}BO_2S$ $[M + H]^+$, 247.0964; found, 247.0953.

Synthesis of nickel(II) aryl complex 3c



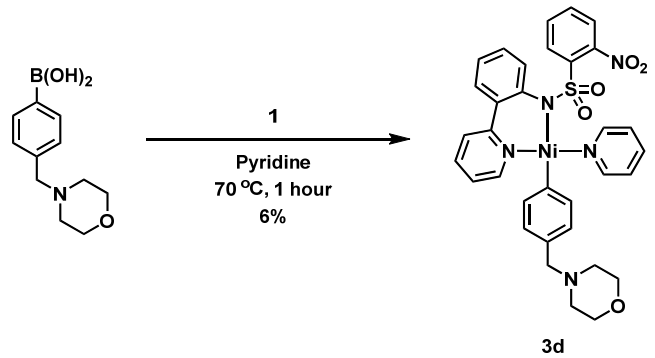
S1 3c

To a 250 mL 2-necked round-bottomed flask were added nickel cube **1** (142 mg, 75.0 μmol, 0.250 equiv.), arylboronic ester **S1** (73.8 mg, 0.300 mmol, 1.00 equiv.), and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a septum, and its atmosphere was evacuated and backfilled with nitrogen three times. Dry pyridine (12 mL) was added by syringe, and the septum was replaced with a glass stopper. The mixture was heated with stirring at 70 ºC

for 1 hour. Once cooled to 23 ºC, hexanes (120 mL) was added with stirring. The mixture was filtered through celite on a glass frit, which was then rinsed with hexanes (20 mL). The hexanes filtrate was discarded, and the collected residue was dissolved in DCM/pyridine 95:5 (v/v), passed through the frit, and concentrated in vacuo to give a residue. The residue was quickly purified by chromatography on silica gel/K_2CO_3 9:1 (w/w) (5.1 g, diameter = 1 cm, length = 14 cm), eluting with DCM/pyridine 95:5 (v/v). The yellow-orange band was collected and concentrated in vacuo to remove DCM, to about 1.5 mL. Hexanes was added dropwise with mixing (2.6 mL), inducing gradual precipitation of yellow solid. A further 5.4 mL of hexanes was added dropwise with mixing. The solid was triturated and centrifuged, the supernatant was decanted, and the residual solid was dried in vacuo. The solid was dissolved in DCM (9.0 mL), and to this solution was added pentane (5 mL) dropwise with mixing, which induced gradual precipitation of a solid. A further 55 mL of pentane was added with mixing. The supernatant was decanted, leaving about 10 mL of mixture, which was triturated with a spatula, sonicated, centrifuged, and the remaining supernatant was decanted. The solid was dried in vacuo (0.2 Torr, 23 ºC, overnight) to afford 111 mg of the title compound as a yellow solid (59% yield).

NMR Spectroscopy: ^{1}H NMR (500 MHz, $CDCl_3$, 23 ºC, δ): 9.13 (d, *J* = 5.1 Hz, 2H), 8.64 (br s, 1H), 8.32 (d, *J* = 5.6 Hz, 1H), 7.66–7.60 (m, 3H), 7.56 (d, *J* = 7.8 Hz, 1H), 7.51–7.42 (m, 2H), 7.22 (ddd, *J* = 7.8, 7.6, 1.5 Hz, 1H), 7.18 (d, *J* = 7.8 Hz, 1H), 7.16–7.04 (m, 7H), 7.01 (ddd, *J* = 7.8, 7.6, 1.2 Hz, 1H), 6.96 (d, *J* = 7.6 Hz, 1H), 6.52 (ddd, *J* = 5.8, 5.8, 1.2 Hz, 1H). ^{13}C NMR (125 MHz, $CDCl_3$, 23 ºC, δ): 155.8, 152.1, 151.5, 147.1, 145.5, 142.5 (br), 140.6, 139.4, 137.3, 136.9, 136.5, 135.9, 132.0, 130.5, 130.3, 130.1, 128.8, 128.3, 126.3, 124.7, 124.1, 123.2, 122.92, 122.85, 122.42, 122.41, 122.0, 117.6. Anal: calcd for $C_{30}H_{22}N_4NiO_4S_2$: C, 57.62; H, 3.55, N, 8.96; found: C, 57.22; H, 3.45; N, 8.83. HRMS-FIA (ESI-TOF) (m/z): calcd for $C_{26}H_{18}N_3NiO_6S_2$ [M – pyridine + HCO_2^-]$^-$, 589.9991; found, 589.9987.

Synthesis of nickel(II) aryl complex 3d



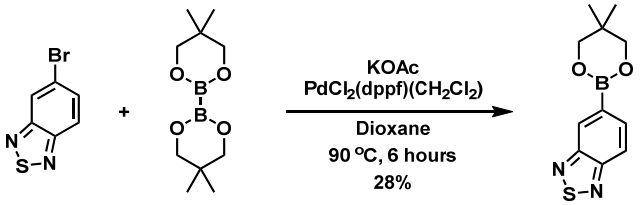
3d

To a 500 mL 2-necked round-bottomed flask were added nickel cube **1** (425 mg, 0.225 mmol, 0.25 equiv.), 4-(morpholinomethyl)phenylboronic acid (199 mg, 0.900 mmol, 1.00 equiv.), and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a septum, and

its atmosphere was evacuated and backfilled with nitrogen three times. Dry pyridine (36 mL) was added by syringe, and the septum was replaced with a glass stopper and Teflon sleeve. The mixture was heated with stirring at 70 ºC for 1 hour. Once cooled to 23 ºC, hexanes (360 mL) was added with stirring. The mixture was filtered through celite on a glass frit, which was then rinsed with hexanes (20 mL). The hexanes filtrate was discarded, and the collected residue was dissolved in DCM/pyridine 95:5 (v/v), passed through the frit, and concentrated in vacuo to a residue that was purified by chromatography on silica gel/K_2CO_3 9:1 (w/w) (4.2 g, diameter = 1 cm, length = 13 cm), eluting with DCM/pyridine 95:5 (v/v). The yellow band was collected and concentrated in vacuo to remove DCM, to about 2 mL. Hexanes (10 mL) was added dropwise with mixing, and a yellow solid precipitated. The precipitate was triturated, the mixture was centrifuged, and the supernatant was decanted. The solid was triturated in hexanes (10 mL), the mixture was centrifuged, and the supernatant was decanted. The solid was dried in vacuo (0.2 Torr, 23 ºC, 9.5 hours) to afford 34.2 mg of the title compound as a yellow solid (6% yield).

NMR Spectroscopy: ^{1}H NMR (600 MHz, $CDCl_3$, 23 ºC, δ): 9.18 (d, *J* = 5.8 Hz, 2H), 8.21 (d, *J* = 5.8 Hz, 1H), 7.57–7.51 (m, 3H), 7.46 (d, *J* = 7.9 Hz, 1H), 7.37–7.32 (m, 3H), 7.32–7.27 (m, 1H), 7.18–7.10 (m, 4H), 7.07 (dd, *J* = 7.9, 1.0 Hz, 1H), 7.02–6.96 (m, 2H), 6.68 (d, *J* = 7.9 Hz, 2H), 6.59 (dd, *J* = 6.0, 5.9 Hz, 1H), 3.64–3.57 (m, 4H), 3.23 (s, 2H), 2.28 (br s, 4H). ^{13}C NMR (125 MHz, $CDCl_3$, 23 ºC, δ): 156.0, 153.6, 152.6, 151.5, 147.1, 141.3, 137.1, 136.7, 136.6, 135.6, 135.2, 131.6, 131.5, 130.4, 130.1, 129.8, 128.8, 128.3, 127.1, 124.3, 124.1, 122.8, 122.6, 121.7, 67.1, 63.5, 53.7. Anal: calcd for $C_{33}H_{31}N_5NiO_5S$: C, 59.30; H, 4.67, N, 10.48; found: C, 59.18; H, 4.48, N, 10.84. HRMS (ESI-TOF) (m/z): calcd for $C_{29}H_{27}N_4NiO_7S$ [M – pyridine + HCO_2]$^-$, 633.0954; found, 633.0948.

5-(5,5-Dimethyl-1,3,2-dioxaborinan-2-yl)benzo[c][1,2,5]thiadiazole (S2)



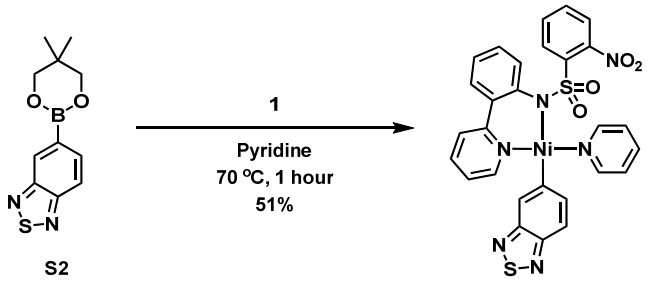
S2

To a 100 mL 2-necked round-bottomed flask were added 5-bromobenzo[c][1,2,5]thiadiazole (430. mg, 2.00 mmol, 1.00 equiv.), 5,5,5',5'-tetramethyl-2,2'-bi(1,3,2-dioxaborinane (497 mg, 2.20 mmol, 1.10 equiv.), palladium dichloride-bis(diphenylphosphino)ferrocene-dichloromethane complex (163 mg, 0.200 mmol, 0.100 equiv.), potassium acetate (393 mg, 4.00 mmol, 2.00 equiv.), and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a septum, and its atmosphere was evacuated and backfilled with nitrogen three times. Dry, degassed dioxane (10 mL) was added by syringe, and the septum was replaced with a glass stopper. The mixture was heated with stirring at 90 ºC for 6 hours. Once cooled to 23 ºC, the

mixture was filtered through celite on a glass frit, and the flask and celite were rinsed with dichloromethane. The combined filtrate was concentrated in vacuo to afford a residue, which was dissolved in dichloromethane and passed through sodium sulfate. The filtrate was concentrated in vacuo to afford a residue, which was purified by chromatography on silica gel (45 g, diameter = 3.5 cm, length = 14 cm), eluting with DCM/methanol 99:1 (v/v), to afford 138 mg of the title compound as a tan solid (28% yield).

NMR Spectroscopy: ^{1}H NMR (500 MHz, $CDCl_3$, 23 ºC, δ): 8.47 (s, 1H), 7.95 (s, 2H), 3.83 (s, 4H), 1.05 (s, 6H). ^{13}C NMR (125 MHz, $CDCl_3$, 23 ºC, δ): 156.3, 155.1, 133.5, 128.4, 120.4, 72.7, 32.1, 22.0. Anal: calcd for $C_{11}H_{13}BN_2O_2S$: C, 53.25; H, 5.28, N, 11.29; found: C, 53.45; H, 5.29; N, 11.25. HRMS (ESI-TOF) (m/z): calcd for $C_{11}H_{14}BN_2O_2S$ $[M + H]^+$, 249.0869; found, 249.0861.

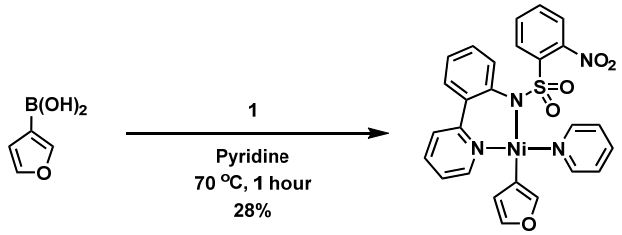
Synthesis of nickel(II) aryl complex 3e



To a 250 mL 2-necked round-bottomed flask were added nickel cube **1** (142 mg, 75.0 μmol, 0.250 equiv.), arylboronic ester **S2** (74.4 mg, 0.300 mmol, 1.00 equiv.), and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a septum, and its atmosphere was evacuated and backfilled with nitrogen three times. Dry pyridine (12 mL) was added by syringe, and the septum was replaced with a glass stopper. The mixture was heated with stirring at 70 ºC for 1 hour. Once cooled to 23 ºC, hexanes (120 mL) was added with stirring. The mixture was filtered through celite on a glass frit, which was then rinsed with hexanes (20 mL). The hexanes filtrate was discarded, and the collected residue was dissolved in DCM/pyridine 95:5 (v/v), passed through the frit, and concentrated in vacuo to give a residue. The residue was quickly purified by chromatography on silica gel/K_2CO_3 9:1 (w/w) (4.7 g, diameter = 1 cm, length = 13 cm), eluting with DCM/pyridine 95:5 (v/v). The yellow-orange band was collected and concentrated in vacuo to remove DCM, to about 0.5 mL. Hexanes was added dropwise with mixing (3 mL), inducing a biphasic mixture to form. The denser phase was ground with a metal spatula until completely converted to a yellow powder. A further 2 mL of hexanes was added. The solid was triturated, centrifuged, and the supernatant was decanted. Hexanes (10 mL) was added, and the mixture was triturated, sonicated for 2 minutes, centrifuged, and the supernatant was decanted. The solid was dried in vacuo (0.2 Torr, 23 ºC, 17 hours) to afford 107 mg of the title

compound (as solvate with 0.8 pyridine and 0.08 hexane molecules) as a yellow solid (51% yield). NMR Spectroscopy: ^{1}H NMR (600 MHz, pyridine-d_5, 23 ºC, δ): 8.54 (s, 1H), 8.46 (d, J = 5.9 Hz, 1H), 8.24 (d, J = 8.8 Hz, 1H), 7.86 (d, J = 7.6 Hz, 1H), 7.84 (dd, J = 7.6, 1.2 Hz, 1H), 7.67 (ddd, J = 7.6, 7.6, 1.2 Hz, 1H), 7.51 (dd, J = 7.6, 7.6 Hz, 1H), 7.47 (d, J = 7.6 Hz, 1H), 7.38 (ddd, J = 7.6, 7.6, 1.2 Hz, 1H), 7.30 (dd, J = 8.2, 1.2 Hz, 1H), 7.25 (d, J = 7.6 Hz, 1H), 7.14 (ddd, J = 7.6, 7.6, 1.2 Hz, 1H), 6.93 (dd, J = 7.6, 7.6 Hz, 1H), 6.65 (ddd, J = 5.9, 5.9, 1.2 Hz, 1H). ^{13}C NMR (125 MHz, pyridine-d_5, 23 ºC, δ): 164.1, 156.3, 154.6, 153.7, 152.8, 148.1, 142.2, 138.8, 137.7, 137.3, 136.6, 136.4, 132.2, 131.4, 131.3, 131.0, 129.5, 129.1, 126.9, 125.4, 123.1, 117.3. Anal: calcd for $C_{28}H_{20}N_6NiO_4S_2(C_5H_5N)_{0.8}(C_6H_{14})_{0.08}$: C, 55.93; H, 3.63, N, 13.66; found: C, 55.98; H, 3.64; N, 13.61. HRMS (ESI-TOF) (m/z): calcd for $C_{28}H_{21}N_6NiO_4S_2$ $[M + H]^+$, 627.0419; found, 627.0410.

Synthesis of nickel(II) aryl complex 3f

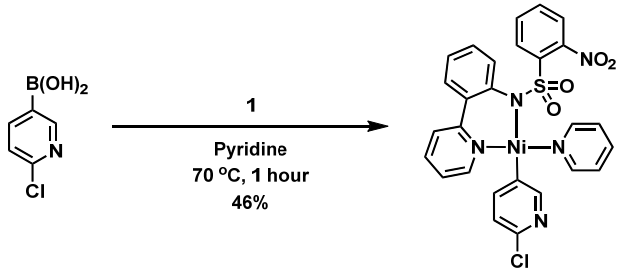


3f

To a 250 mL 2-necked round-bottomed flask were added nickel cube **1** (142 mg, 75.0 μmol, 0.250 equiv.), furan-3-boronic acid (33.6 mg, 0.300 mmol, 1.00 equiv.), and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a septum, and its atmosphere was evacuated and backfilled with nitrogen three times. Dry pyridine (12 mL) was added by syringe, and the septum was replaced with a glass stopper. The mixture was heated with stirring at 70 ºC for 1 hour. Once cooled to 23 ºC, hexanes (120 mL) was added with stirring. The mixture was filtered through celite on a glass frit, which was then rinsed with hexanes (20 mL). The hexanes filtrate was discarded, and the collected residue was dissolved in DCM, passed through the frit, and concentrated by rotary evaporation to a residue that was treated with hexanes (15 mL), sonicated. The mixture was then concentrated in vacuo. The resulting residue was quickly purified by chromatography on silica gel/K_2CO_3 9:1 (w/w) (5.4 g, diameter = 1 cm, length = 13 cm), eluting with DCM/pyridine 95:5 (v/v). The yellow-orange band was collected and concentrated in vacuo to remove DCM, to about 0.5 mL. Hexanes (7 mL) was added dropwise with mixing, which caused a precipitate to form, which was triturated. The supernatant was decanted, the precipitate was dried in vacuo, and then dissolved in DCM (1 mL). To the resulting solution was added hexanes (10 mL) slowly, with mixing, to give a precipitate that was triturated. The mixture was centrifuged, the supernatant was decanted, and the solid was dried in vacuo (0.2 Torr, 23 ºC, 15 hours) to afford 49.0 mg of the title compound (as a solvate with 0.32 dichloromethane molecules) as an orange solid (28% yield).

NMR Spectroscopy: ^{1}H NMR (600 MHz, $CDCl_3$, 23 ºC, δ): 9.14 (d, *J* = 5.3 Hz, 2H), 8.45 (dd, *J* = 5.9, 1.2 Hz, 1H), 7.62 (dd, *J* = 7.6, 7.6 Hz, 1H), 7.57–7.50 (m, 3H), 7.39–7.33 (m, 2H), 7.25–7.20 (m, 2H), 7.16–7.12 (m, 2H), 7.11–7.09 (m, 1H), 7.06 (d, *J* = 7.6 Hz, 1H), 7.03–6.97 (m, 2H), 6.72 (ddd, *J* = 5.9, 5.8, 1.2 Hz, 1H), 6.23 (s, 1H), 5.58 (d, *J* = 1.2 Hz, 1H). ^{13}C NMR (100 MHz, $CDCl_3$, 23 ºC, δ): 155.8, 154.1, 152.1, 147.0, 141.3, 140.8, 139.8, 137.5, 137.0, 136.2, 135.8, 131.8, 130.4, 130.3, 130.0, 128.8, 128.2, 124.5, 124.1, 122.8, 122.1, 121.6, 116.1, 113.3. Anal: calcd for $C_{26}H_{20}N_4NiO_5S(CH_2Cl_2)_{0.32}$: C, 53.91; H, 3.55, N, 9.55; found: C, 54.30; H, 3.15; N, 9.54. HRMS (ESI-TOF) (m/z): calcd for $C_{22}H_{16}N_3NiO_7S$ [M – pyridine + HCO_2^-]$^-$, 524.0063; found, 524.0065.

Synthesis of nickel(II) aryl complex 3g



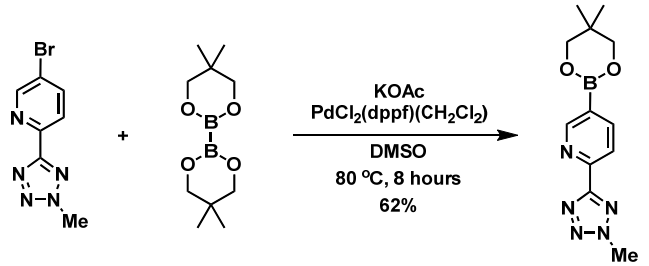
3g

To a 250 mL 2-necked round-bottomed flask were added nickel cube **1** (142 mg, 75.0 μmol, 0.250 equiv.), 6-chloropyridine-3-boronic acid (47.2 mg, 0.300 mmol, 1.00 equiv.), and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a septum, and its atmosphere was evacuated and backfilled with nitrogen three times. Dry pyridine (12 mL) was added by syringe, and the septum was replaced with a glass stopper. The mixture was heated with stirring at 70 ºC for 1 hour. Once cooled to 23 ºC, hexanes (120 mL) was added with stirring. The mixture was filtered through celite on a glass frit, which was then rinsed with hexanes (20 mL). The hexanes filtrate was discarded, and the collected residue was dissolved in DCM/pyridine 95:5 (v/v), passed through the frit, and concentrated in vacuo to give a residue. The residue was quickly purified by chromatography on silica gel/K_2CO_3 9:1 (w/w) (4.75 g, diameter = 1 cm, length = 13 cm), eluting with DCM/pyridine 95:5 (v/v). The yellow-orange band was collected and concentrated in vacuo to remove DCM, to about 1 mL. Hexanes was added dropwise with mixing until turbid (0.3 mL), inducing precipitation of a yellow solid. A further 4.7 mL of hexanes was added dropwise with mixing. The solid was triturated, centrifuged, and the supernatant was decanted. Hexanes (5 mL) was added, and the mixture was triturated, centrifuged, and the supernatant was decanted. The solid was dried in vacuo (0.2 Torr, 23 ºC, 14 hours) to afford 82.9 mg of the title compound as a yellow solid (46% yield).

NMR Spectroscopy: ^{1}H NMR (600 MHz, pyridine-d_5, 23 ºC, δ): 8.83 (d, *J* = 1.8 Hz, 1H), 8.27 (d, *J* = 5.3 Hz, 1H), 7.89 (dd, *J* = 8.2, 1.7 Hz, 1H), 7.81 (t, *J* = 9.1 Hz, 2H), 7.63 (t, *J* = 7.6 Hz, 1H), 7.49–7.44 (m, 2H), 7.41 (t, *J* = 7.6 Hz, 1H), 7.28–7.23 (m, 2H), 7.14 (t, *J* = 7.7 Hz, 1H), 6.97 (d,

J = 8.2 Hz, 1H), 6.92 (t, *J* = 7.7 Hz, 1H), 6.69 (t, *J* = 6.5 Hz, 1H). ^{13}C NMR (125 MHz, pyridine-d_5, 23 ºC, δ): 156.2, 155.2, 153.1, 149.2, 148.5, 148.0, 146.9, 141.8, 138.9, 137.1, 136.6, 132.3, 131.5, 131.4, 131.0, 129.4, 129.1, 125.4, 123.2, 123.0. Anal: calcd for $C_{27}H_{20}ClN_5NiO_4S$: C, 53.63; H, 3.33, N, 11.58; found: C, 53.75; H, 3.00; N, 11.49. HRMS-FIA(ESI-TOF) (m/z): calcd for $C_{22}H_{16}ClN_4NiO_4S$ [M – pyridine + H]$^+$, 524.9935; found, 524.9916.

5-(5,5-Dimethyl-1,3,2-dioxaborinan-2-yl)-2-(2-methyl-2H-tetrazol-5-yl)pyridine (S3)

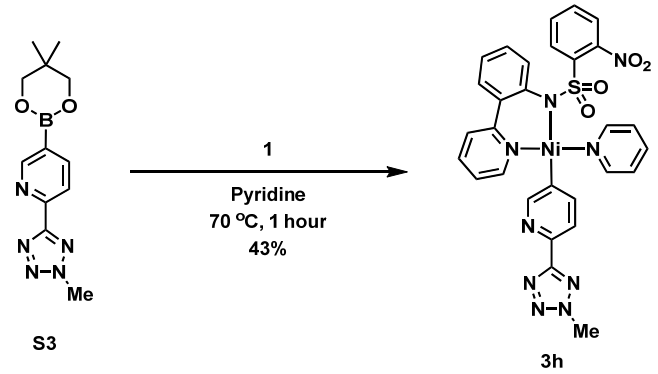


S3

To a 2-necked round-bottomed flask were 5-bromo-2-(2-methyl-2H-tetrazol-5-yl)pyridine (717 mg, 3.00 mmol, 1.00 equiv.), 5,5,5',5'-tetramethyl-2,2'-bi(1,3,2-dioxaborinane) (1.02 g, 4.50 mmol, 1.50 equiv.), palladium dichloride-bis(diphenylphosphino)ferrocene-dichloromethane complex (243 mg, 0.298 mmol, 0.099 equiv.), potassium acetate (883 mg, 9.00 mmol, 3.00 equiv.), and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a septum, and its atmosphere was evacuated and backfilled with nitrogen three times. Dry, degassed DMSO (15 mL) was added by syringe, and the septum was replaced with a glass stopper. The mixture was heated with stirring at 80 ºC for 8 hours. Once cooled to 23 ºC, 15 mL of EtOAc and 15 mL of water were added, and the mixture was transferred to a separatory funnel and the phases were separated. The aqueous layer was extracted with EtOAc (2 × 10 mL), the combined organic layers were dried (Na_2SO_4), filtered, and concentrated in vacuo. The residue thus afforded was purified by chromatography on sodium sulfate (mesh size >50) (60 g, diameter = 4.5 cm, length = 13 cm), eluting with DCM/methanol 96:4 (v/v) to afford a solid that was triturated and sonicated in hexanes (4 mL), and filtered. The solid was triturated twice more with hexanes (2 × 4 mL), then washed with pentane (2 mL), and dried in vacuo to afford 508 mg of the title compound as a tan solid (62% yield).

NMR Spectroscopy: ^{1}H NMR (500 MHz, $CDCl_3$, 23 ºC, δ): 9.10 (s, 1H), 8.20 (dd, *J* = 7.7, 1.3 Hz, 1H), 8.17 (d, *J* = 7.7 Hz, 1H), 4.43 (s, 3H), 3.78 (s, 4H), 1.03 (s, 6H). ^{13}C NMR (125 MHz, $CDCl_3$, 23 ºC, δ): 165.3, 155.5, 148.1, 142.7, 128.6 (br), 121.5, 72.5, 39.8, 32.1, 22.0. HRMS-FIA(ESI-TOF) (m/z): calcd for $C_{12}H_{17}BN_5O_2$ [M + H]$^+$, 274.1476; found, 274.1472.

Synthesis of nickel(II) aryl complex 3h

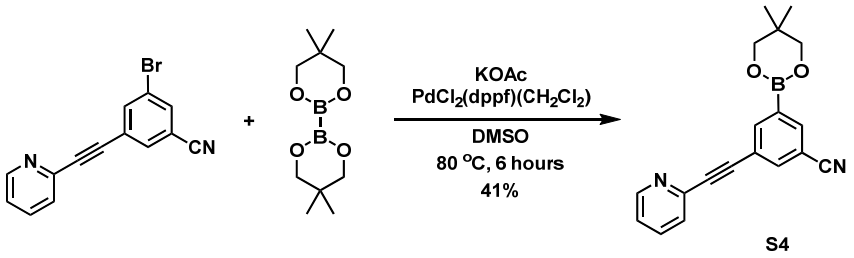


To a 250 mL 2-necked round-bottomed flask were added nickel cube **1** (182 mg, 96.2 μmol, 0.250 equiv. (1.00 equiv. of Ni)), arylboronic ester **S3** (109 mg, 0.400 mmol, 1.04 equiv.), and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a septum, and its atmosphere was evacuated and backfilled with nitrogen three times. Dry pyridine (16 mL) was added by syringe, and the septum was replaced with a glass stopper. The mixture was heated with stirring at 70 ºC for 1 hour. Once cooled to 23 ºC, hexanes (160 mL) was added with stirring. The mixture was filtered through celite on a glass frit, which was then rinsed with hexanes (20 mL). The hexanes filtrate was discarded, and the collected residue was dissolved in DCM/pyridine 95:5 (v/v), passed through the frit, and concentrated in vacuo to give a residue. The residue was quickly purified by chromatography on silica gel/K_2CO_3 9:1 (w/w) (4.5 g), which was layered on top of basic alumina (2.1 g) within the same column (diameter = 1 cm), eluting with DCM/pyridine 95:5 (v/v). The yellow-orange band was collected and concentrated in vacuo to remove DCM, to about 3 mL. Hexanes was added dropwise with mixing (17 mL), the mixture was thoroughly ground with a metal spatula, and the supernatant was decanted. This process was repeated with 13 mL of hexanes. A further 10 mL of hexanes was added, and the solid was triturated, sonicated, centrifuged, and the supernatant was decanted. The solid was dried in vacuo (0.2 Torr, 23 ºC, 16 hours) to afford 110. mg of the title compound (as a solvate with 0.18 hexane molecules) as a yellow solid (43% yield). The product, a fine powder, was not heated under vacuum to further remove hexanes because of the possibility for decomposition at high temperature.

NMR Spectroscopy: ^{1}H NMR (500 MHz, pyridine-d_5, 23 ºC, δ): 9.35 (s, 1H), 8.36 (d, J = 5.9 Hz, 1H), 8.12 (d, J = 7.8 Hz, 1H), 7.89 (d, J = 7.8 Hz, 1H), 7.84 (d, J = 7.8 Hz, 1H), 7.81 (d, J = 7.8 Hz, 1H), 7.64 (dd, J = 7.9, 7.5 Hz, 1H), 7.50–7.43 (m, 2H), 7.40 (dd, J = 7.8, 7.7 Hz, 1H), 7.29 (d, J = 7.9 Hz, 1H), 7.25 (d, J = 8.2 Hz, 1H), 7.14 (dd, J = 7.8, 7.6 Hz, 1H), 6.93 (dd, J = 7.8, 7.6 Hz, 1H), 6.70–6.64 (m, 1H), 4.21 (s, 3H). ^{13}C NMR (125 MHz, pyridine-d_5, 23 ºC, δ): 166.7, 156.4, 156.3, 155.4, 153.1, 148.1, 145.0, 143.3, 141.9, 138.8, 137.2, 136.6, 132.3, 131.44, 131.35, 131.0, 129.4, 129.1, 125.4, 123.2, 121.3, 39.7. Anal: calcd for $C_{29}H_{23}N_9NiO_4S(C_6H_{14})_{0.18}$: C, 54.10; H,

3.85, N, 18.88; found: C, 54.26; H, 3.26; N, 18.34. HRMS-FIA(ESI-TOF) (m/z): calcd for $C_{24}H_{19}N_8NiO_4S$ [M – pyridine + H]$^+$, 573.0604; found, 573.0587.

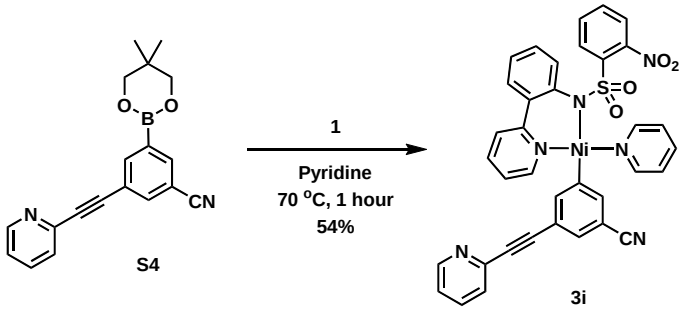
3-(5,5-Dimethyl-1,3,2-dioxaborinan-2-yl)-5-(pyridin-2-ylethynyl)benzonitrile (S4)



To a 50 mL 2-necked round-bottomed flask were added 3-bromo-5-(pyridin-2-ylethynyl)benzonitrile (400. mg, 1.41 mmol, 1.00 equiv.), 5,5,5',5'-tetramethyl-2,2'-bi(1,3,2-dioxaborinane) (479 mg, 2.12 mmol, 1.50 equiv.), palladium dichloride-bis(diphenylphosphino)ferrocene-dichloromethane complex (115 mg, 0.141 mmol, 0.100 equiv.), potassium acetate (416 mg, 4.24 mmol, 3.00 equiv.), and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a glass stopper, and its atmosphere was evacuated and backfilled with nitrogen three times. Dry, degassed DMSO (7.1 mL) was added. The mixture was heated with stirring at 80 ºC for 6 hours. Once cooled to 23 ºC, 7 mL of EtOAc and 7 mL of water were added, and the mixture was transferred to a separatory funnel and the phases were separated. The aqueous layer was extracted with 7 mL EtOAc, the combined organic layers were washed with 7 mL water, dried over anhydrous sodium sulfate, filtered, and concentrated in vacuo. The residue thus afforded was purified by chromatography on silica gel (diameter = 3 cm, length = 11 cm), eluting with DCM/methanol 97:3 (v/v). The residue thus obtained was dissolved in DCM (1 mL), and hexanes (14 mL) was added. The mixture was concentrated to about 5 mL, hexanes (5 mL) was added with mixing, and the supernatant was decanted. The remaining solid was dissolved in ether, and this solution was passed through sodium sulfate and concentrated in vacuo to afford 183 mg of the title compound as a beige solid (41% yield). The product was contaminated with minor impurities as evidenced by ^{1}H NMR. However, purity is sufficient for use in the synthesis of **3i**.

NMR Spectroscopy: ^{1}H NMR (600 MHz, $CDCl_3$, 23 ºC, δ): 8.62 (d, J = 4.7 Hz, 1H), 8.21 (s, 1H), 8.03 (s, 1H), 7.88 (s, 1H), 7.69 (ddd, J = 7.6, 7.6, 1.8 Hz, 1H), 7.51 (d, J = 7.6 Hz, 1H), 7.26 (dd, J = 7.6, 4.7 Hz, 1H), 3.77 (s, 4H), 1.01 (s, 6H). ^{13}C NMR (100 MHz, $CDCl_3$, 23 ºC, δ): 150.3, 142.9, 141.6, 137.5, 136.7, 136.4, 134.5 (br), 127.4, 123.3, 123.1, 118.4, 112.4, 90.3, 87.0, 72.5, 32.0, 21.9. HRMS-FIA(ESI-TOF) (m/z): calcd for $C_{19}H_{18}BN_2O_2$ [M + H]$^+$, 317.1462; found, 317.1469.

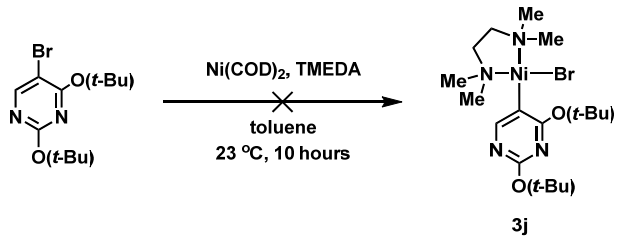
Synthesis of nickel(II) aryl complex 3i



To a 250 mL 2-necked round-bottomed flask were added nickel cube **1** (118 mg, 62.5 μmol, 0.250 equiv.), arylboronic ester **S4** (79.0 mg, 0.250 mmol, 1.00 equiv.), and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a septum, and its atmosphere was evacuated and backfilled with nitrogen three times. Dry pyridine (10 mL) was added by syringe, and the septum was replaced with a glass stopper. The mixture was heated with stirring at 70 ºC for 1 hour. Once cooled to 23 ºC, hexanes (100 mL) was added with stirring. The mixture was filtered through celite on a glass frit, which was then rinsed with hexanes (20 mL). The hexanes filtrate was discarded, and the collected residue was dissolved in DCM/pyridine 95:5 (v/v), passed through the frit, and concentrated in vacuo to give a residue. The residue was quickly purified by chromatography on silica gel/K_2CO_3 9:1 (w/w) (4.9 g, diameter = 1 cm, length = 13 cm), eluting with DCM/pyridine 95:5 (v/v). The yellow-orange band was collected and concentrated in vacuo to remove DCM, to about 0.5 mL. Hexanes (5 mL) was added with mixing, and a biphasic mixture formed, which was thoroughly ground with a metal spatula until the denser phase was converted to a yellow solid. The mixture was sonicated for 5 minutes, centrifuged, and the supernatant was decanted. The solid was triturated in hexanes (10 mL), the mixture was sonicated for 3 minutes, centrifuged, and the supernatant was decanted. The solid was dried in vacuo (0.2 Torr, 23 ºC, 15 hours) to afford 95.4 mg of the title compound (as a solvate with 0.18 hexane molecules) as a yellow solid (54% yield). The product, a fine powder, was not heated under vacuum to further remove hexanes because of the possibility for decomposition at high temperature.

NMR Spectroscopy: ^{1}H NMR (500 MHz, pyridine-d_5, 23 ºC, δ): ^{13}C NMR (125 MHz, pyridine-d_5, 23 ºC, δ): 160.4, 156.2, 152.7, 151.1, 148.1, 144.0, 143.2, 141.9, 139.4, 138.9, 137.1, 136.9, 136.5, 132.3, 131.5, 131.4, 131.0, 130.9, 129.5, 129.1, 128.1, 125.5, 124.3, 123.3, 120.6, 119.8, 110.2, 90.9, 88.7. Anal: calcd for $C_{36}H_{24}N_6NiO_4S(C_6H_{14})_{0.18}$: C, 62.65; H, 3.76, N, 11.82; found: C, 62.26; H, 3.47; N, 11.59. HRMS (ESI-TOF) (m/z): calcd for $C_{31}H_{20}N_5NiO_4S$ $[M - \text{pyridine} + H]^+$, 616.0590; found, 616.0581.

Attempted synthesis of nickel(II) complex 3j by oxidative addition

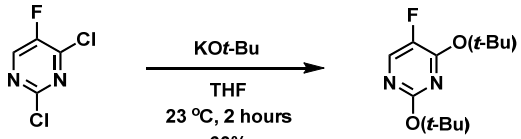


3j

This procedure was attempted with 2,4-di-tert-butoxy-5-bromopyrimidine (a known compound[4]), following a procedure for the synthesis of other nickel(II) σ-aryl complexes by oxidative addition.[1] In a N_2-filled glovebox, 2,4-di-tert-butoxy-5-bromopyrimidine (50.0 mg, 0.165 mmol, 1.00 equiv.) and nickel(0) bis-cyclooctadiene (45.4 mg, 0.165 mmol, 1.00 equiv.) were treated with a solution of tetramethylethylenediamine (25.0 μL, 0.167 mmol, 1.01 equiv.) in toluene (10 mL). The solution was stirred at 23 ºC for 10 hours. After addition of excess pentane, an oily precipitate was observed. The mixture was subjected to centrifugation and the supernatant was decanted. The residue was washed with ether to afford a brown gooey residue. The desired product **3j** was not present based on analysis of the residue by ^{1}H NMR spectroscopy.

Preparation of Authentic Aryl Fluorides

2,4-Di-tert-butoxy-5-fluoropyrimidine (4a)



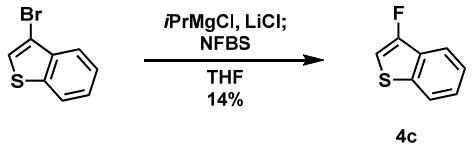
4a

To a 50 mL round-bottomed flask were added 2,4-dichloro-5-fluoropyrimidine (334 mg, 2.00 mmol, 1.00 equiv.) and a Teflon-coated stirbar. The flask was fitted with a septum and connection to a vacuum manifold via a needle, and its atmosphere was evacuated and backfilled with nitrogen three times. Dry THF (10 mL) was added, resulting in a colorless solution. A solution of potassium *tert*-butoxide (561 mg, 5.00 mmol, 2.50 equiv.) in 10 mL of THF was added dropwise over 8 minutes. The reaction mixture was stirred at 23 ºC for 2 hours, and then 10 mL of EtOAc and 10 mL of water were added. The phases were separated, the aqueous layer was extracted with 10 mL of EtOAc, and the organic layers were combined and dried over $MgSO_4$, filtered, and concentrated in vacuo. The residue was purified by chromatography on silica gel (diameter = 2.5 cm, length = 24 cm), eluting with hexanes/EtOAc 96:4 (v/v) to afford 289 mg of the title compound as a colorless oil (60% yield).

NMR Spectroscopy: ^{1}H NMR (400 MHz, $CDCl_3$, 23 ºC, δ): 7.98 (d, J = 2.6 Hz, 1H), 1.64 (s, 9H), 1.57 (s, 9H). ^{13}C NMR (125 MHz, $CDCl_3$, 23 ºC, δ): 159.1 (d, J = 3.7 Hz), 158.9 (d, J = 10.3 Hz),

143.6 (d, J = 252.5 Hz), 143.0 (d, J = 21.2 Hz), 83.3, 80.5, 28.51, 28.45. ^{19}F NMR (376 MHz, $CDCl_3$, 23 ºC, δ): –163.6. HRMS (ESI-TOF) (m/z): calcd for $C_{12}H_{20}FN_2O_2$ $[M + H]^+$, 243.1509; found, 243.1497.

3-Fluorobenzo[*b*]thiophene (4c)[5]



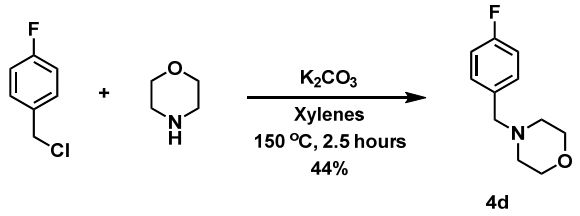
4c

To a 100 mL 2-necked round-bottomed flask were added lithium chloride (890. mg, 21.0 mmol, 2.33 equiv.) and a Teflon-coated stirbar. The flask was fitted in one neck with a Teflon sleeve and glass joint to tube adapter (which was connected to a vacuum manifold), and in the other neck with a septum, and flame-dried under high vacuum to dry the LiCl. After cooling to room temperature and backfilling the atmosphere with N_2, isopropyl magnesium chloride (6.0 mL, 2.0M solution in THF, 12 mmol, 1.3 equiv.) was added, and the mixture was stirred at 23 ºC for 1 hour, and then cooled to -16 ºC (bath temperature) in a NaCl/ice bath. To the stirring mixture was added 3-fluorobenzo[*b*]thiophene (1.18 mL, 9.02 mmol, 1.00 equiv.) over 3 minutes. The mixture was then stirred in the -16 ºC bath for 10 minutes, then warmed to 0 ºC (ice/water bath) and stirred for 75 minutes, then the ice bath was removed, and the mixture was stirred for 45 minutes, and then cooled to 0 ºC. To the stirring mixture was added *N*-fluorobenzenesulfonimide (3.88 g, 12.3 mmol, 1.36 equiv., as a solution in 10 mL THF) over 7 minutes. After an additional 3 minutes at 0 ºC, the ice bath was removed, and the mixture was stirred for an additional 30 minutes. Water (20 mL) was then added, followed by 10 mL pentane, and after shaking, the phases were separated. The aqueous layer was extracted with pentane (3 × 15 mL), and the combined organic layers were dried ($MgSO_4$), filtered, and concentrated in vacuo. The residue was purified by chromatography on silica gel (diameter = 5.5 cm, length = 22 cm), eluting with pentane, to afford 336 mg of the title compound as a mixture with unreacted starting material (5.3:1 molar ratio). To a 100 mL round-bottomed flask was added 336 mg of this mixture, together with DMF (11.1 mL), palladium on activated carbon (235 mg, 10 wt% Pd, 23.5 mg Pd), and a Teflon stirbar. The flask was fitted with a septum and hydrogen balloon, and the headspace was purged with hydrogen. The mixture was stirred at 23 ºC for 65 minutes, and then water (20 mL) was added. The mixture was extracted with pentane (4 × 10 mL), and the combined organic layers were dried ($MgSO_4$), filtered, and concentrated in vacuo. The residue was purified by chromatography on silica gel (diameter = 5.5 cm, length = 18 cm), eluting with pentane, to afford 196 mg of the title compound as a colorless oil (14% yield).

NMR Spectroscopy: ^{1}H NMR (500 MHz, $CDCl_3$, 23 ºC, δ): 7.83–7.76 (m, 2H), 7.46–7.38 (m, 2H), 6.87 (d, J = 2.2 Hz, 1H). ^{13}C NMR (125 MHz, $CDCl_3$, 23 ºC, δ): 152.1 (d, J = 262.5 Hz), 137.0 (d, J = 8.1 Hz), 129.2 (d, J = 24.6 Hz), 125.6, 124.6, 123.3, 120.2 (d, J = 2.5 Hz), 103.5 (d, J = 20.0 Hz). ^{19}F NMR (376 MHz, $CDCl_3$, 23 ºC, δ): –135.7. These spectroscopic data correspond

to previously reported data.[4]

4-(4-Fluorobenzyl)morpholine (4d)[6]



To a 25 mL round-bottomed flask were added potassium carbonate (415 mg, 3.00 mmol, 1.00 equiv.), a Teflon-coated stirbar, xylenes (4.5 mL), morpholine (0.263 mL, 3.00 mmol, 1.00 equiv.), and 4-fluorobenzyl chloride (0.360 mL, 3.01 mmol, 1.00 equiv.). The flask was fitted with reflux condenser, which was fitted with a glass joint to tube adapter (that connected to a vacuum manifold). The flask headspace was purged with nitrogen, and the reaction mixture was heated with magnetic stirring at 150 ºC for 2.5 hours. After cooling to 23 ºC, the reaction mixture was purified by chromatography on silica gel (diameter = 2.5 cm, length = 16 cm), eluting with DCM/methanol/triethylamine 97:3:0.2 (v/v/v) to afford 259 mg of the title compound as a yellow oil (44% yield).

NMR Spectroscopy: ^{1}H NMR (500 MHz, $CDCl_3$, 23 ºC, δ): 7.31–7.26 (m, 2H), 7.03–6.97 (m, 2H), 3.73–3.68 (m, 4H), 3.46 (s, 2H), 2.42 (br s, 4H). ^{13}C NMR (125 MHz, $CDCl_3$, 23 ºC, δ): 162.1 (d, J = 245.0 Hz), 133.6 (d, J = 3.1 Hz), 130.7 (d, J = 8.1 Hz), 115.1 (d, J = 21.2 Hz), 67.1, 62.7, 53.7. ^{19}F NMR (376 MHz, $CDCl_3$, 23 ºC, δ): –115.8. (HRMS (ESI-TOF) (m/z): calcd for $C_{11}H_{15}FNO$ $[M + H]^+$, 196.1138; found, 196.1132. The spectroscopic data correspond to previously reported data.[5]

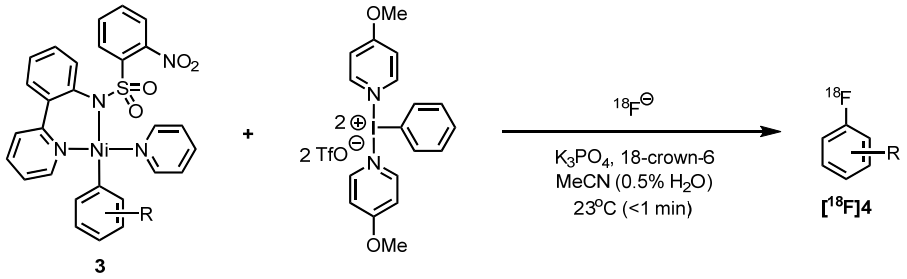
Radiochemistry

General methods

No-carrier-added [^{18}F]fluoride was produced from water 97% enriched in ^{18}O (ISOFLEX, USA) by the nuclear reaction ^{18}O(p,n)^{18}F using a Siemens Eclipse HP cyclotron and a silver-bodied target at Massachusetts General Hospital Athinoula A. Martinos Center for Biomedical Imaging. The produced [^{18}F]fluoride in water was transferred from the cyclotron target by helium push. Radioactivity was measured in a Capintec, Inc. CRC-25PET ion chamber.

Solvents and reagents for radiochemical experiments: Acetonitrile (>99.9%, extra dry, <0.005% water) was purchased from Acros in bottles with a needle-penetrable barrier (AcroSeal®). Water was obtained from a Millipore Milli-Q Integral Water Purification System. 18-crown-6 (99%) was purchased from Alfa Aesar. Potassium phosphate tribasic (Reagent grade, ≥98%) was purchased from Sigma Aldrich.

Radiosynthesis of ^{18}F-labeled molecules



In a nitrogen-filled glovebox, to an oven-dried 1-dram (4 mL) glass vial was added a nickel(II) aryl complex **3** and oxidant (1,1'-(phenyl-λ^3-iodanediyl)bis(4-methoxypyridinium)bis(trifluorome-thanesulfonate))[1] in a 1:1 mass ratio, and the two solids were mixed gently with a metal spatula to give a light yellow or light orange (depending on the color of the starting nickel complex) homogeneous admixture. To an oven-dried 1-dram glass vial was added 2.0 mg of this admixture, and the vial was sealed with a screw cap with a Teflon-lined septum insert under nitrogen, and removed from the glovebox.

An [^{18}F]fluoride solution with 18-crown-6 and potassium phosphate tribasic was prepared as follows. To an oven-dried 1-dram (4 mL) glass vial was added dry 18-crown-6 (20.0 – 44.0 mg) under nitrogen, and this vial was sealed with a Teflon-lined cap. The vial was opened under air, dry MeCN (1.0 mL per 10.0 mg of 18-crown-6) was added quickly, and the vial was sealed and mixed until all 18-crown-6 had dissolved. The vial was opened, aqueous potassium phosphate (0.561M K_3PO_4 in water, 2.0 μL per 10.0 mg of 18-crown-6) was added quickly, and the vial was sealed, shaken, and then vortexed for 10 seconds. The vial was opened, aqueous [^{18}F]fluoride from the cyclotron (3.0 μL per 10.0 mg of 18-crown-6) was added quickly, and the vial was sealed, shaken, and then vortexed for 10 seconds.

The resulting solution (0.50 mL) was quickly added, with a 1-mL plastic syringe with 18-G disposable metal needle, to the vial containing nickel(II) aryl complex and oxidant through the septum. After 1 minute at 23 ºC, the radiochemical yield was then measured (see Measurement of Radiochemical yield, below), and HPLC analysis was also performed (see Characterization of ^{18}F Labeled Molecules, below).

Measurement of radiochemical conversion by radio TLC

Radiochemical conversion (RCC) was determined by multiplying the percentage of radioactivity in the solution and the relative peak integrations of a radio TLC scan (radioTLC yield):

RCC = (radioTLC yield) * (radioactivity in solution)

The radioactivity of the reaction vial was measured. An aliquot of the solution was then taken with a capillary and spotted on a TLC plate. The remaining reaction solution was transferred to another vial, the reaction vial was rinsed with MeCN (0.5 – 1 mL) in order to remove residual reaction solution, and the radioactivity of the empty reaction vial was again measured to

determine the radioactivity of ^{18}F that was left on the walls of the reaction vial, and therefore was not in solution. The fraction of ^{18}F not in solution was determined by dividing the radioactivity of the empty reaction vial by the initial radioactivity of the reaction vial + reaction solution (the second radioactivity measurement was decay corrected to the timepoint of the first, because there was a small delay time between measurements). This number was converted to % of ^{18}F in solution by subtracting the number from 1, and multiplying the result by 100% to convert to percentage units.

The TLC plate was eluted with an appropriate solvent mixture, and then the TLC plate was scanned with a Bioscan AR-2000 RadioTLC Imaging Scanner. The Radiochemical TLC (RTLC) yield was calculated by dividing the area of the product peak by the total area of all peaks, and multiplying by 100% to convert to percentage units.

The radiochemical yield (RCC) was determined by multiplying the RTLC yield by the fraction of radioactivity in solution (typically 0.75–0.95).

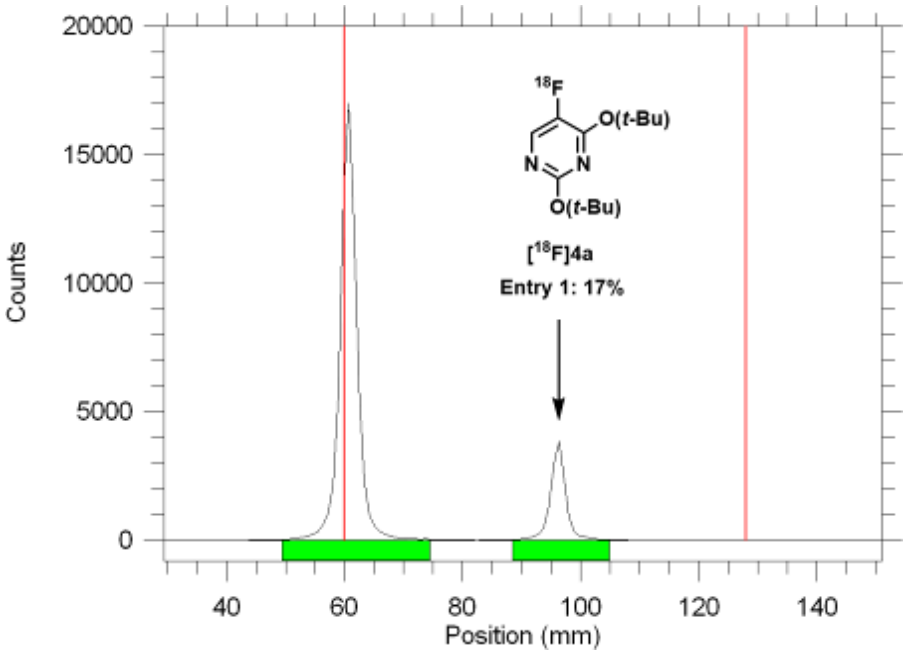
Table S1. Radiochemical conversion data

Entry	Molecule	RTLC yield (%)	^{18}F in solution (%)	RCC (%)	Average RCC (%)
1	**[^{18}F]4a**	17	75	13	15
2		20	77	15	
3		22	78	17	
4		19	79	15	
5		18	78	14	
6		20	78	16	
7	**[^{18}F]4b**	41	94	39	39
8		43	91	39	
9		40	89	36	
10		43	89	38	
11		45	91	41	
12		48	91	44	
13	**[^{18}F]4c**	22	86	19	17
14		18	90	16	
15		18	92	17	
16		21	85	18	
17		17	92	16	
18		19	91	17	
19	**[^{18}F]4d**	47	93	44	41
20		43	89	38	
21		43	91	39	
22		47	91	43	
23		46	91	42	
24	**[^{18}F]4e**	22	91	20	22
25		26	92	24	
26		22	90	20	
27		29	90	26	
28		24	90	22	
29		22	91	20	

Example radioTLC scans

(Note: The startpoint and endpoint of the TLC elution path is approximately indicated by the vertical red lines).

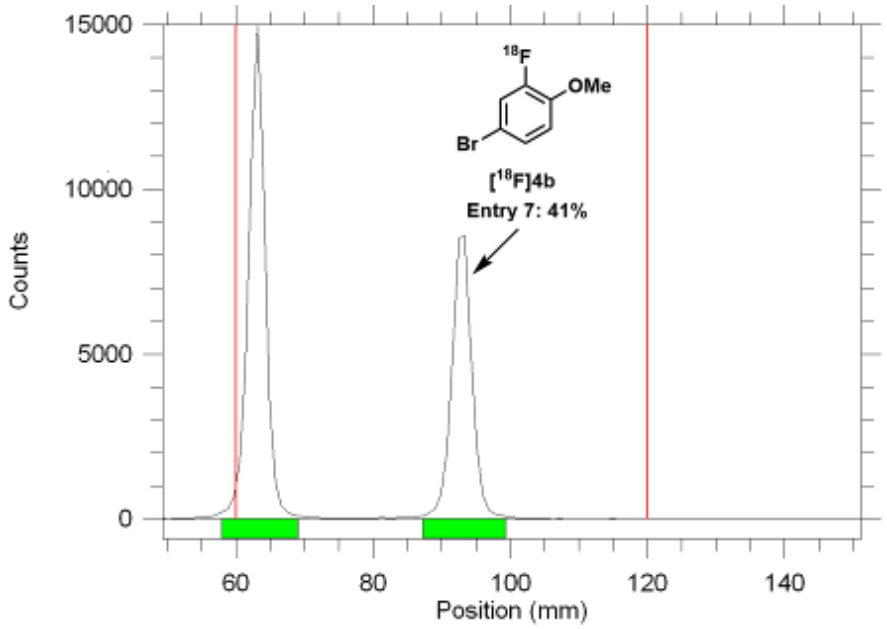
Figure S1. Example radioTLC scan of [^{18}F]4a



Entry 1 of Table S1. TLC eluent: 9:1 hexanes/EtOAc (v/v).

Percent of total integration is listed for **[^{18}F]4a**.

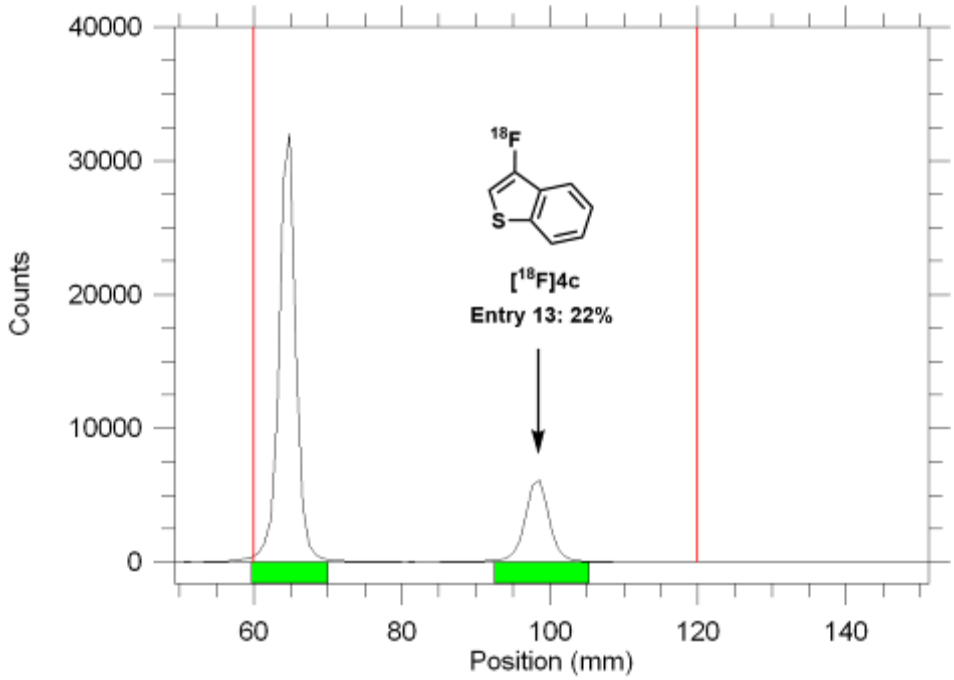
Figure S2. Example radioTLC scan of [^{18}F]4b



Entry 7 of Table S1. TLC eluent: 9:1 hexanes/EtOAc (v/v).

Percent of total integration is listed for **[^{18}F]4b**.

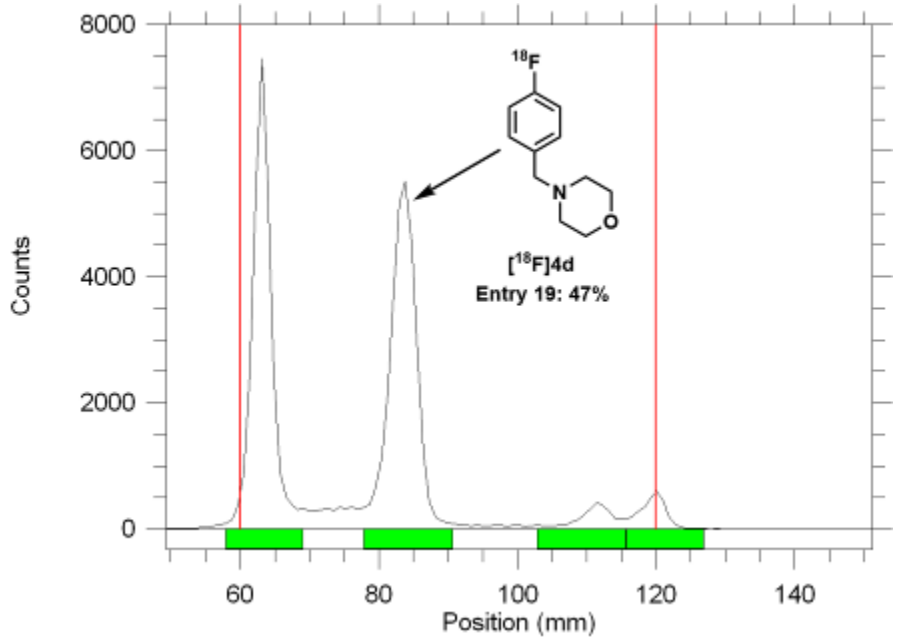
Figure S3. Example radioTLC scan of [^{18}F]4c



Entry 13 of Table S1. TLC eluent: hexanes.

Percent of total integration is listed for **[^{18}F]4c**.

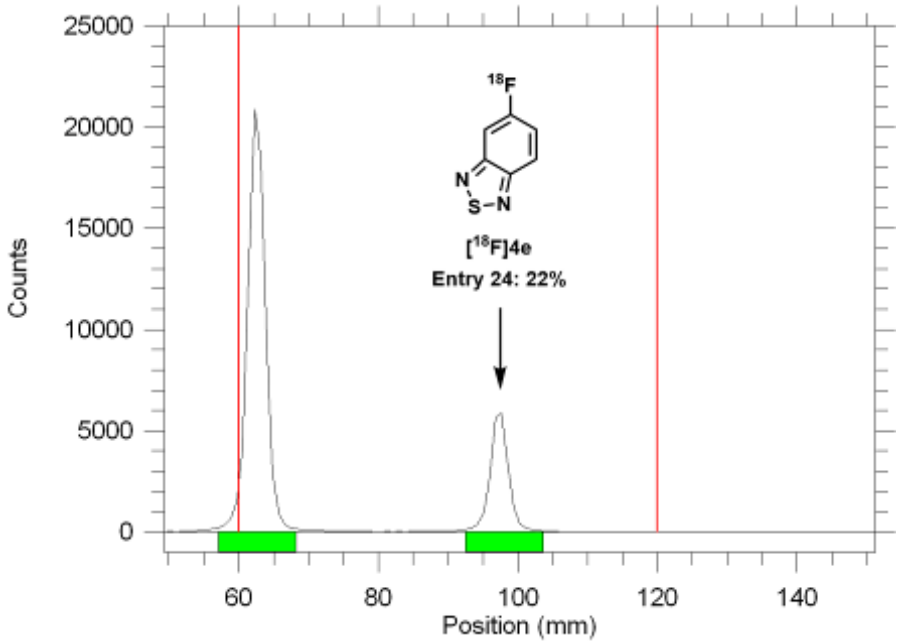
Figure S4. Example radioTLC scan of [^{18}F]4d



Entry 1 of Table S1. TLC eluent: 92:8:0.2 DCM/MeOH/Et_3N (v/v/v).

Percent of total integration is listed for **[^{18}F]4d**.

Figure S5. Example radioTLC scan of [^{18}F]4e



Entry 1 of Table S1. TLC eluent: 9:1 hexanes/EtOAc (v/v).

Percent of total integration is listed for **[^{18}F]4e**.

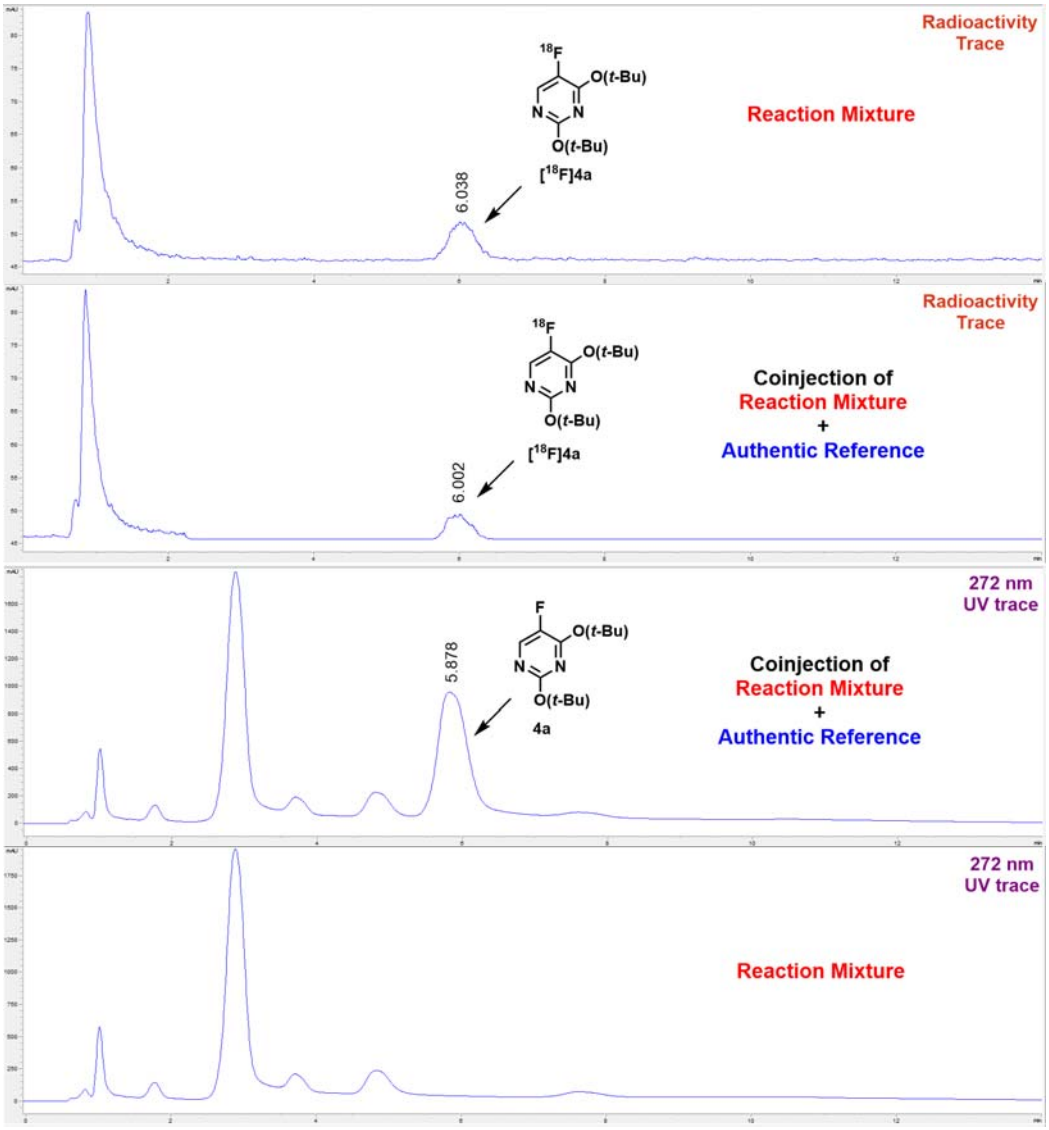
Characterization of ^{18}F-labeled molecules

All ^{18}F-labeled molecules were characterized by comparison of HPLC retention times to those of the corresponding authentic ^{19}F-containing reference samples by coinjection analysis.

An Eclipse XDB-C18 HPLC column (5 µm, 4.6 x 150 mm) was used, with an Agilent / HP Series 1100 HPLC instrument for analytical HPLC analysis.

Note: Radioactivity chromatographs are delayed by +0.125 min (for 2 mL/minute flow rates) in comparison to the UV chromatographs, since the radioactivity detector is positioned after the UV diode array detector in the flow path of the analytical HPLC instrument.

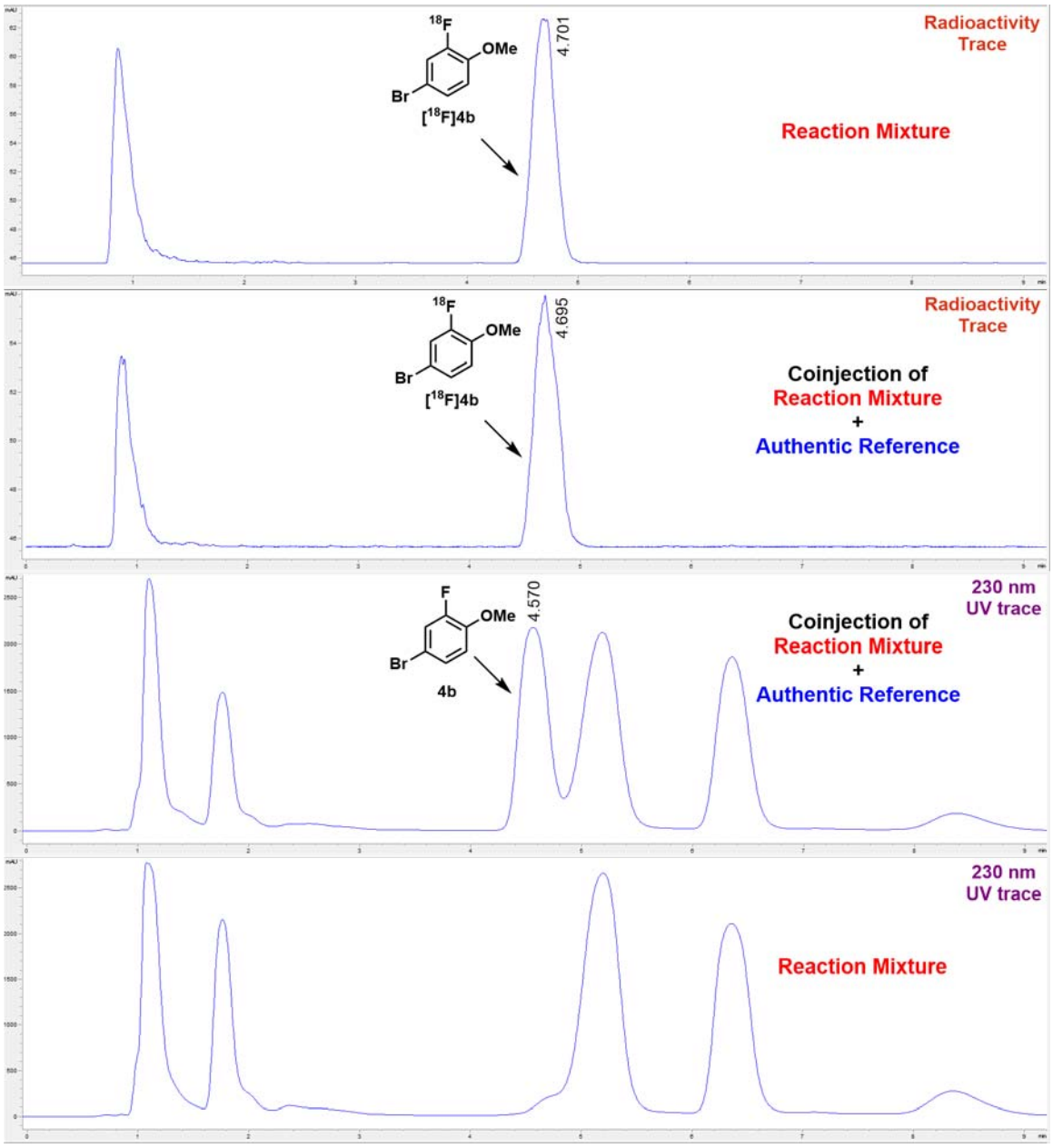
Figure S6. Characterization of [^{18}F]4a



HPLC Method: 50% MeCN, 50% H_2O (10 mM ammonium formate), flow rate = 2 mL/min.

Note: The radioactivity signal is delayed by +0.125 minutes relative to the UV signal.

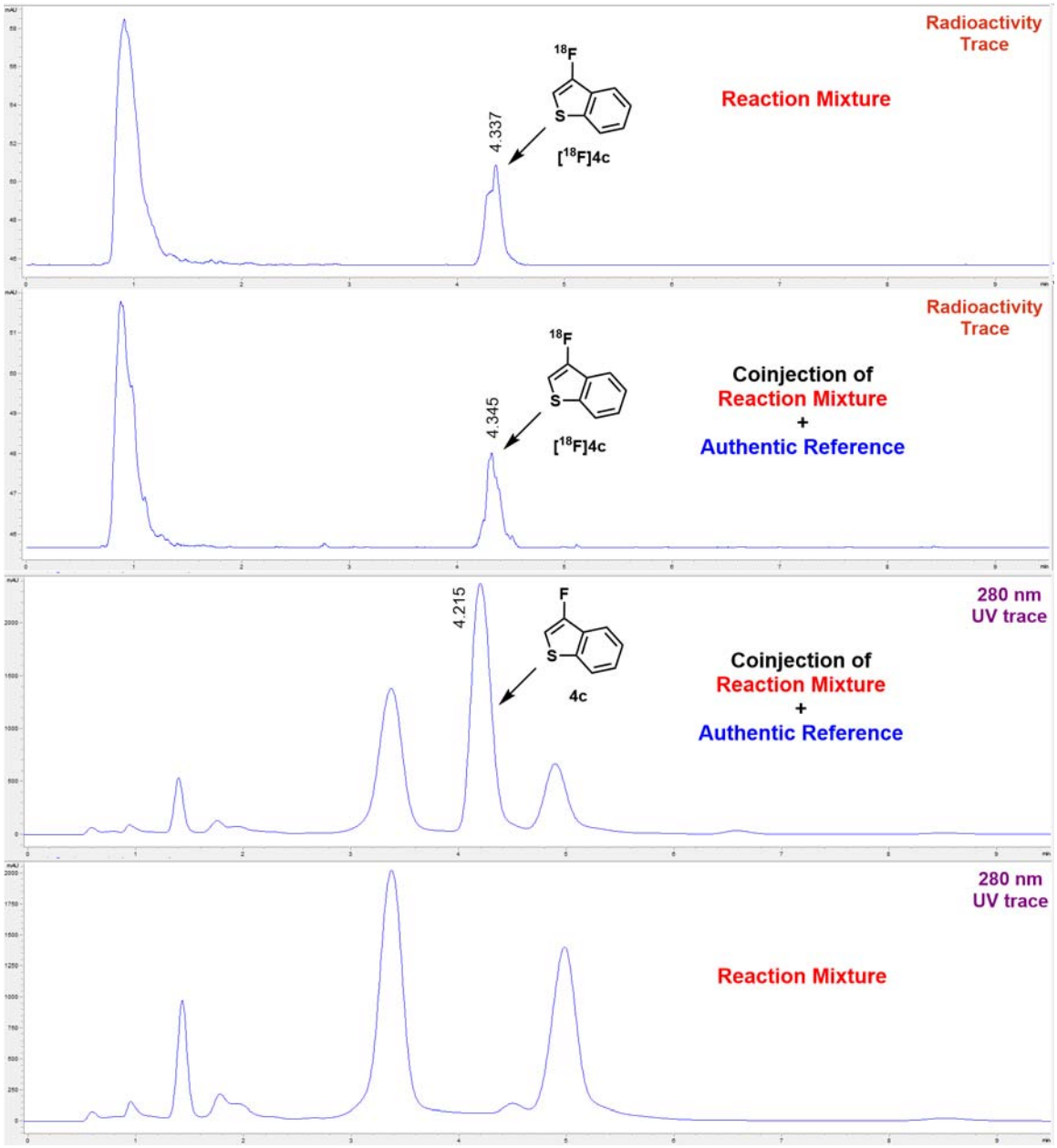
Figure S7. Characterization of [^{18}F]4b



HPLC Method: 45% MeCN, 55% H_2O (10 mM ammonium formate), flow rate = 2 mL/min.

Note: The radioactivity signal is delayed by +0.125 minutes relative to the UV signal.

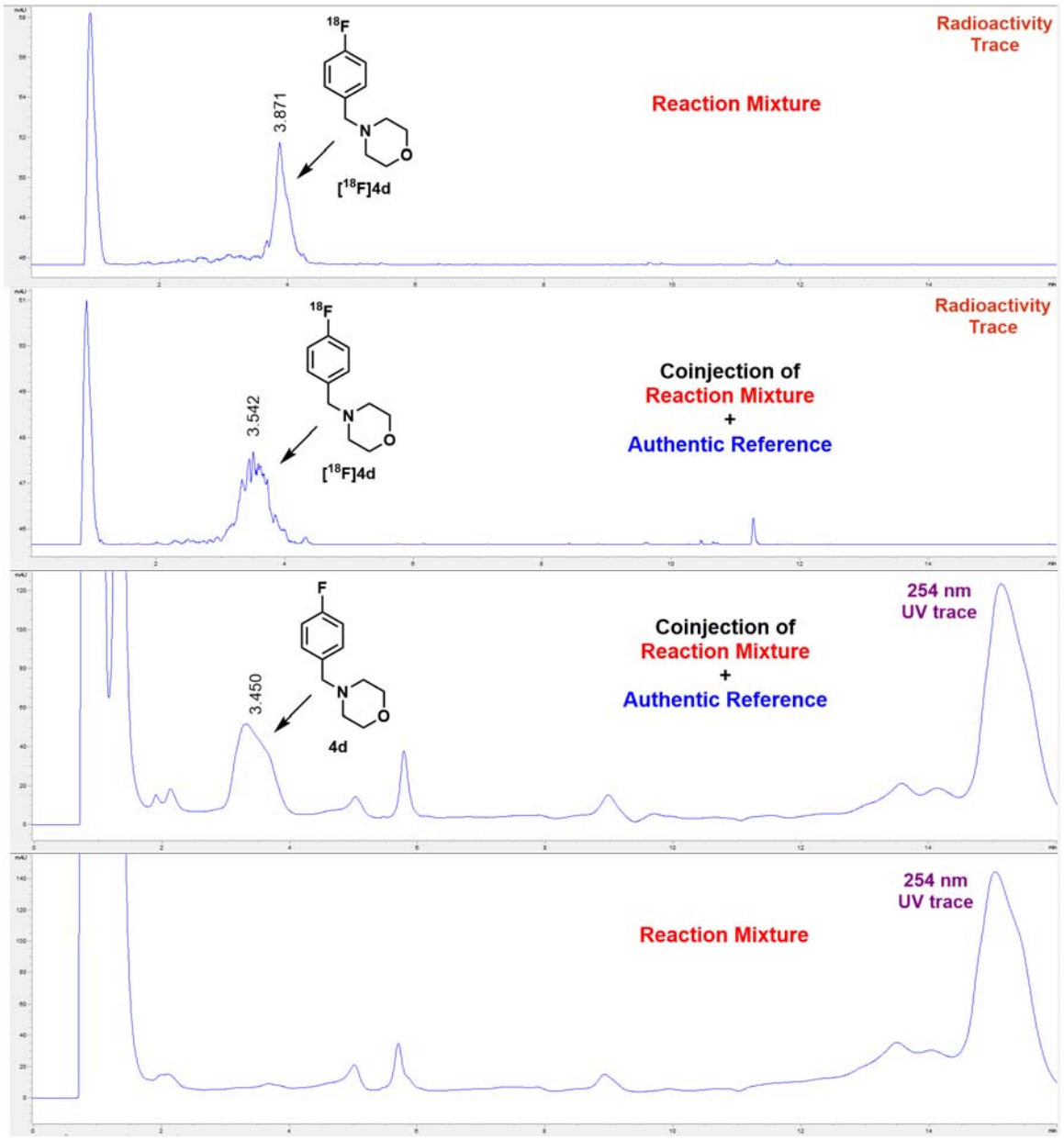
Figure S8. Characterization of [^{18}F]4c



HPLC Method: 50% MeCN, 50% H_2O (10 mM ammonium formate), flow rate = 2 mL/min.

Note: The radioactivity signal is delayed by +0.125 minutes relative to the UV signal.

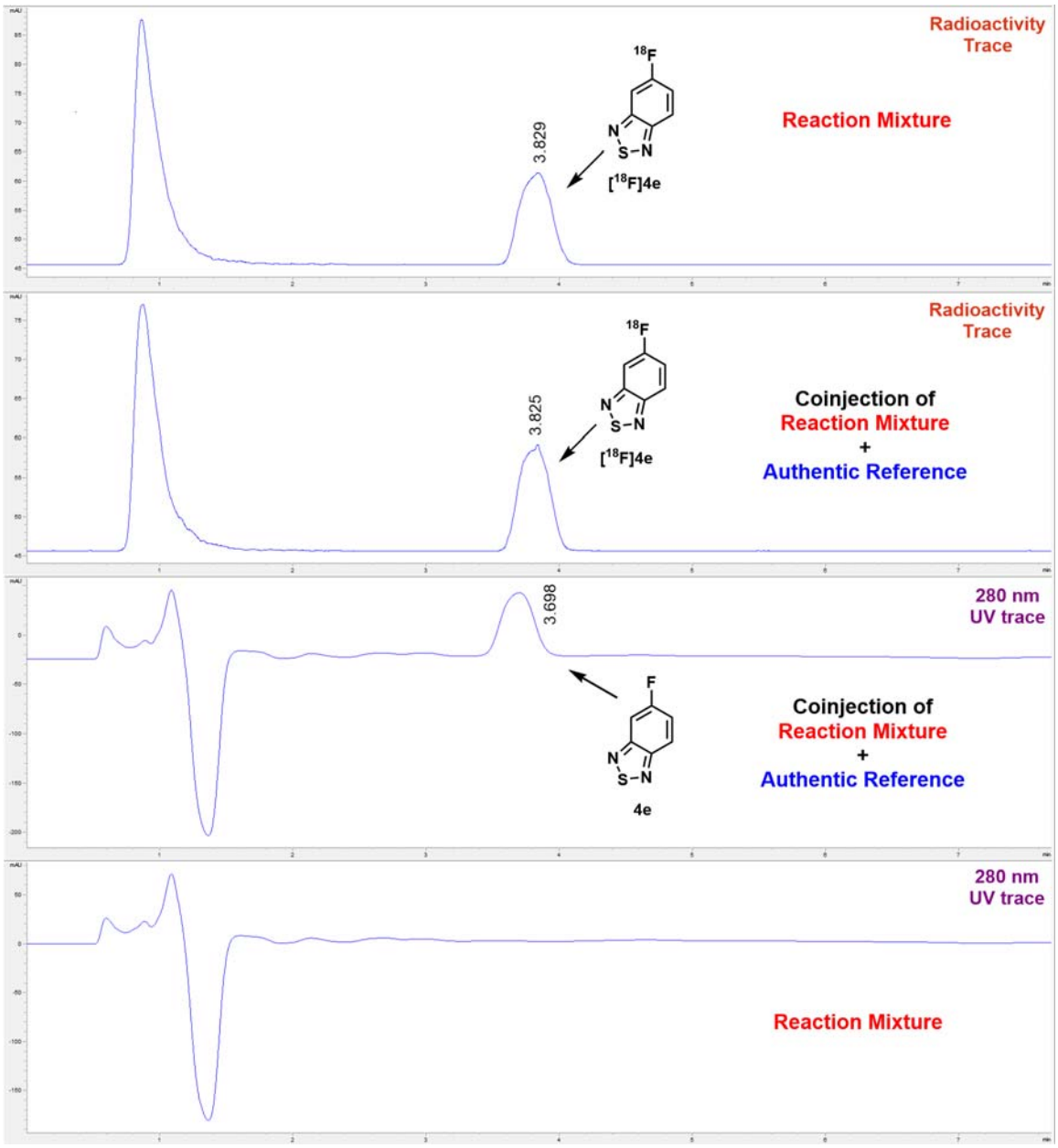
Figure S9. Characterization of [^{18}F]4d



HPLC Method: Gradient: Started with 5% A (A = 99.9:0.1 MeCN/TFA, v/v) and 95% B (B = 99.9:0.1 water/TFA, v/v) at time = 0; from 0 – 10 minutes, increase linearly to 20% A and 80% B. Flow rate = 2 mL/min.

Notes: The radioactivity signal is delayed by +0.125 minutes relative to the UV signal. The broadening of [^{18}F]**4d** peak shape in the coinjection compared to the unaltered reaction mixture is likely due to the increased amount of **4d** injected, and may reflect a change in the protonation state of [^{18}F]**4d**.

Figure S10. Characterization of [^{18}F]4e



HPLC Method: 35% MeCN, 65% H_2O (10 mM ammonium formate), flow = 2 mL/min.

Note: The radioactivity signal is delayed by +0.125 minutes relative to the UV signal.

First generation (preliminary, not cGMP) synthesis and isolation of [^{18}F]4a and [^{18}F]5-fluorouracil

Synthesis was accomplished using a Siemens Explora GN module in a lead-lined hot cell. The synthesis method was adapted from the previously reported synthesis of [^{18}F]MDL100907.[7]

Chemicals Preparation (Important: Unless otherwise specified, all chemicals should be prepared

on the synthesis day within two hours.)

• Prepare and condition Chromafix PS-HCO_3 IEX cartridge using aqueous 5 mg/mL TBAB as follows: Take out 30 mg of filling powder from Chromafix PS-HCO3 IEX cartridge; condition the cartridge with 2 mL of 5 mg/mL aqueous TBAB solution followed by rinsing the cartridge with 10 mL of SWFI.

• Prepare a PTC solution utilizing the following masses and volumes: Transfer prepackaged 26 mg of TBAB (purchased from ABX) in 1.2 mL of water (in two plastic vials) to a 10 mL sterile vial; Add another 2.8 mL of SWFI and 6 mL of anhydrous MeCN to the solution.

• Prepare the PPTS anhydrous MeCN solution as follows: add 4.0 μL of a 0.25 M PPTS anhydrous MeCN stock solution to a conical GC vial; add 0.6 mL of anhydrous MeCN to the conical GC vial. The GC vial was then capped and sealed with parafilm. (0.25 M PPTS stock solution was prepared as follows: dissolve 63 mg of PPTS solid in 1.0 mL of anhydrous MeCN.)

Explora GN Modules Cleaning and Preparation (IMPORTANT: Precursor and transfer lines need to be cleaned within two hours of synthesis with FRESH anhydrous MeCN!)

• Clean Explora modules and F18 line with SWFI (sterile water for injection) and acetonitrile.

• Clean precursor line 2 as follows: Load FRESH anhydrous MeCN to reagent 2 position; Use a new conical GC vial and clean the line with 10 mL of MeOH; Clean the line with 10 mL of acetone; Clean the line with 15 mL of anhydrous MeCN.

• Clean transfer line 2 as follows: Load FRESH anhydrous MeCN to reagent 2 position; Use a NEW test tube; Clean the line with anhydrous acetonitrile.

• Fill the Explora GN reagent vials with the following solvents: Reagent 1: SWFI; Reagent 2: MeCN; Reagent 3: Empty; Reagent 4: PTC; Reagent 5: Empty; Reagent 6: Empty.

• Prime the Explora GN with the “PTC” at position 4.

• Attach the IEX cartridge to F-18 line 2 on the Explora GN. Replace the old reaction vessel with a clean one.

Addition lines Cleaning and Preparation (IMPORTANT: Addition lines need to be cleaned within two hours of synthesis with FRESH anhydrous MeCN!)

• Clean main addition loop line as follows: Clean the line with 10 mL of anhydrous MeCN and then dry the line by passing high-flow N_2 through for 20 minutes to make sure the line is dry and there is NO residue MeCN left in the line. Purification Modules Cleaning (HPLC and columns may be cleaned one day prior to the synthesis.)

• Clean HPLC semiprep column and analytical column with a mixture of MeCN and H_2O (90:10 v/v).

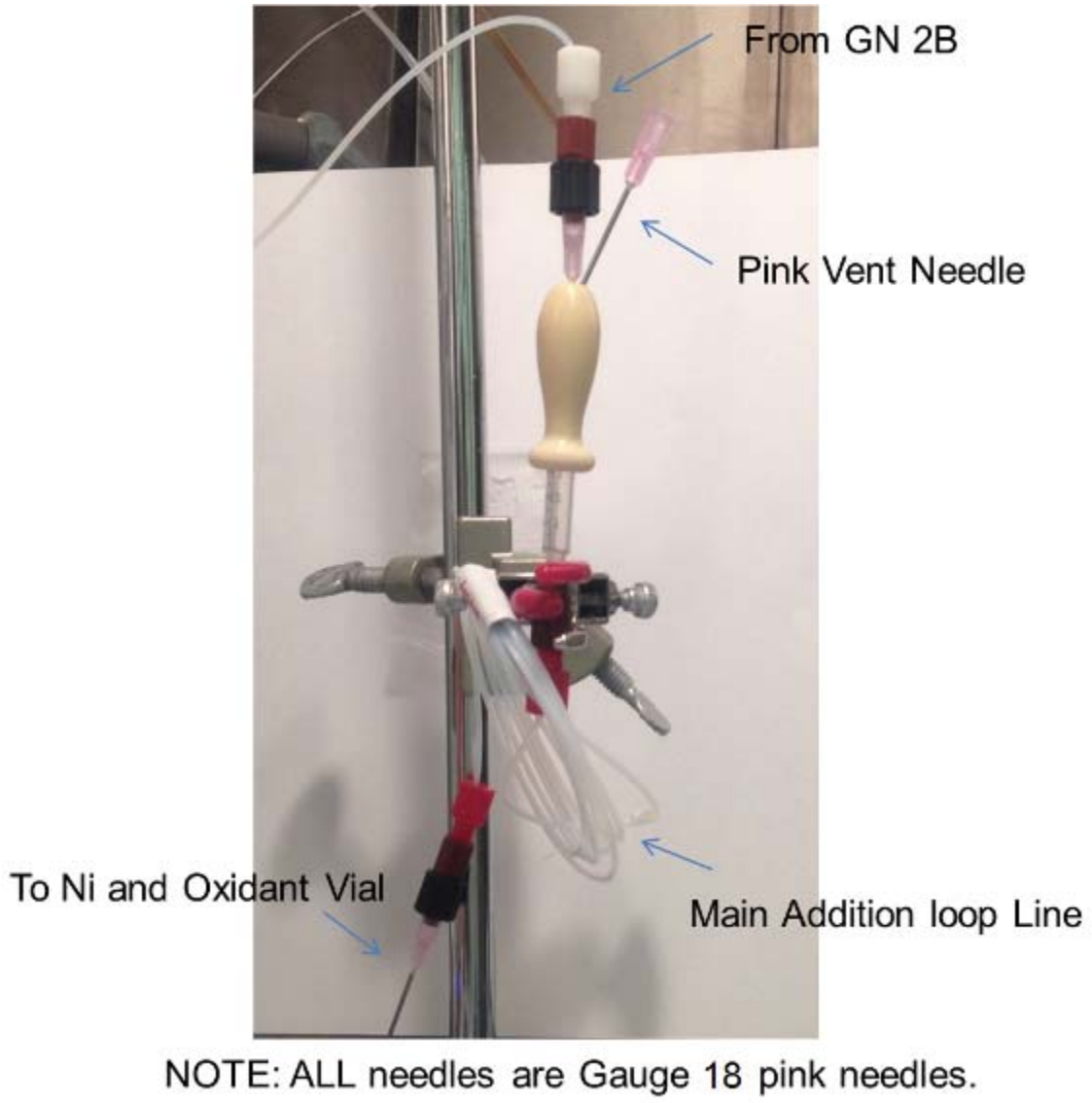
• Rinse the semiprep column with 60% MeCN, 40% (10 mM ammonium formate in H_2O).

Main Addition Line Setup

Connect a 1 mL syringe to the main addition loop line, seal the 1 mL syringe with a new rubber bulb and place a G18 pink vent needle on the rubber bulb in accordance with Figure S11. The

rubber bulb is then connected to "GN 2B line" from Explora GN in the hot cell. The main addition line is clamped on a stand as shown in Figure S11.

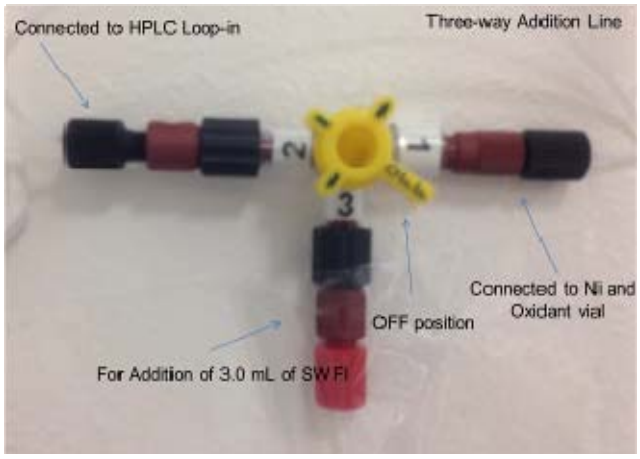
Figure S11. Main addition line



Three-Way Addition Line Setup

Attach a three-way stopcock with two female luers and 1 male luer to an HPLC Loop-in port, a line leading to a syringe with 3.0 mL of H_2O, and a line leading to a needle that will later be inserted to the reaction vial (containing Ni and oxidant).

Figure S12. Three-Way Addition Line

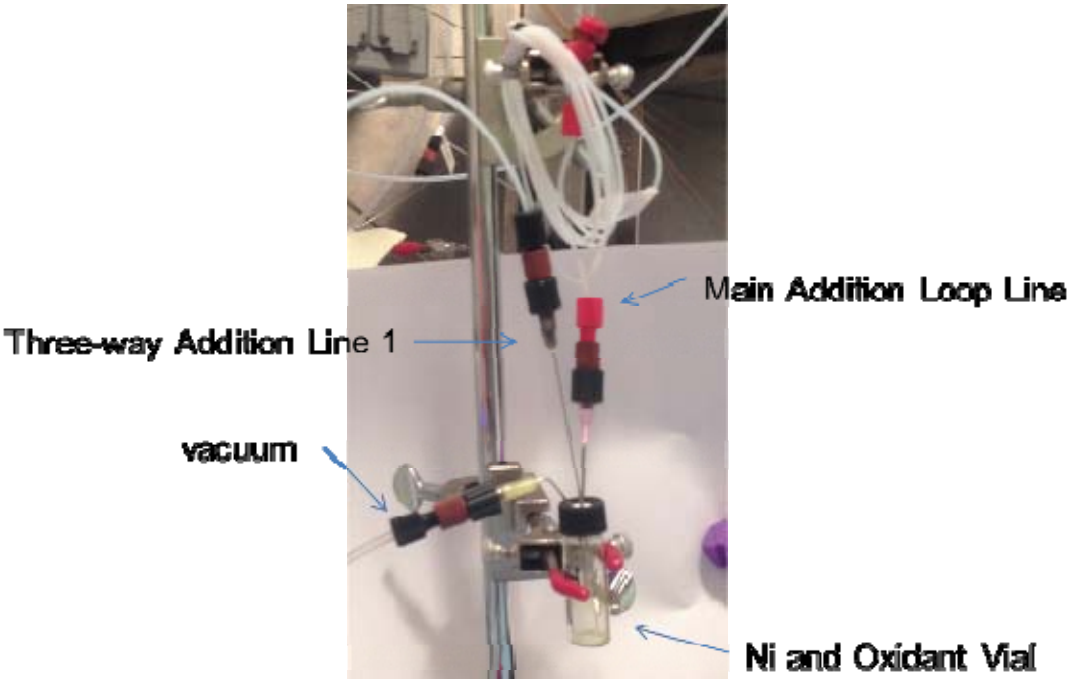


Reaction Vial Setup

In a glovebox under an atmosphere of dry nitrogen, to an oven-dried 4 mL 1-dram glass vial, add 2.0 mg of **2** and 2.0 mg of 1,1'-(phenyl-λ^3-iodanediyl)bis(4-methoxypyridinium)bis(trifluoromethanesulfonate)[1]. Seal the vial with a screw cap containing an inserted Teflon-lined septum. Remove the vial from the glovebox.

Immediately prior to execution of the synthesis, perforate the reaction vial with needles leading to the Main Addition Loop Line, Three-Way Addition Line 1, and vacuum line (Figure S13). At this time, also attach precursor line 2 to the PPTS anhydrous MeCN solution.

Figure S13. Reaction Vial Setup



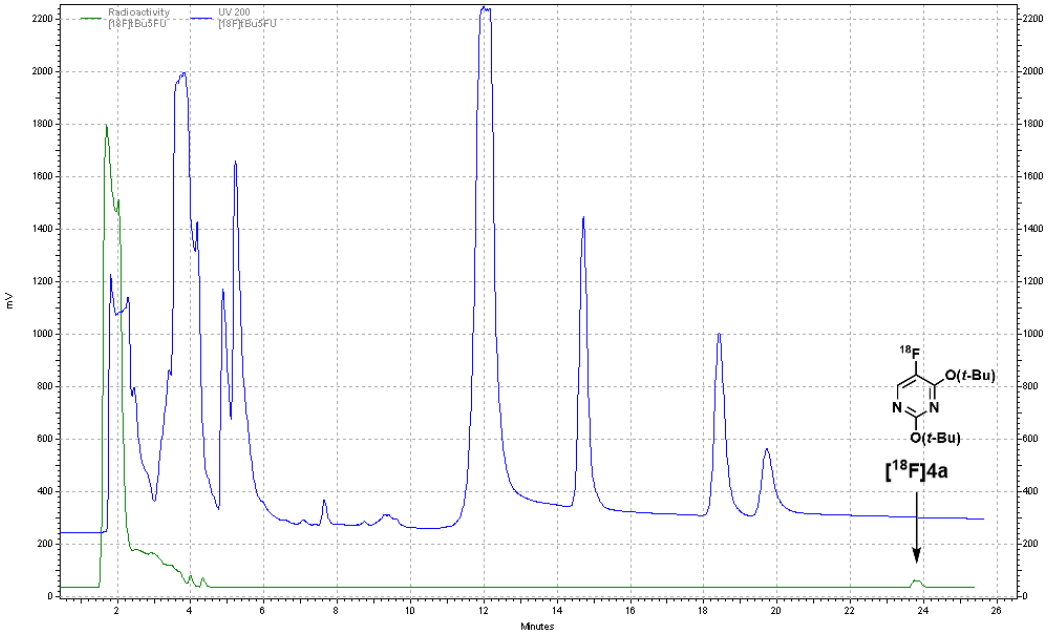
Synthesis Execution

The synthesis began when 0.98 Ci of [^{18}F]fluoride was loaded directly from the cyclotron to the IEX cartridge in the synthesis module. 0.6 Ci was eluted from the cartridge after 4 minutes. Azeotropic drying was then performed, and then 0.25 Ci of [^{18}F]fluoride as a solution in dry MeCN with TBAB and PPTS was passed through Transfer Line 2 into the Main Addition Loop Line. After this transfer was complete, vacuum was applied via the vacuum line leading to the reaction vial, which caused rapid (<1 second) addition of the [^{18}F]fluoride solution to the reaction vial (this rapid addition of the entire solution at once is essential) (addition occurred at 45 minutes after start of synthesis). Water (3.0 mL) was then added via Three-way Addition Line 1, resulting in a yellow mixture, which was then drawn to the HPLC Loop-in via the Three-way Addition Line 1, by slowly pulling on a syringe connected to the other side of HPLC Loop-in, so that the reaction mixture was loaded into HPLC Loop-in with minimal bubble formation.

Semiprep HPLC purification was conducted with a Phenomenex Luna C18(2) column (5 µm, 10.00 x 250 mm), eluting with 60:40 MeCN/(10 mM ammonium formate in water) v/v, flow rate = 5 mL/minute (Figure S14).

Compound **[^{18}F]4a** was collected from the semiprep HPLC purification (at 23.6 minutes after injection, 0.6 minute collection window, Figure S14) in 2.9 mL of HPLC mobile phase. In order to remove MeCN and ammonium formate, the collected product was diluted with H_2O (20.0 mL) and passed through a Waters C18 Sep-Pak Plus (which had previously been preconditioned by washing first with ethanol and then water). The product was eluted from the Sep-Pak with ethanol (2.0 mL), to afford a solution containing 2.37 mCi of **[^{18}F]5** (at 95 minutes after the start of the synthesis).

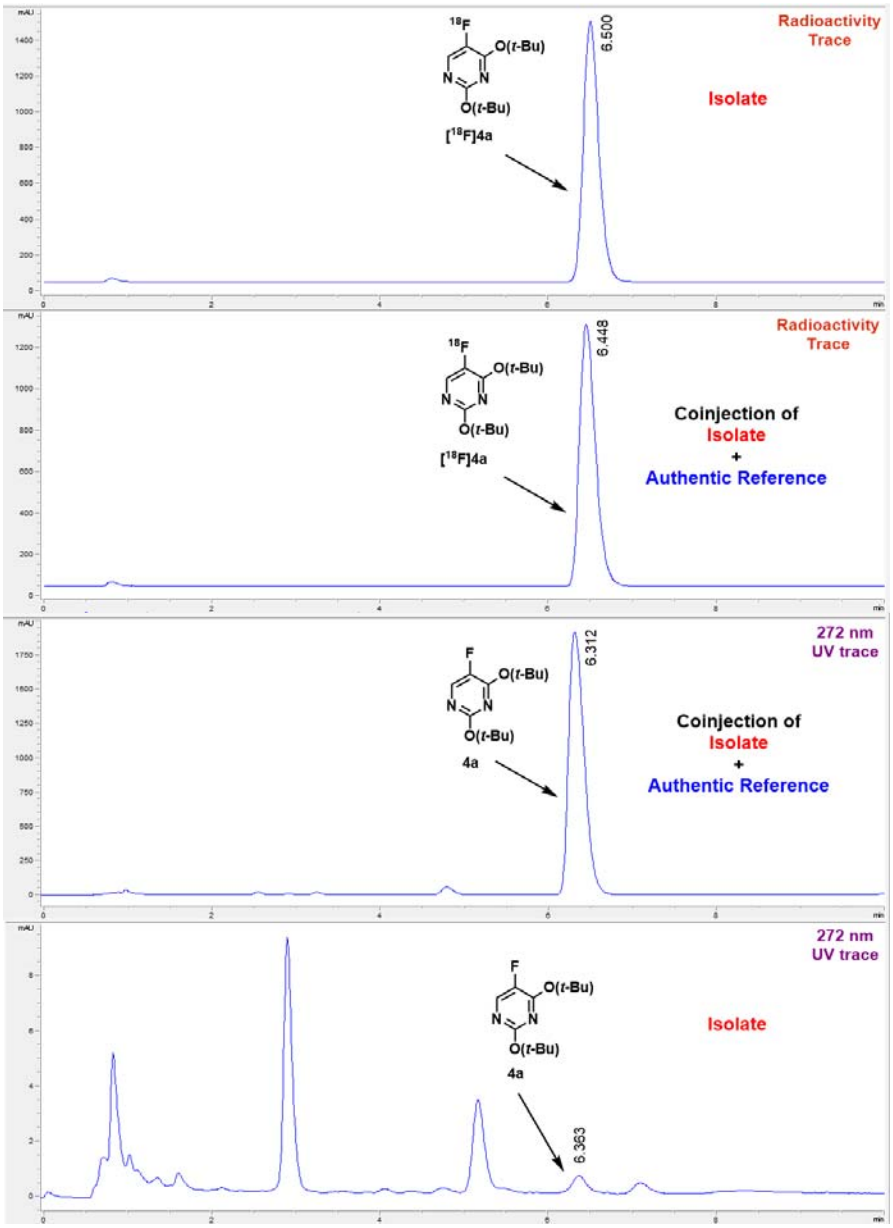
Figure S14. Semipreparative HPLC purification of [^{18}F]4a.



HPLC method: 60:40 MeCN/(10 mM ammonium formate in water) v/v, flow rate = 5 mL/minute

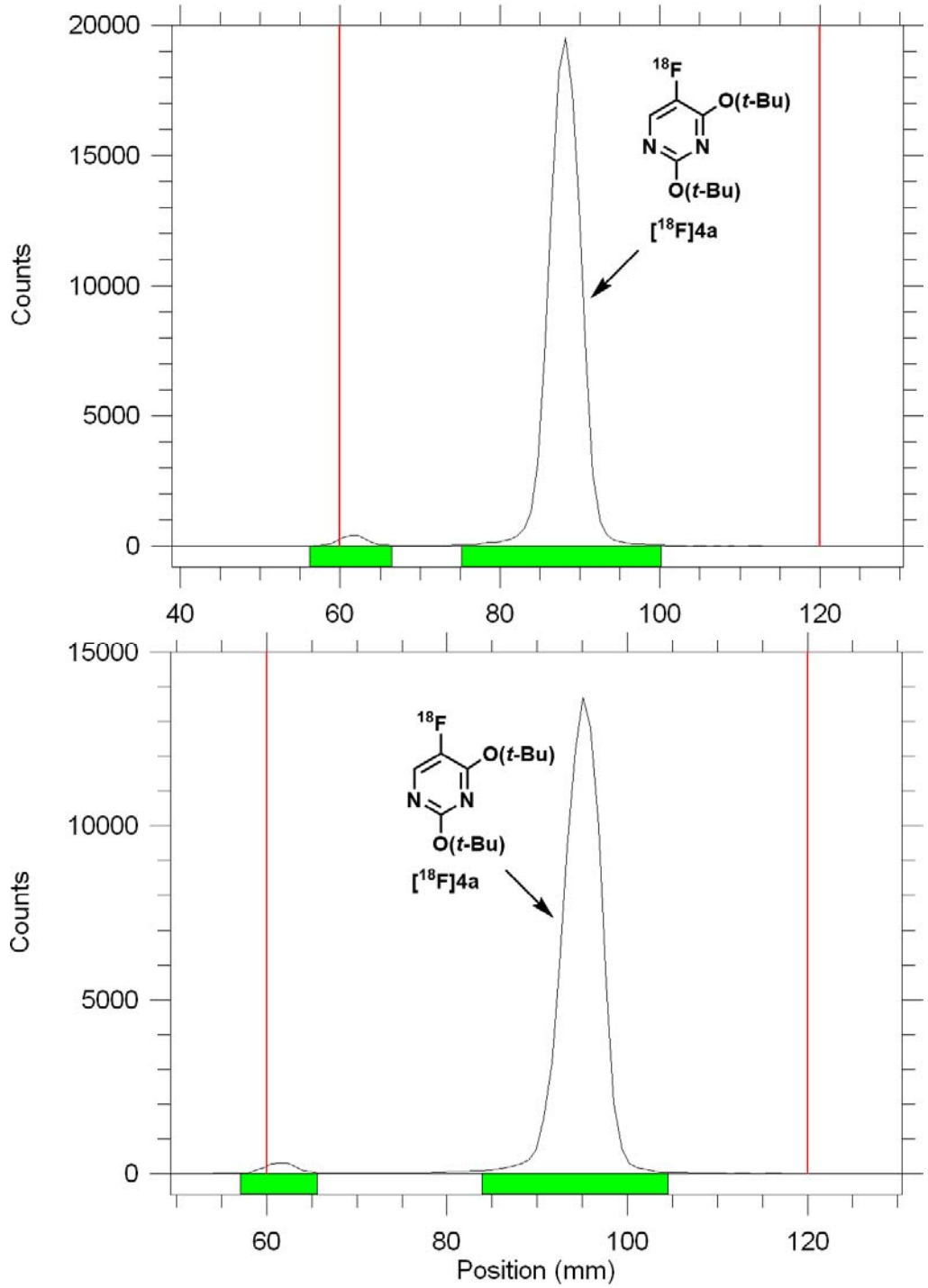
Deprotection was carried out by mixing the ethanol solution of **[^{18}F]4a** with conc. HCl (0.23 mL, 37% by weight in water, 2.8 mmol HCl). The resulting solution was allowed to stand at 23 ºC for 2 minutes, and was then neutralized with $NaHCO_3$ (2.9 mL, 1.0 M solution, 2.9 mmol $NaHCO_3$), and diluted with water (14.9 mL) to afford a solution of [^{18}F]5-fluorouracil (quantitative conversion).

Figure S15. HPLC characterization of isolated [^{18}F]4a.



Method: 60:40 MeCN/(10 mM ammonium formate in water) v/v, flow rate = 2 mL/minute
Column: Eclipse XDB-C18 (5 µm, 4.6 x 150 mm)

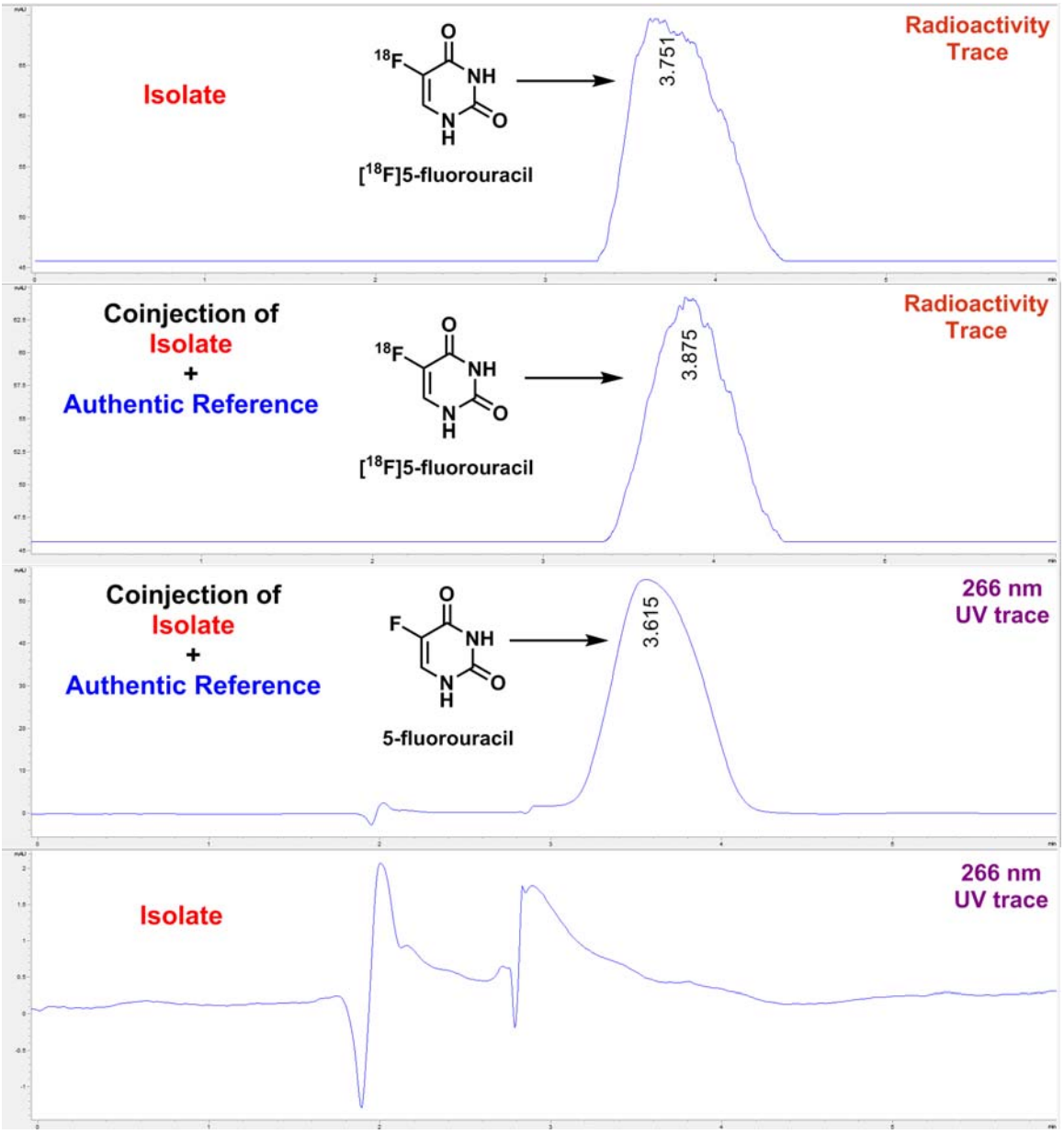
Figure S16. TLC characterization of isolated [^{18}F]4a.



Top: RadioTLC of isolated **[^{18}F]5**. Eluent: 95:5 Hexanes/EtOAc (v/v). **[^{18}F]4a**: R_f = 0.47. **4a** (Authentic Reference): R_f = 0.48. Radiochemical purity > 98%.

Bottom: RadioTLC of isolated **[^{18}F]5**. Eluent: 96:4 Hexanes/THF (v/v). **[^{18}F]4a**: R_f = 0.58. **4a** (Authentic Reference): R_f = 0.59. Radiochemical purity > 98%.

Figure S17. HPLC characterization of isolated [^{18}F]5-fluorouracil.

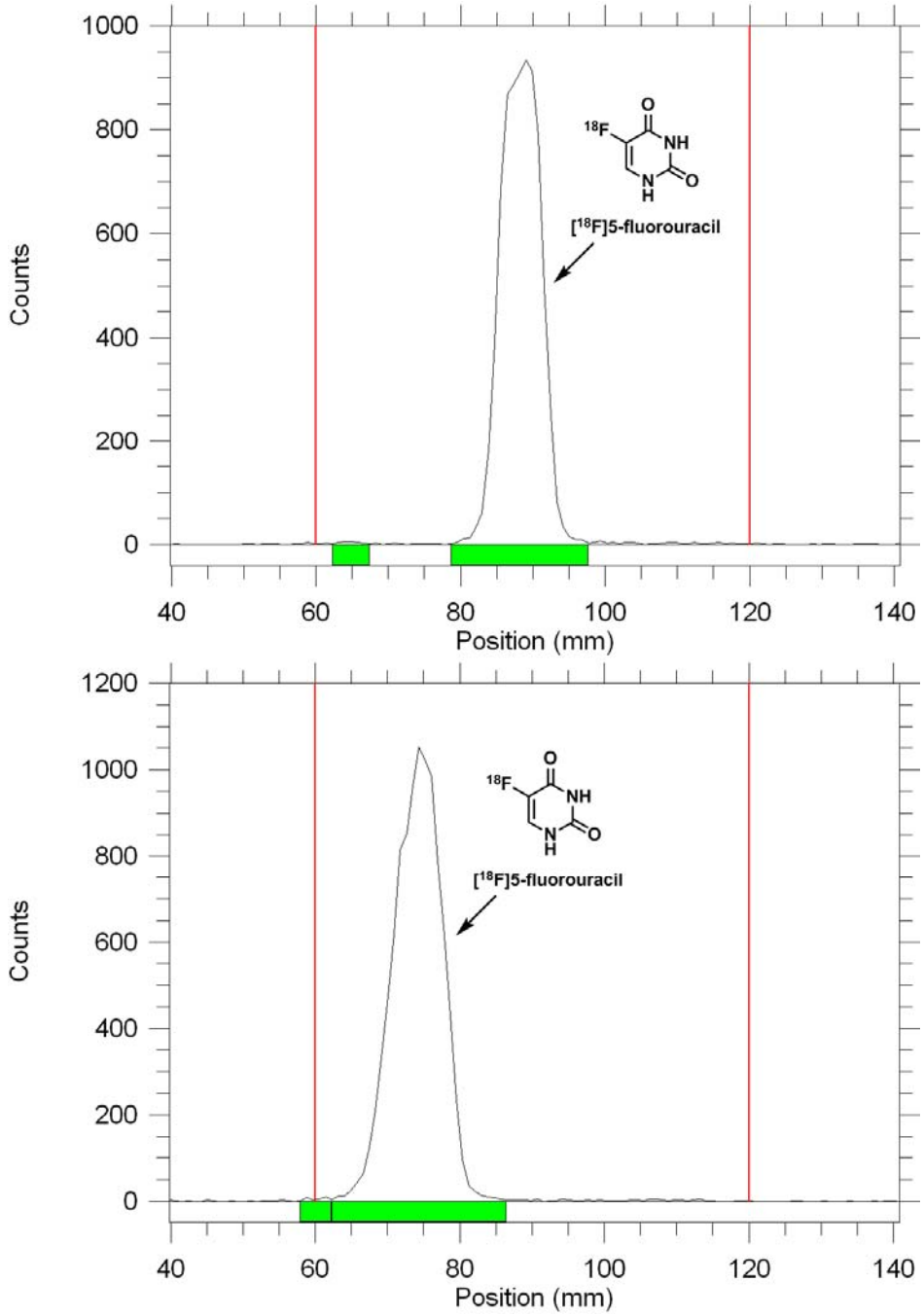


HPLC Method: 65 mM ammonium formate in water, flow = 1 mL/min.

Column: Luna Pentafluorophenyl (PFP) (2) (5 µm, 4.6 mm x 150 mm).

Note: The radioactivity signal is delayed by +0.250 minutes relative to the UV signal.

Figure S18. TLC characterization of isolated [^{18}F]5-fluorouracil.



Top: RadioTLC of isolated [^{18}F]5-fluorouracil. Eluent: 85:15:2 MeCN/H_2O/NH_3(25% aq.) (v/v/v). [^{18}F]5-Fluorouracil: R_f = 0.47. 5-Fluorouracil (Authentic Reference): R_f = 0.46. Radiochemical purity > 99%.

Bottom: RadioTLC of isolated [^{18}F]5-fluorouracil. Eluent: 75:25:2 DCM/MeOH/NH_3(25% aq.) (v/v/v). [^{18}F]5-Fluorouracil: R_f = 0.24. 5-Fluorouracil (Authentic Reference): R_f = 0.23. Radiochemical purity > 99%.

Determination of specific activity of [^{18}F]4a

Specific activity of **[^{18}F]4a** was determined by dividing the radioactivity of a sample of **[^{18}F]4a** by the amount of (**[^{18}F]4a + 4a**) in the sample. The number of moles of (**[^{18}F]4a + 4a**) in an isolated sample was determined by measuring the UV signal at 272 nm, and converting the UV signal intensity to number of moles using a standard curve.

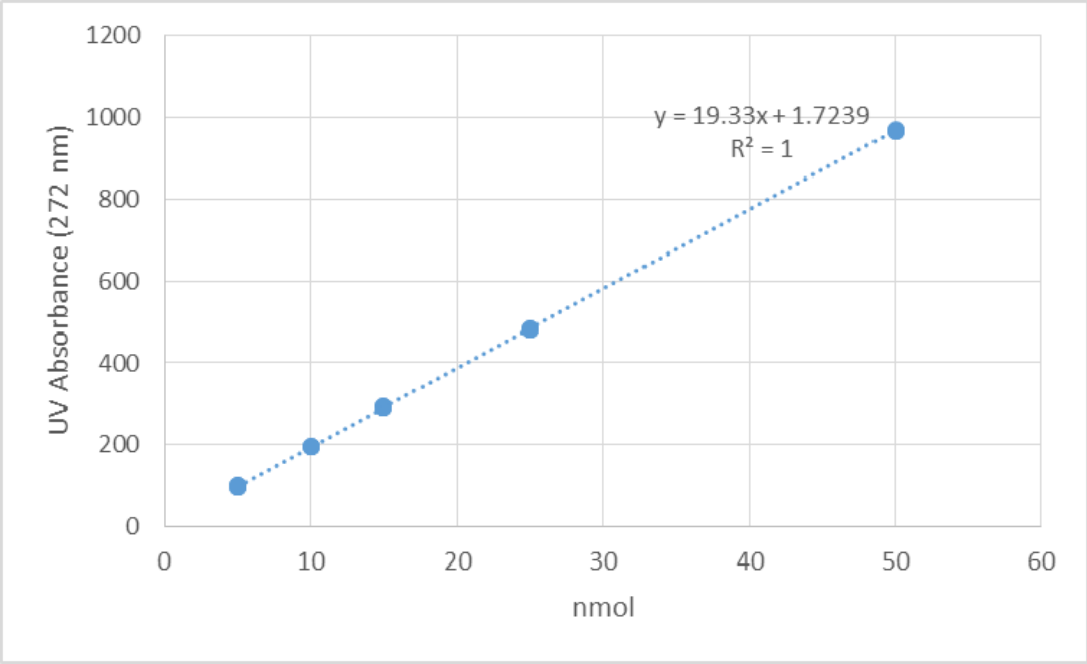
For 105 μCi of **[^{18}F]4a** a UV absorbance (at 272 nm) of 8.2 was measured, corresponding to 0.335 nmol, for a specific activity of 0.31 Ci/μmol (12 GBq/μmol) at time of injection (TOI) to the analytical HPLC instrument.

The calibration curve was generated by integration of the UV absorbance signal intensity (at 272 nm) of different known amounts of **4a**, in triplicate for each amount (see Table S2 and Figure S19).

Table S2. Data for the standard curve of UV absorbance vs. amount of 4a

Amount of **5** (nmol)	UV absorbance (272 nm)
5	98.2
5	98.5
5	98.1
10	195.3
10	194.4
10	195.3
15	293.4
15	293.3
15	292.4
25	483.9
25	483.3
25	482.7
50	969.5
50	968.2
50	968.3

Figure S19. Calibration curve for UV absorbance vs. amount of 4a



cGMP synthesis of [^{18}F]5-fluorouracil: general information

The large scale synthesis of [^{18}F]5-FU was accomplished using a new concentrator instrument that is diagrammed in Figure S20. The instrument was assembled from Rheodyne 6-port switching valves, a 10-port Vici selector valve, and the following Upchurch Scientific tubings: 0.01”ID, 1/16”OD PEEK tubing (for eluent loop and anion exchange cartridge lines); 0.02”ID, 1/16”OD PEEK tubing (for line from common vial to valve, and eluent spout); 0.02”ID, 1/16”OD PFA HP plus tubing (for lines with liquid sensors); 0.03”ID, 1/16”OD ETFE tubing (for other liquid lines). 1/16”ID, 1/8”OD polyurethane tubing from McMaster-Carr was used for gas lines. The electronic valves were controlled via LabView® on a computer.

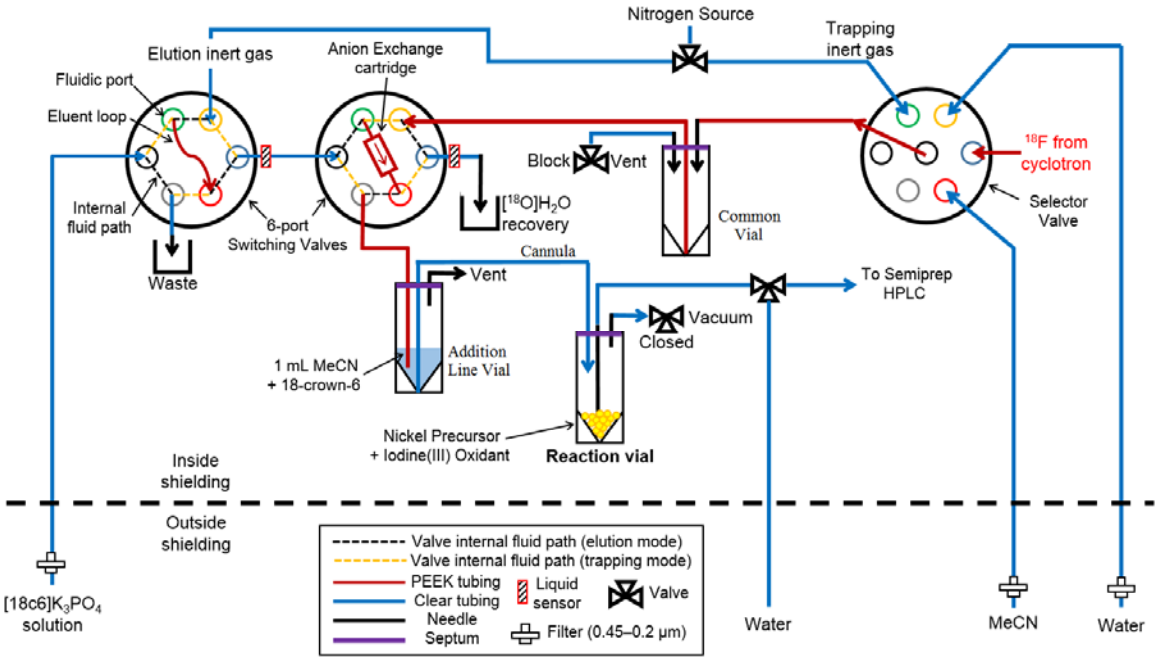
Wheaton V-Vials (3 mL) were used in combination with 20 mm butyl rubber septa and 20 mm aluminum crimp caps for the Common Vial and Addition Line Vial . For the Reaction Vial, a 10 mL 24-400 Wheaton V-Vial with a screw-on cap and insertable PTFE-lined septum was used.

The Opti-Lynx micro-anion exchange cartridge (and its holster with adapters to microfluidic lines) was purchased from Optimize Technologies, and filled with BioRad® AGMP-1 Resin by the manufacturer.

The radiopharmaceutical production facility, reagents, documentation, etc. were controlled in a cGMP environment. Ethanol, 37% aq. HCl, and 1M aq. sodium bicarbonate were USP grade. Sterile Water for Injection (SWFI) was used in purification and formulation. All liquids and air introduced to the concentrator instrument were filtered through 0.45 micron Phenomenex filters

(RC for water or 4:1 MeCN/water (v/v) solutions (including eluent for [^{18}F]fluoride elution); PTFE for MeCN or 1 M NaOH). Dry MeCN was purchased from Acros in bottles sealed with a needle-permeable membrane. Aristar Ultra water was used for instrument cleaning, cartridge conditioning, and eluent solution preparation.

Figure S20. Schematic of F18 concentrator and completed setup for production of [^{18}F]5-fluorouracil

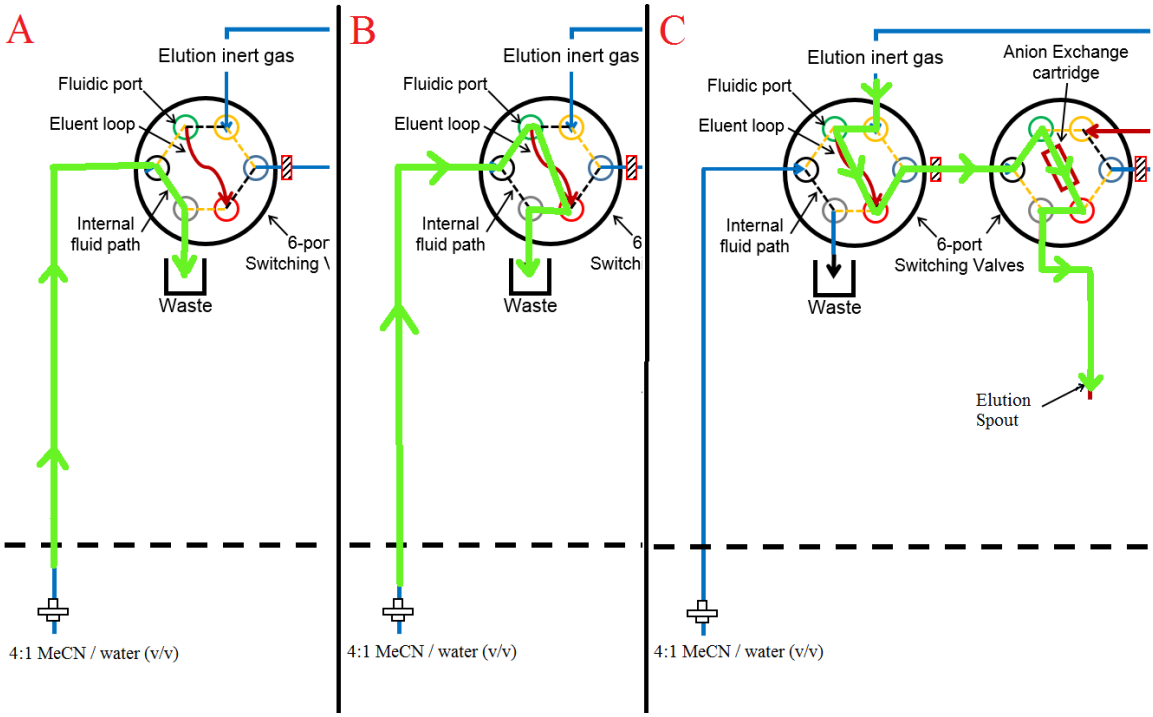


Full procedure for synthesis of [^{18}F]5-fluorouracil

1. The instrument modules were cleaned in preparation for [^{18}F]5-Fluorouracil production.

1.1 The flow pathways for elution were cleaned.

Figure S21. Flow pathways for elution.



1.1.1 The green lines in Figure S21 panel A were cleaned with 4:1 MeCN / Aristar Ultra water (v/v).

1.1.2 The green lines in Figure S21 panel B were filled with 4:1 MeCN / Aristar Ultra water (v/v).

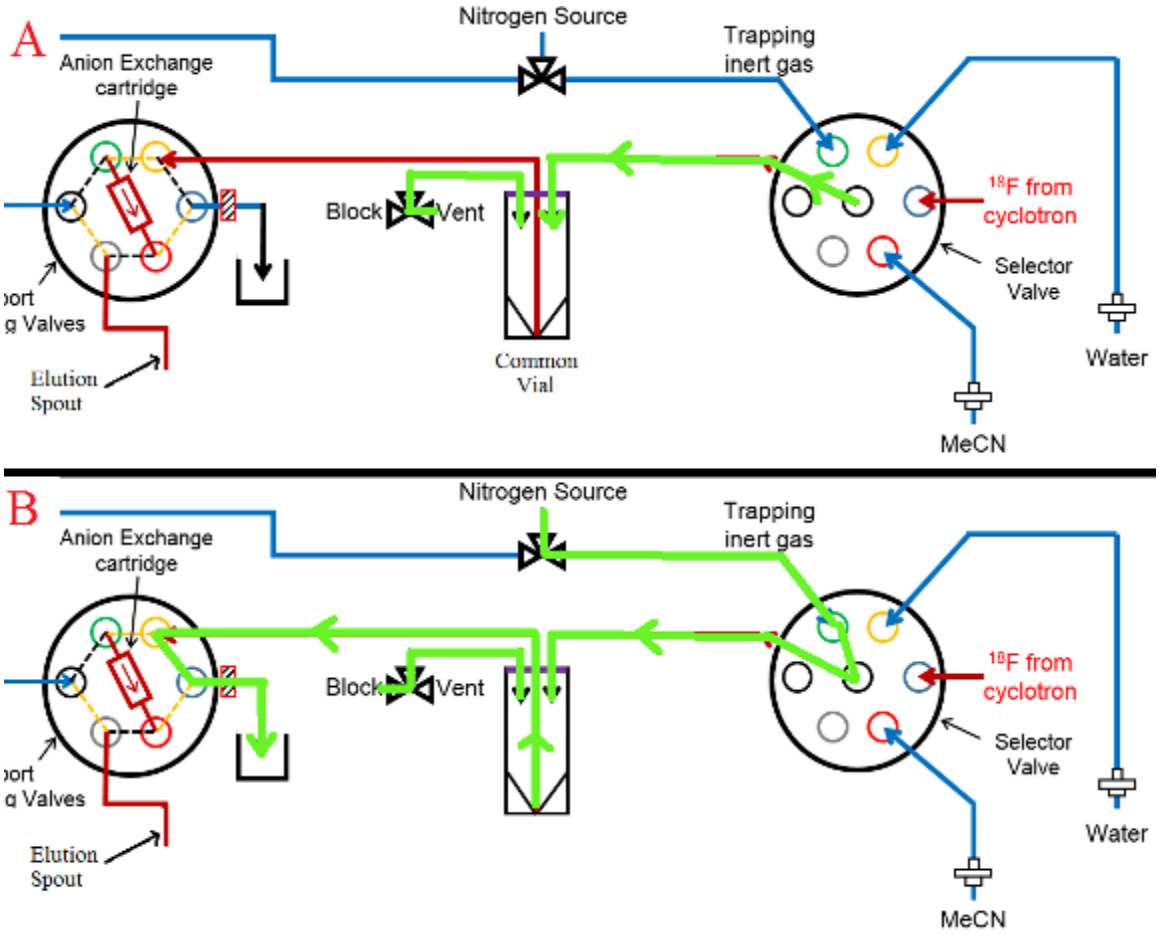
1.1.3 Nitrogen gas was applied as shown in the green lines in Figure S21 panel C, to clean the Elution Spout.

1.1.4 Steps 1.1.2 and 1.1.3 were repeated 9 times.

1.1.5 The lines in Figure S21 panels A, B and C were emptied by passage of air or nitrogen.

1.2 The common vial and associated flow pathways were assembled and cleaned (Figure S22).

Figure S22. Common vial and associated flow pathways



1.2.1 The Common Vial was assembled from a 3 mL V-vial, septum, and crimped cap. The common vial was perforated with the PEEK tubing leading to the 6-port switching valve containing the trapping cartridge. The common vial septum was also perforated with needles on the termini of tubing leading to the block/vent line, and the selector valve.

Important: 18G needles must be used for attachment to the common vial, to prevent pressure build-up during F18 delivery from cyclotron.

1.2.2 Aristar Ultra Water (1 mL) was added through the selector valve to the common vial, through the pathway highlighted in green in Figure S22 panel A.

1.2.3 Nitrogen gas was applied (15 psi) to push the liquid from common vial to waste, as shown in Figure S22 panel B.

1.2.4 Steps 1.2.2 and 1.2.3 were repeated, but with MeCN (1 mL), and the lines were dried by passage of nitrogen.

1.3 The Addition Line Vial was assembled from a 3 mL V-vial, septum, and crimped cap. The cannula was perforated through the septum of the MeCN dilution vial (see Figure S22). Both the

vial and cannula were cleaned with 4:1 MeCN / Aristar Ultra water (v/v), then with anhydrous MeCN, and dried with a stream of nitrogen until no trace of liquid remains.

1.4 The water dilution / HPLC loading line valve was assembled and cleaned as follows.

1.4.1 A Teflon three-way stopcock was attached to three lines as follows (Figure S23). **Position 1** was attached to tubing that leads to a 16G spinal needle. **Position 2** was connected via tubing to a female Luer so that wash solvents could be passed through the valve via syringe. **Position 3** was closed off, so that only **Position 1** and **Position 2** were connected.

Figure S23. Assembly of three-way water dilution / HPLC loading line valve

1.4.2 A solution of 4:1 MeCN / Aristar Ultra water (v/v) was passed through **Position 2** to **Position 1**, to clean the valve and the 16 G needle. Then, anhydrous MeCN was passed through the same line. The 16 G spinal needle was thoroughly scrubbed with MeCN to remove residues from needle manufacture. The line was then dried with a stream of nitrogen until no trace of liquid remained.

2. Component Preparation

2.0 The flow rate of water, when passed through the Opti-Lynx cartridge as shown in Figure S29 (F18 trapping pathway), should be at least 2.4 mL per 10 minutes at nitrogen pressure of 15 psi. When flow rate was not adequate for a new cartridge, the cartridge was filled with water, placed in a 20 mL scintillation vial filled with water, and the vial was sonicated. Generally, water flow rate through the cartridge improved after sonication. Additionally, to improve flow rate, the lines can be disconnected from the ports on the switching valves, and MeCN can be passed through the valve ports (in the opposite direction to the usual direction of flow) to flush out accumulated particulates.

2.1 The Opti-Lynx miniature ion exchange cartridge was preconditioned as follows. To avoid introducing salt into the F18 trapping line, the Opti-Lynx cartridge inlet port was disconnected from the F18 trapping line. The eluent waste line (cleaned during step 1.1) was unscrewed, and then screwed into the inlet port to the Opti-Lynx cartridge. A 3 mL conical vial with septum and

crimped cap was attached to the needle of the eluent waste line. The block/vent line, as well as the nitrogen line from the selector valve, were removed from the common vial septum, and affixed to the 3 mL vial via needles through septum. Sodium hydroxide solution (1M in water, 1 mL) was added via syringe, and was pushed with nitrogen (15 psi) through the Opti-Lynx cartridge. This process was then repeated, but with Aristar Ultra water (2 times 1 mL), then with MeCN (1 mL), followed by nitrogen to dry the lines and cartridge. The eluent waste line and F18 trapping line were then reconfigured to their original connectivities prior to cartridge preconditioning. The needles on the block/vent line and selector valve line were replaced with clean 18G needles prior to reattachment to the common vial.

2.2 Three Waters® C18 Sep-Pak Plus SPE Cartridges were prepared, by rinsing each one, separately, with the following: 5 mL EtOH, then 10 mL of SWFI. Cartridges were left wet after conditioning. Two of the cartridges were attached so that they were in series.

2.3 One Waters® HLB Plus LP Extraction SPE Cartridge was prepared by rinsing with 5 mL EtOH, 10 mL of SWFI, and pass 3 mL of air through the cartridge.

2.4 One Grace Alltech® Maxi-Clean IC-Chelate SPE Cartridge was prepared as follows. The cartridge was filled with EtOH, shaken so that the contents were uniformly slurried in the EtOH, to ensure complete rinsing. EtOH was then passed through (total 1 mL). The cartridge was then rinsed with 20 mL of SWFI, and left wet.

2.5 K_3PO_4 / 18-crown-6 eluent solution (which will be used to elute [^{18}F]fluoride) was prepared as follows.

2.5.1 K_3PO_4 (286 mg) was weighed to a 1-dram vial, and Aristar Ultra water (3.00 mL) was added. The vial was sealed with a PTFE-lined cap, and vortexed until contents were homogeneous with no solids.

2.5.2 To a 1-dram vial containing 400.0 mg of anhydrous 18-crown-6 was added 561 uL of the K_3PO_4 solution from step 2.5.1. Anhydrous MeCN (2.24 mL) was added, and the vial was sealed with a PTFE-lined cap, and vortex until the contents were homogeneous with no solids.

2.6 A solution of 18-crown-6 in anhydrous MeCN (which will be used as the oxidative fluorination reaction solvent) was prepared as follows. To a 1-dram vial containing 40.0 mg of anhydrous 18-crown-6 was added 4.0 mL of anhydrous MeCN. The vial was sealed quickly with a PTFE-lined cap to avoid absorption of water from the external atmosphere, and vortex until contents were homogeneous with no solids.

2.7 The Reaction Vial, containing **3a** + I(III) oxidant, was prepared as follows. *To ensure success, test that the RCC of oxidative fluorination of complex* **3a** *is at least 10% (by the procedure described in* Radiosynthesis of ^{18}F-labeled Molecules, S23), *within 2 weeks of production.*

The following steps may be performed the day before production.

A clean 1-dram vial, and a clean 10 mL V-vial were dried in a 200 ^{0}C oven for at least 6 hours. While still hot, these materials were transfered into a nitrogen-filled glovebox, and cooled to room temperature in the glovebox under argon or nitrogen. To the 1-dram vial were added 13.0

mg of **3a**, and 13.0 mg of I(III) Oxidant ([PhI(4-OMe-pyridine)$_2$][2OTf]). The two solids were gently mixed with a spatula, to make a homogeneous admixture. 20.0 mg of this admixture was added to the 10 mL V-vial, which was then sealed with a PTFE-lined septum and cap in the glovebox under nitrogen. The vial was then removed from the glovebox and transported under ambient atmosphere to the site of radiopharmaceutical production.

3. Hot cell set up

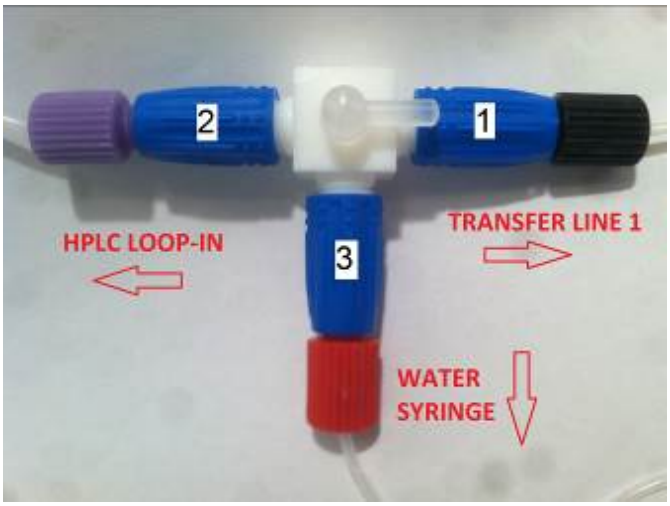
3.1 The hot cell was set up as follows. First, the Addition Line Vial was assembled as shown in Figure S24.

Figure S24. Assembly of addition line vial, cannula, and eluent tube.

The Addition Line Vial and cannula (which were assembled and cleaned as described in step 1.3) is attached to the eluent tube, with the eluent tube end near the bottom of the V vial. The end of the cannula was positioned at the bottom of the V-vial. An 18G vent needle was inserted to the septum.

3.2 The water dilution / HPLC loading valve and lines were assembled as follows (Figure S25).

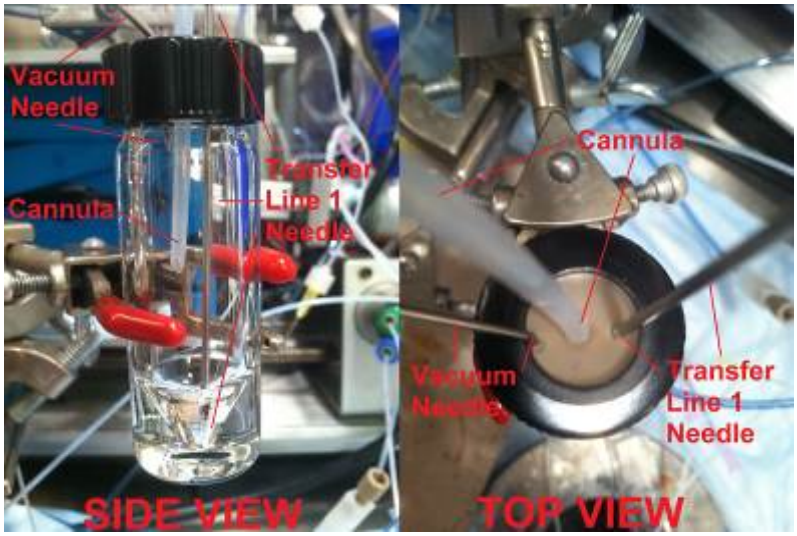
Figure S25. Assembly of water dilution and HPLC loading valve and lines.



As described in 1.4.1, **Position 1** leads to a clean 16G spinal needle (Transfer Line 1). **Position 1** was closed, so that only **Positions 2 and 3** were connected. To **Position 2**, the line leading to the semiprep HPLC loop-in was attached. To **Position 3**, a line leading to a syringe containing 4.0 mL of SWFI was attached.

3.3 Assemble the reaction vial as shown in **Figure S26.**

Figure S26. Reaction vial assembly

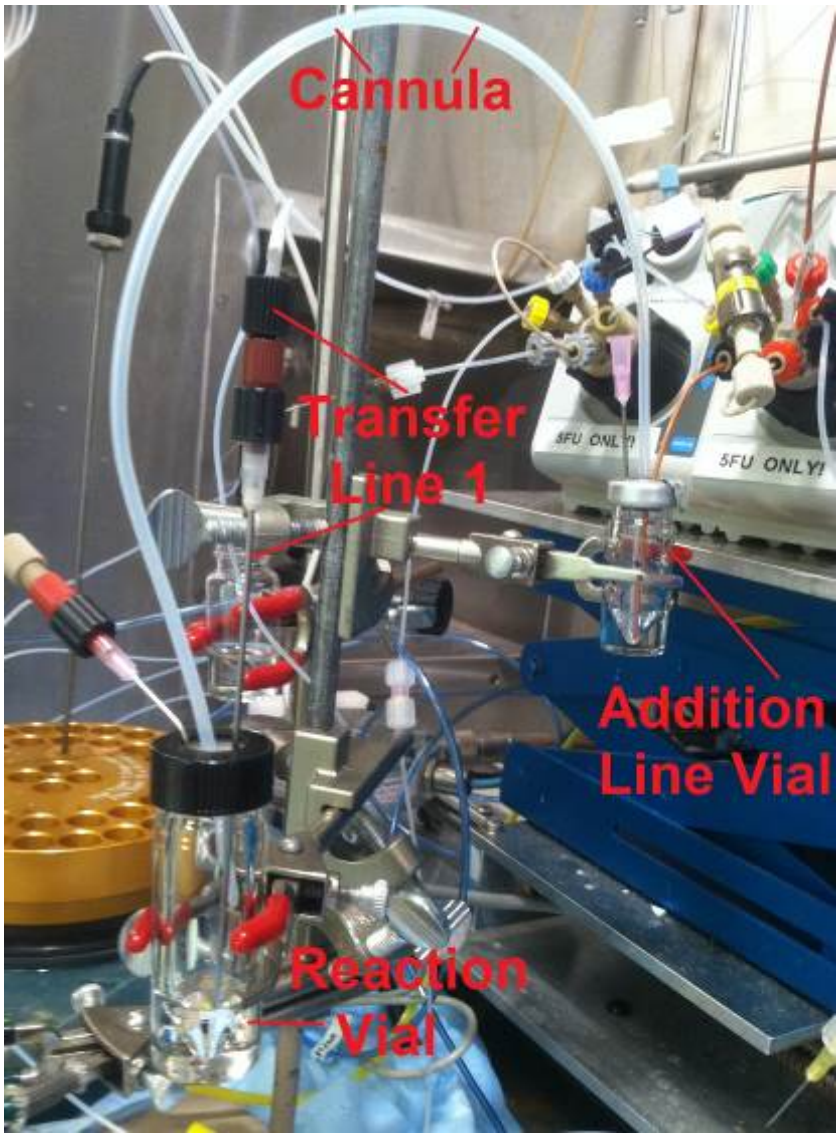


The ***center*** of the reaction vial septum was perforated with the cannula that is connected on its other side to the Addition Line Vial. The ***sides*** of the septum were perforated with the Transfer Line 1 16G spinal needle (connected to **Position 1** of the HPLC loading line valve, see Figure S25), and vacuum needle. Only the transfer line 1 needle touched the bottom of the V-vial. The Transfer Line 1 must be **closed off** from the three-way valve (**Position 1** must be closed off, see

Figure S25). ***The cannula and needles fit tightly through the septum, with no open space around where they pass through the septum -- the setup must be airtight, to prevent premature addition of MeCN to the reaction vial during fluoride elution.***

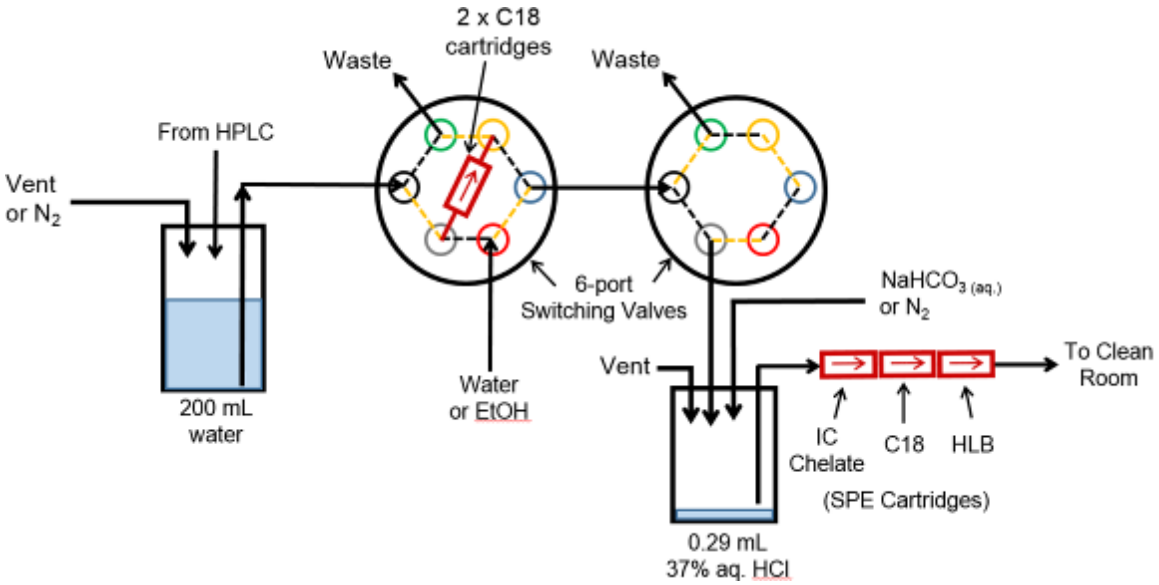
3.4 The final complete hot cell setup for synthesis is displayed in **Figure S27**.

Figure S27. Completed assembly



3.5 The purification and formulation module was prepared as shown in Figure S28.

Figure S28. Purification and formulation module



As shown in Figure S28, a bottle connected to the HPLC collection line was filled with 200 mL of SWFI. Two C18 Sep Pak Plus cartridges connected in series (prepared in step 2.2) were connected to the valve. A 30 mL sterile vial was charged with 0.29 mL of 37% HCl. Additionally, 5 mL of SWFI, 2.0 mL of EtOH, and a solution of 18.1 mL of SWFI with 4.1 mL of 1M sodium bicarbonate were prepared for application as described in steps 6.3, 7.1 and 7.2. One Maxi Clean IC-Chelate, one conditioned C18 Sep pak and one conditioned HLB cartridge were connected in series to the product delivery line leading to the clean room.

4. Preparation for Synthesis

4.1 A syringe with 1 mL of anhydrous MeCN was attached to the MeCN input line.

4.2 The common vial (with attached PEEK tubing line, nitrogen, and vent lines), was placed in the dose calibrator, in order to measure the amount of F18 activity delivered from the cyclotron.

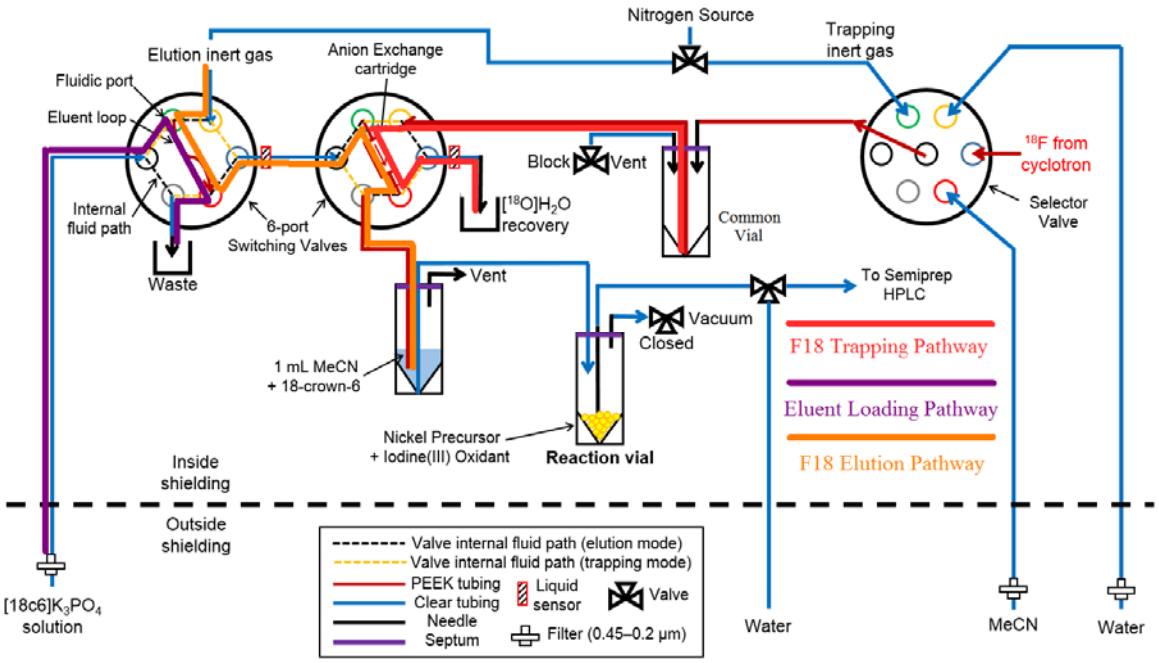
4.3 The F18 delivery line (from the cyclotron) was attached to the selector valve. The selector valve position was changed to match the position where the F18 line is connected, so that F18 could later be delivered from the cyclotron, to the common vial.

4.4 The anhydrous MeCN + 18-crown-6 solution (1.00 mL, prepared in step 2.6) was added to the Addition Line Vial (within 5 minutes before transfer of F18 from cyclotron, to prevent excessive wetting by exposure to air through the vent needle in the Addition Line Vial).

5. Synthesis Execution.

5.1 The [^{18}F]fluoride was delivered from the cyclotron to the common vial. The amount of activity delivered to the common vial was monitored by reading the dose calibrator.

Figure S29. Pathways for trapping, eluent loop filling, and elution.



5.2 The [^{18}F]fluoride was trapped as follows. After the delivery of [^{18}F]fluoride in step 5.1 was complete, the selector valve was switched to the position for nitrogen gas. The [^{18}F]fluoride was pushed with 15 psi of nitrogen through the ion exchange cartridge (Figure S29, red line for F18 Trapping Pathway). To the common vial was added MeCN (1.0 mL) through the selector valve, and this MeCN was pushed through the cartridge as above, followed with nitrogen to dry the lines and cartridge.

5.3 The [^{18}F]fluoride was eluted as follows. The eluent line was primed with K_3PO_4 / 18-crown-6 eluent (prepared in step 2.5.2). The eluent loop was then filled (Figure S29, purple line for Eluent Loading Pathway), the [^{18}F]fluoride was eluted by pushing (with nitrogen pressure) the eluent, from the eluent loop, through the cartridge, and out of the eluent spout into the Addition Line Vial (Figure S29, orange line for F18 Elution Pathway). This step was repeated two times, for a total of 3 elutions. At the end of the third elution, nitrogen was bubbled through the eluent spout at 15 psi, to thoroughly mix the eluted [^{18}F]fluoride with the MeCN in the Addition Line Vial.

5.4 When the elution finished, vacuum was applied to the needle connected to the Reaction Vial. Important: ***When properly configured, the [^{18}F]fluoride solution is transferred from the Addition Line Vial, through the cannula, to the Reaction vial, instantaneously (< 1 second).*** The vacuum was turned off once the addition was complete. A yellow homogeneous solution was observed in the Reaction Vial. After 1 minute, step 6.1 was initiated.

6. Purification

6.1 The reaction mixture was diluted with water as follows. The three-way water dilution / HPLC transfer line valve (Figure S25) was adjusted to connect the water addition line (**Position 3**) to the

Transfer Line 1 (**Position 1**). SWFI (4.0 mL) was added to the reaction vial, followed by at least 20 mL of air, which was bubbled gently to mix the reaction mixture. A yellow mixture formed that should be mixed, with air bubbling, to homogeneity, to resemble freshly mixed orange juice.

6.2 Promptly, without letting the precipitate settle, the three-way valve was adjusted to connect the Transfer Line 1 (**Position 1**) with the HPLC loop-in line (**Position 2**), the valve was placed behind shielding, and the crude product was loaded into the loop-in line, and injected onto the semiprep HPLC (column: Phenomenex Luna C18(2) (5 μm, 10.00 x 250 mm)). Elution occurred with 60:40 MeCN/(10 mM ammonium formate in water) v/v, flow rate = 5 mL/minute, for 13 minutes, then 70:30 MeCN/(10 mM ammonium formate in water) v/v, flow rate = 5 mL/minute. The product **[^{18}F]4a** eluted at approximately 18 minutes.

6.3 The **[^{18}F]4a** was collected from the HPLC collection line into 200 mL of SWFI. After completion of collection, the diluted **[^{18}F]4a** was passed (with nitrogen push) through two C18 Sep Pak cartridges (conditioned in step 2.2, and assembled in step 3.5, see Figure S28), in series, to trap the **[^{18}F]4a** and remove MeCN and salt from the HPLC mobile phase. The cartridges were rinsed with SWFI (5 mL). EtOH (2.0 mL) was then passed through the cartridges, in order to elute **[^{18}F]4a** into a 30 mL vial that contained 0.29 mL of 37% aq. HCl. The liquids were mixed as thoroughly as possible with a gentle nitrogen stream, and were allowed to sit for two minutes at 23 ºC. Deprotection of **[^{18}F]4a** occured during this time to form [^{18}F]5-FU.

7. Formulation

7.1 A solution prepared from 18.1 mL of SWFI and 4.1 mL of 1M sodium bicarbonate was added to the [^{18}F]5-FU solution.

7.2 The resulting solution was passed (with nitrogen push, after removal of the delivery and vent lines) through the IC-Chelate, C18, and HLB cartridges (conditioned in steps 2.2, 2.3, 2.4), in series, into a line that lead to an intermediate sterile vial that was located in a clean room for sterile radiopharmaceutical packaging.

7.3 In the clean room, the collected [^{18}F]5-FU solution was passed from the intermediate vial, through a sterilizing Millex GP 0.22 micron filter, into a sterile 30 mL vial to afford pure, formulated, sterile [^{18}F]5-FU.

8. Quality control

8.1 Radiochemical Identity, Chemical & Radiochemical Purity, and Specific Activity of [^{18}F]5-FU were measured by HPLC. The column was a Phenomenex Luna® 5 μm PFP(2) 100 Å, 250 x 4.6 mm analytical column. The mobile phase was 65 mM ammonium formate in water, flow rate = 1 mL/minute.

8.2 The formulated [^{18}F]5-FU was visually inspected.

8.3 The pH was measured by coating pH indicator paper with several drops of the solution.

8.4 The formulated [^{18}F]5-FU was tested for bacterial endotoxins.

8.5 Radionuclidic Half-Life was measured.

8.6 The formulated [^{18}F]5-FU was analyzed for sterility.

8.7 The Sterilizing Filter Integrity Test was performed.

8.8 Radiochemical identity and purity were analyzed by Radio TLC.

8.9 Residual solvents were assayed by gas chromatography.

8.10 Nickel content was measured by ICP-MS.

Table S3. Results of [^{18}F]5-fluorouracil synthesis

	Synthesis 1	Synthesis 2	Synthesis 3
Isolated yield (mCi) of [^{18}F]5-FU at EOS [a]	13.48	18.83	12.85
Starting [^{18}F]fluoride activity (mCi) [b]	1785	1698	1433
Percent Yield	0.7552%	1.109%	0.8967%
Average % Yield ± Std. Dev.	0.92% ± 0.18%		
Area of 5FU Peak (UV, 266 nm)	1.82437	0.927105	0.510297
Specific Activity [c] (SA) at EOS (Ci / μmol)	14.48	38.95	49.34
Average SA ± Std. Dev. at EOS (Ci / μmol)	34.3 ± 17.9		
Synthesis Time (minutes)	120 (85[d])	84	90
Solution Volume (mL)	23.2	23.7	23.2
pH	7.5	7.5	7.5
Ni content (ppb)	45.7	94.2	40.3
Unknown Impurities (μg) [e]	3.91	5.91	2.29

[a] EOS refers to the end of synthesis, when the completely purified, formulated product in a sterile vial in a clean room, obtained after Step 7.3 of the synthesis, was measured for radioactivity.

[b] Radioactivity of [^{18}F]fluoride solution (in about 2.4 mL [^{18}O]water) delivered from cyclotron to the common vial (Step 5.1 of synthesis), immediately after the end of bombardment.

[c] Amount of 5FU, for the SA measurement, was calculated as follows: nmol 5FU = (5FU peak area) * (calibration curve slope) * (Solution Volume) / (Injection Volume). Injection volume = 0.100 mL; See Figure S30 for Calibration Curve.

[d] Corrected synthesis time for Synthesis 1, after subtracting extra time elapsed due to unexpected clogging, which did not occur in subsequent runs, after implementation of preventative measures.

[e] Estimated from UV areas (266 nm), as described above[c], and using the molecular weight of 5-FU.

Table S4. Data for the standard curve of UV absorbance vs. amount of 5-FU

Amount of **5-FU** (nmol)	UV absorbance (266 nm)
0.192189	72.7
0.192189	73.6
0.192189	75.3
0.384379	153.6
0.384379	156.1
0.384379	158.5
0.768758	321.8
0.768758	325.7
0.768758	328.8
1.153137	503.2
1.153137	499.8
1.153137	506.8
1.921894	855.9
1.921894	855.9
1.921894	862.5

Figure S30. Calibration curve for nmol of 5-FU vs UV absorbance at 266 nm

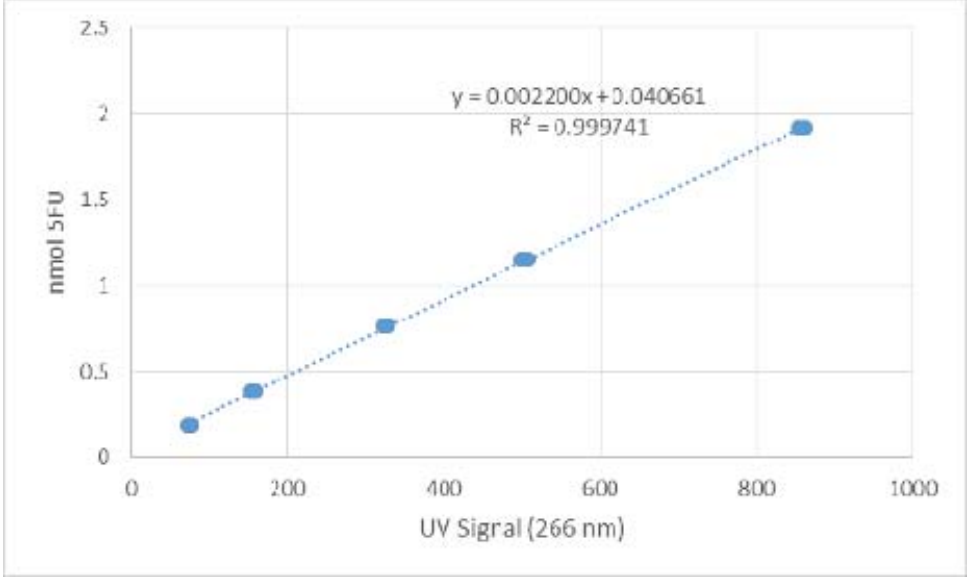


Table S5. Summary of quality control for produced [^{18}F]5-Fluorouracil (for doses produced from 3 validation syntheses)

Analytical Test Method	*Specifications*	*Result (PASS or FAIL)*
Visual Inspection	Clear and colorless solution, free from visible particles	**PASS**
Radionuclidic Purity of [^{18}F] by Gamma Spectroscopy	The final product centroid value is within 1% of the ^{68}Ge calibration standard.	**PASS**
Radionuclidic Identity of [^{18}F] by Half-Life	Half-Life of 105 – 115 minutes	**PASS**
Radiochemical Purity [^{18}F] 5FU (TLC)	≥ 90% as [^{18}F] 5FU	**PASS**
Radiochemical Purity [^{18}F] 5FU (HPLC)	≥ 90% as [^{18}F] 5FU	**PASS**
Radiochemical Identity of [^{18}F] 5FU (HPLC)	Retention time of Standard and the Final product is ±5% of each other	**PASS**
pH	4.5-8.5	**PASS**
Filter Integrity	Meets Manufacture's Bubble Point Specification	**PASS**
Chemical Purity (HPLC)	≤10µg of Total Impurity	**PASS**
Residual Solvents (GC)	Ethanol ≤ 10 % Acetonitrile ≤ 0.04 % (observed peak ≤ standard peak) IPA (observed peak ≤ standard peak)	**PASS**
Bacterial Endotoxin	<6 EU /mL (≤175EU/vial)	**PASS**
POSTRELEASE		
Sterility	No turbidity or growth (Sterile)	**PASS**
Nickel Content	≤ 500ppb	**PASS**
Radionuclidic Purity of [^{18}F] by Gamma Spectroscopy	Main photopeak at 0.551± 0.02 MeV; ≥ 99.5% ^{18}F	**PASS**

Measurement of trapped and eluted [^{18}F]fluoride, and RCC for [^{18}F]4a formation, with the new concentrator instrument

The synthesis was performed with the new instrument (steps 1.1 – 5.4), with modifications A and E (below) and additional steps B – D to obtain the desired data:

A. In order to perform multiple experiments with one cyclotron bombardment, instead of using the entire 2.4 mL of [^{18}F]fluoride in [^{18}O]water, an aliquot (between 0.2 mL and 1.0 mL of the 2.4

mL [^{18}F]fluoride solution) was injected directly into the common vial by syringe, followed by a sufficient amount of Aristar Ultra water to bring the total volume in the common vial to 2.4 mL.

B. In order to measure trapped [^{18}F]fluoride, immediately after trapping [^{18}F]fluoride on the Opti-Lynx cartridge (step 5.2), the cartridge was removed from its holder, its radioactivity was measured, and then it was placed back into its holder.

C. In order to measure eluted [^{18}F]fluoride, after synthesis completion (step 5.4), the radioactivity of the Addition Line Vial, cannula, Reaction Vial, and Transfer Line 1 were measured and summed (since these are all of the components that came into contact with [^{18}F]fluoride after elution was completed, as defined by leaving the elution spout).

D. In order to measure RCC for conversion of [^{18}F]fluoride to **[^{18}F]4a**, after completion of step 5.4, the yellow solution in the Reaction Vial was assayed as described in Measurement of Radiochemical Yield by radio TLC (in this case, the Reaction Vial was rinsed with MeCN (2 times 1.0 mL, introduced via Transfer Line 1) to remove residual reaction solution from the walls of the vial). In all cases, radioactivity in solution was >90%.

E. In the case of Run #4 (Table SX), in order to test the effect of PPTS buffer on RCC, PPTS was added to the reaction solvent (prepared as follows instead of as described in step 2.6): 37.6 mg of 18-crown-6 was dissolved in 3.76 mL dry MeCN. A 1.00 M PPTS solution in dry MeCN was prepared, and 22.56 μL of this solution was added to the 18-crown-6 solution to give a homogeneous colorless solution.

Table S6. Data for trapping, elution, and RCC in synthesis with the concentrator instrument.

	Run #				
	1	2	3	4	
Cyclotron Target Water / Aristar Water (mL / mL)	0.2 / 2.2	0.4 / 2.0	0.8 / 1.6	1.0 / 1.4	
Starting [^{18}F]fluoride activity (mCi) [b]	14.13	11.76	9.91	6.81	
Trapped [^{18}F]fluoride activity (mCi), dc	13.53	11.59	9.69	6.44	
Eluted [^{18}F]fluoride activity (mCi), dc	10.81	10.99	9.14	6.17	
Elapsed Time for steps 5.2 – 5.4 (min)	15	11	13	12	
Eluted [^{18}F]fluoride activity (mCi), ndc	9.84	10.25	8.42	5.72	
					Avg. ± Std. Dev.
Trapping Efficiency, %	95.75	98.55	97.78	94.57	96.7 ± 1.8

Elution Efficiency, %	79.90	94.82	94.32	95.81	91.2 ± 7.6
Overall Yield, % (ndc)	69.64	87.16	84.96	83.99	81.4 ± 8.0
RCC, %	3.41	2.90	2.34		2.89 ± 0.54
RCC, % (with PPTS)				2.96	

On large scale, the trapping and elution measurements were performed as follows. To the common vial was delivered [^{18}F]fluoride (1708 mCi; the timepoint of this measurement was defined as t_0. Hereafter, t_0 denotes decay correction to time = t_0). After trapping on the cartridge, 32.06 mCi (t_0) remained in the common vial. The trapping waste ([^{18}O]water recovery) contained 0.193 mCi (t_0). Therefore, 1708 mCi – 32.06 mCi – 0.193 mCi = 1675.75 mCi (t_0) of [^{18}F]fluoride was trapped on the cartridge. After elution, 94.82 mCi (t_0) remained on the cartridge. Therefore, 1675.75 mCi – 94.82 mCi = 1580.93 mCi was eluted (t_0). The total time for trapping and elution was 25 minutes. Applying 25 minutes of decay (half life = 109.771 minutes) to 1580.93 mCi, the non-decay-corrected amount of eluted [^{18}F]fluoride was 1350.06 mCi.

Trapping efficiency = 1675.75 mCi / 1708 mCi * 100% = 98.1%

Elution efficiency = 1580.93 / 1675.75 * 100% = 94.3%

Overall not-decay-corrected yield = 1350.06 mCi / 1708 mCi * 100% = 79.0%

Measurement of elution volume

The eluent loop was filled with water, and an elution was performed by passing the contents of the eluent loop through the Opti-Lynx ion exchange cartridge, out of the elution spout, into a pre-weighed vial. This process was repeated (two elutions were performed in total). For two elutions, the measured volume was 12.4 ± 0.4 µL (average of four measurements).

Determination of K_3PO_4 and 18-crown-6 concentration in eluent

As described in section 2.5 of the cGMP [^{18}F]5-FU synthesis, an aqueous K_3PO_4 solution was prepared from 286 mg of K_3PO_4 and 3.00 mL of water. A 1.00 mL portion of the aqueous K_3PO_4 solution weighed 1.077 g. To prepare the eluent solution, a portion of the aqueous K_3PO_4 solution (561 µL) was added to a vial containing 400.0 mg of 18-crown-6, dry MeCN (2.24 mL) was added, and the resulting mixture was sonicated until a homogeneous solution was obtained. A 1.00 mL portion of the eluent solution weighed 0.872 g. Based on these measured densities and component mass ratios, 1.00 mL of the eluent solution contains 78.1 µmol of K_3PO_4 and 477 µmol of 18-crown-6.

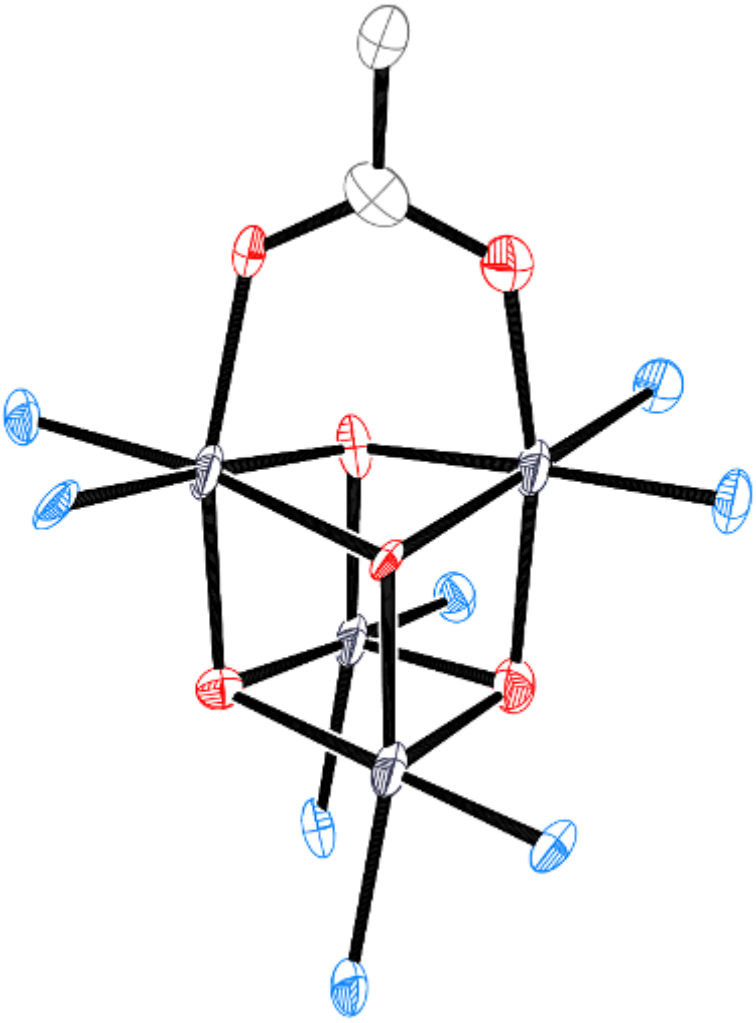
X-ray Crystallographic Analysis

Experimental

A crystal was mounted on a diffractometer, and data was collected at 100 K. The intensities of the reflections were collected by means of a Bruker APEX II DUO CCD diffractometer ($Mo_{K\alpha}$

radiation, λ=0.71073 Å), and equipped with an Oxford Cryosystems nitrogen flow apparatus. The collection method involved 0.5° scans in ω at 28° in 2θ. Data integration down to 0.84 Å resolution was carried out using SAINT V7.46 A (Bruker diffractometer, 2009) with reflection spot size optimization. Absorption corrections were made with the program SADABS (Bruker diffractometer, 2009). The structure was solved by the direct methods procedure and refined by least-squares methods again F^2 using SHELXS-97 and SHELXL-97 (Sheldrick, 2008) with OLEX 2 interface (Dolomanov, et al., 2009). Non-hydrogen atoms were refined anisotropically, and hydrogen atoms were allowed to ride on the respective atoms. Crystal data as well as details of data collection and refinement are summarized in Tables S3-S5.

Figure S31. The structure of 1. The atoms are depicted with 50% probability ellipsoids.



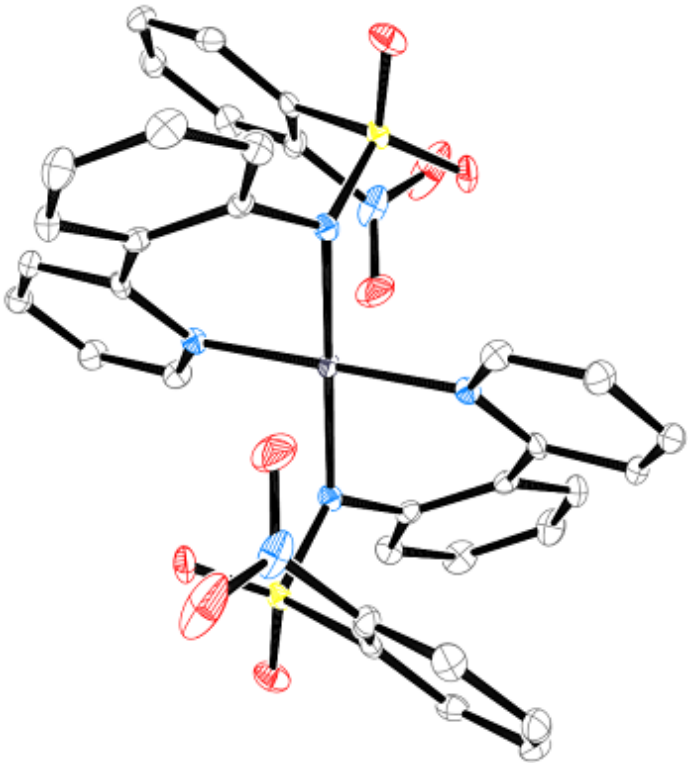
The solvent, potassium, hydrogen, and pyridylsulfonamide ligand atoms (except nitrogens bound to nickel) present in the crystal are omitted for clarity in Figure S31.

Table S7. Experimental details

1	**CCDC 1413389**
Crystal data	
Chemical formula	$C_{84}H_{77.50}KN_{12}Ni_4O_{25.50}S_4$
M_r	2065.26
Crystal system, space group	Triclinic, *P*-1
Temperature (K)	100
a, *b*, *c* (Å)	16.705 (3), 16.794 (3), 18.748 (4)
α, β, γ (°)	86.637 (4), 66.457 (4), 68.899 (4)
V (Å^3)	4476.0 (15)
Z	2
Radiation type	Mo *K*α
μ (mm^{-1})	1.05
Crystal size (mm)	0.28 × 0.12 × 0.10
Data collection	
Diffractometer	Bruker D8 goniometer with CCD area detector diffractometer
Absorption correction	Multi-scan TWINABS
T_{min}, T_{max}	0.757, 0.902
No. of measured, independent and observed [$I > 2\sigma(I)$] reflections	15845, 15845, 6266
R_{int}	0.0000
$(\sin \theta/\lambda)_{max}$ (Å^{-1})	0.601
Refinement	
$R[F^2 > 2\sigma(F^2)]$, $wR(F^2)$, S	0.074, 0.181, 0.85
No. of reflections	15845
No. of parameters	1232
No. of restraints	237
H-atom treatment	H-atom parameters constrained
$\Delta\rho_{max}$, $\Delta\rho_{min}$ (e Å^{-3})	1.24, -1.23

Computer programs: *APEX2* v2014.3.0 (Bruker-AXS, 2014), *SAINT* 8.30C (Bruker-AXS, 2014), *SHELXS97* (Sheldrick, 2008), *SHELXL97* (Sheldrick, 2008), Bruker *SHELXTL* (Sheldrick, 2008).

Figure S32. The structure of 2. The atoms are depicted with 50% probability ellipsoids.



The solvent and hydrogen atoms present in the crystal of **2** are omitted for clarity in Figure S32.

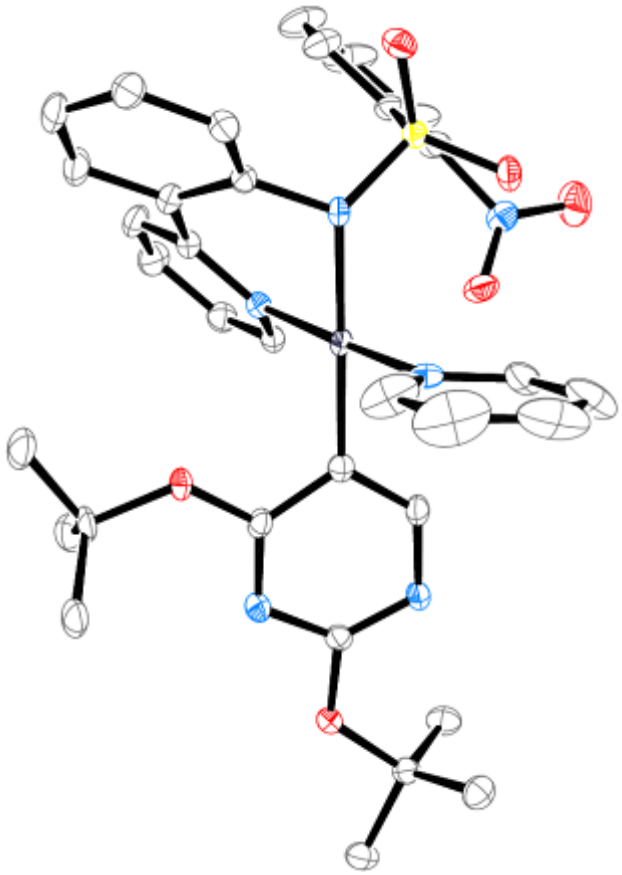
Table S8. Experimental details

2	**CCDC 1413390**
Crystal data	
Chemical formula	$C_{35}H_{26}Cl_2N_8NiO_8S_2$
M_r	880.37
Crystal system, space group	Triclinic, $P\bar{1}$
Temperature (K)	100
a, *b*, *c* (Å)	9.134 (2), 9.293 (2), 12.052 (3)
α, β, γ (°)	103.787 (5), 98.329 (5), 112.385 (5)
V (Å^3)	886.5 (4)
Z	1
Radiation type	Mo *K*α
μ (mm^{-1})	0.88

Crystal size (mm)	0.40 × 0.30 × 0.10
Data collection	
Diffractometer	CCD area detector diffractometer
Absorption correction	Multi-scan *SADABS* (Sheldrick, 2009)
T_{min}, T_{max}	0.719, 0.917
No. of measured, independent and observed [$I > 2\sigma(I)$] reflections	10289, 3355, 2686
R_{int}	0.049
$(\sin \theta/\lambda)_{max}$ (Å^{-1})	0.610
Refinement	
$R[F^2 > 2\sigma(F^2)]$, $wR(F^2)$, S	0.045, 0.139, 1.08
No. of reflections	3355
No. of parameters	259
No. of restraints	1
H-atom treatment	H-atom parameters constrained
$\Delta\rangle_{max}$, $\Delta\rangle_{min}$ (e Å^{-3})	0.69, -0.57

Computer programs: *APEX2* v2009.3.0 (Bruker-AXS, 2009), *SAINT* 7.46A (Bruker-AXS, 2009), *SHELXS97* (Sheldrick, 2008), *SHELXL97* (Sheldrick, 2008), Bruker *SHELXTL*.

Figure S33. The structure of 3a. The atoms are depicted with 50% probability ellipsoids.



The solvent and hydrogen atoms present in the crystal of **3a** are omitted for clarity in Figure S33.

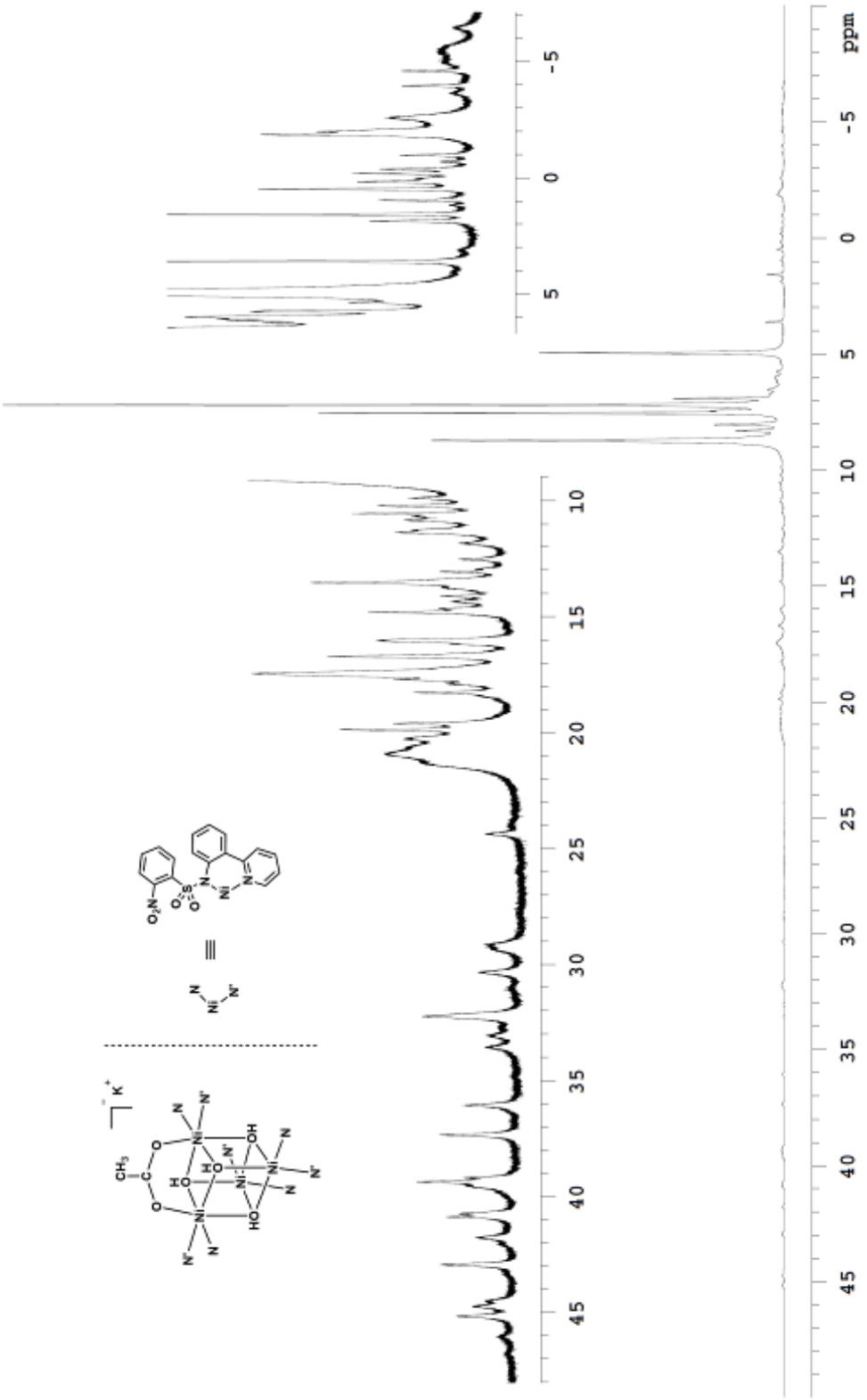
Table S9. Experimental details

3a	**CCDC 1413391**
Crystal data	
Chemical formula	$C_{39}H_{41}N_7NiO_6S$
M_r	794.56
Crystal system, space group	Monoclinic, *C*2/*c*
Temperature (K)	100
a, *b*, *c* (Å)	37.9478 (15), 9.6576 (4), 20.4981 (8)
β (°)	95.0533 (13)
V (Å^3)	7483.0 (5)
Z	8
Radiation type	Mo *K*α

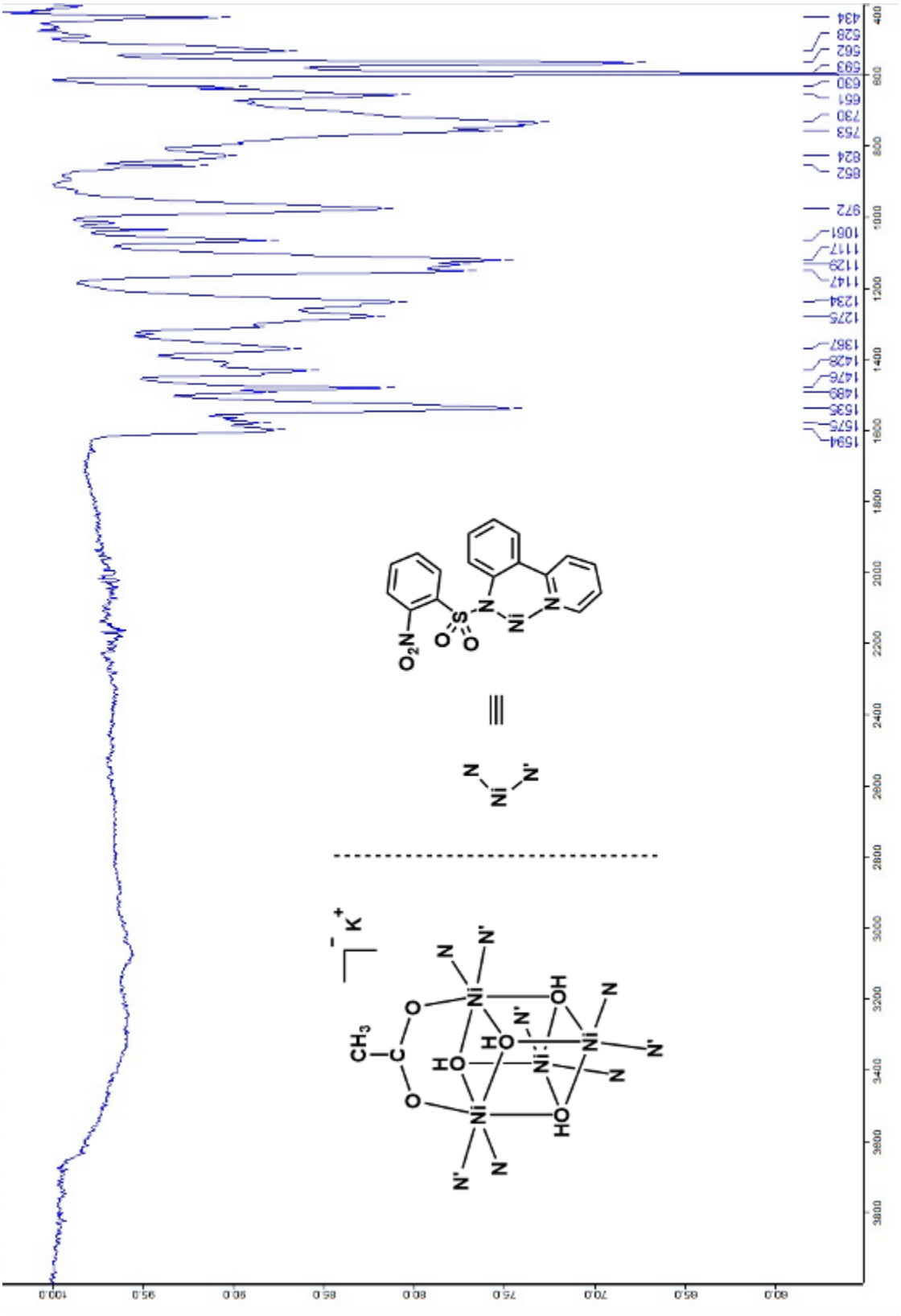
μ (mm^{-1})	0.63
Crystal size (mm)	0.16 × 0.12 × 0.03
Data collection	
Diffractometer	Bruker D8 goniometer with CCD area detector diffractometer
Absorption correction	Multi-scan *SADABS*
T_{min}, T_{max}	0.683, 0.746
No. of measured, independent and observed [$I > 2\sigma(I)$] reflections	56017, 8271, 6074
R_{int}	0.073
$(\sin \theta/\lambda)_{max}$ ($Å^{-1}$)	0.642
Refinement	
$R[F^2 > 2\sigma(F^2)]$, $wR(F^2)$, S	0.055, 0.119, 1.03
No. of reflections	8271
No. of parameters	507
No. of restraints	73
H-atom treatment	H-atom parameters constrained
$\Delta\rho_{max}$, $\Delta\rho_{min}$ (e $Å^{-3}$)	1.00, -0.68

Computer programs: *APEX2* v2014.3.0 (Bruker-AXS, 2014), *SAINT* 8.34C (Bruker-AXS, 2014), SHELXT-2014 (Sheldrick, 2015), *SHELXL2014* (Sheldrick, 2015), Bruker *SHELXTL* (Sheldrick, 2015).

Spectroscopic Data

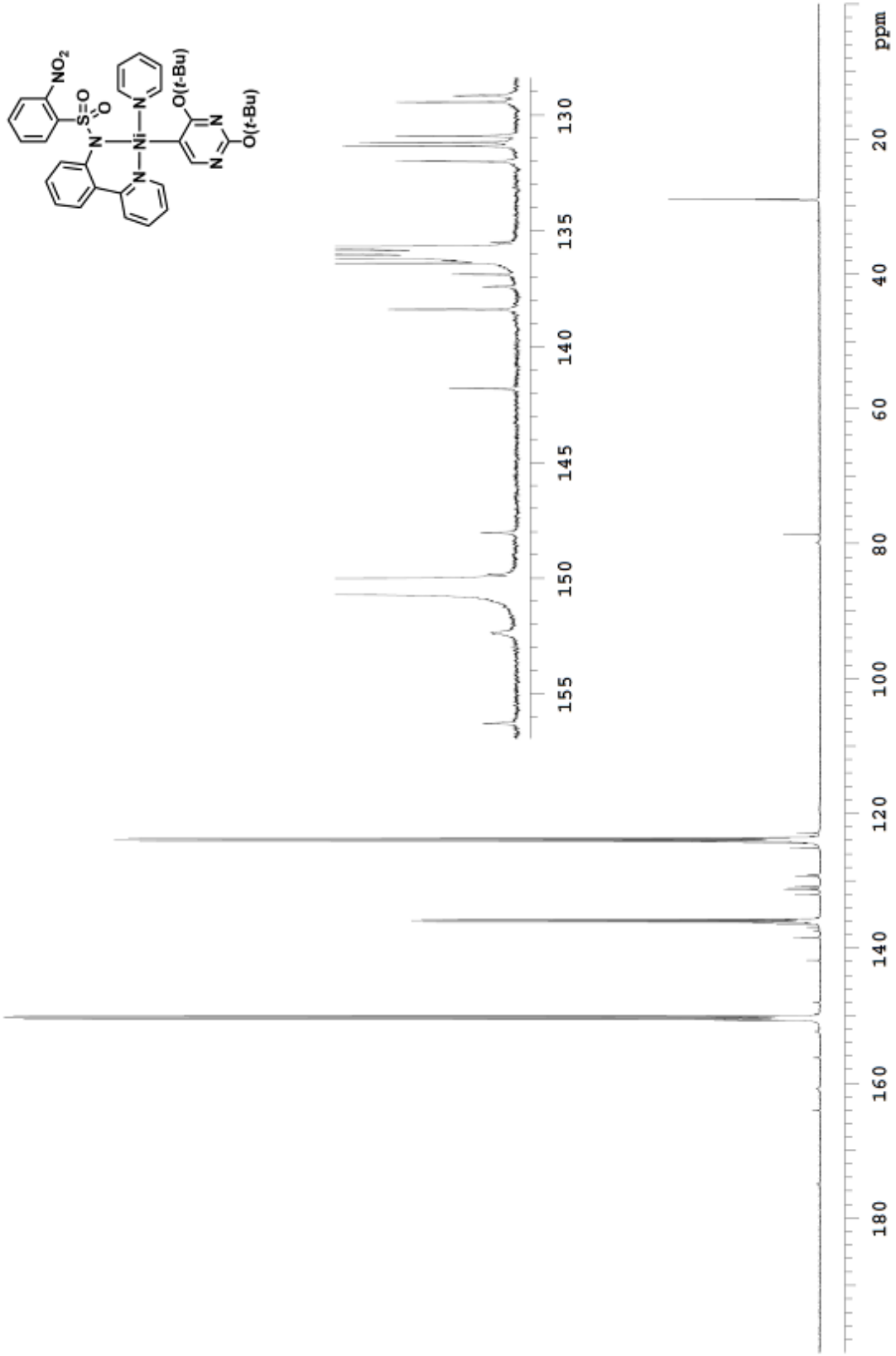


^{1}H NMR of **1**, pyridine-d_5, 600 MHz, 23 ºC

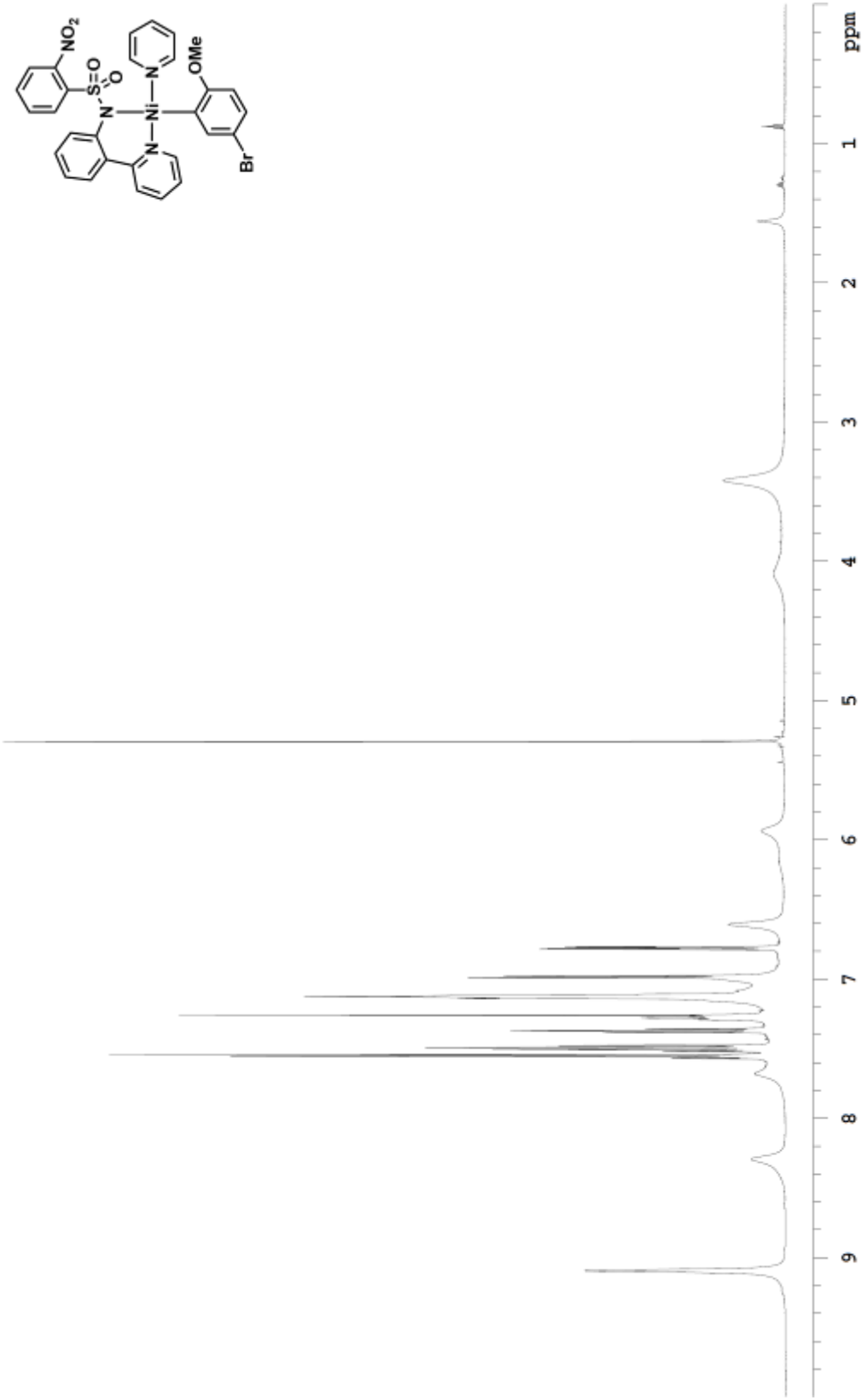


IR spectrum of **1**, neat, 23 ºC

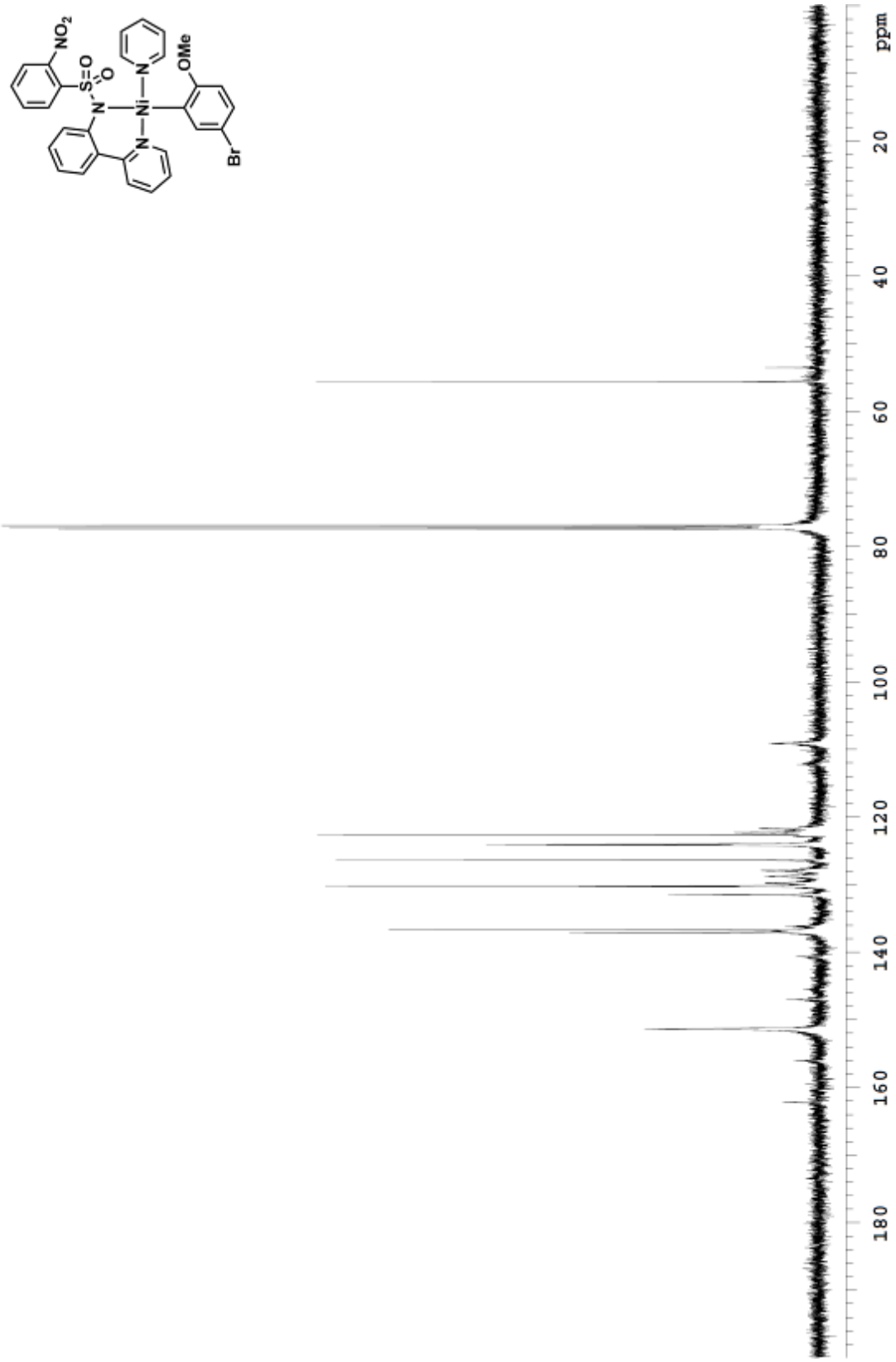
^{1}H NMR of **3a**, pyridine-d_5, 600 MHz, 23 ºC



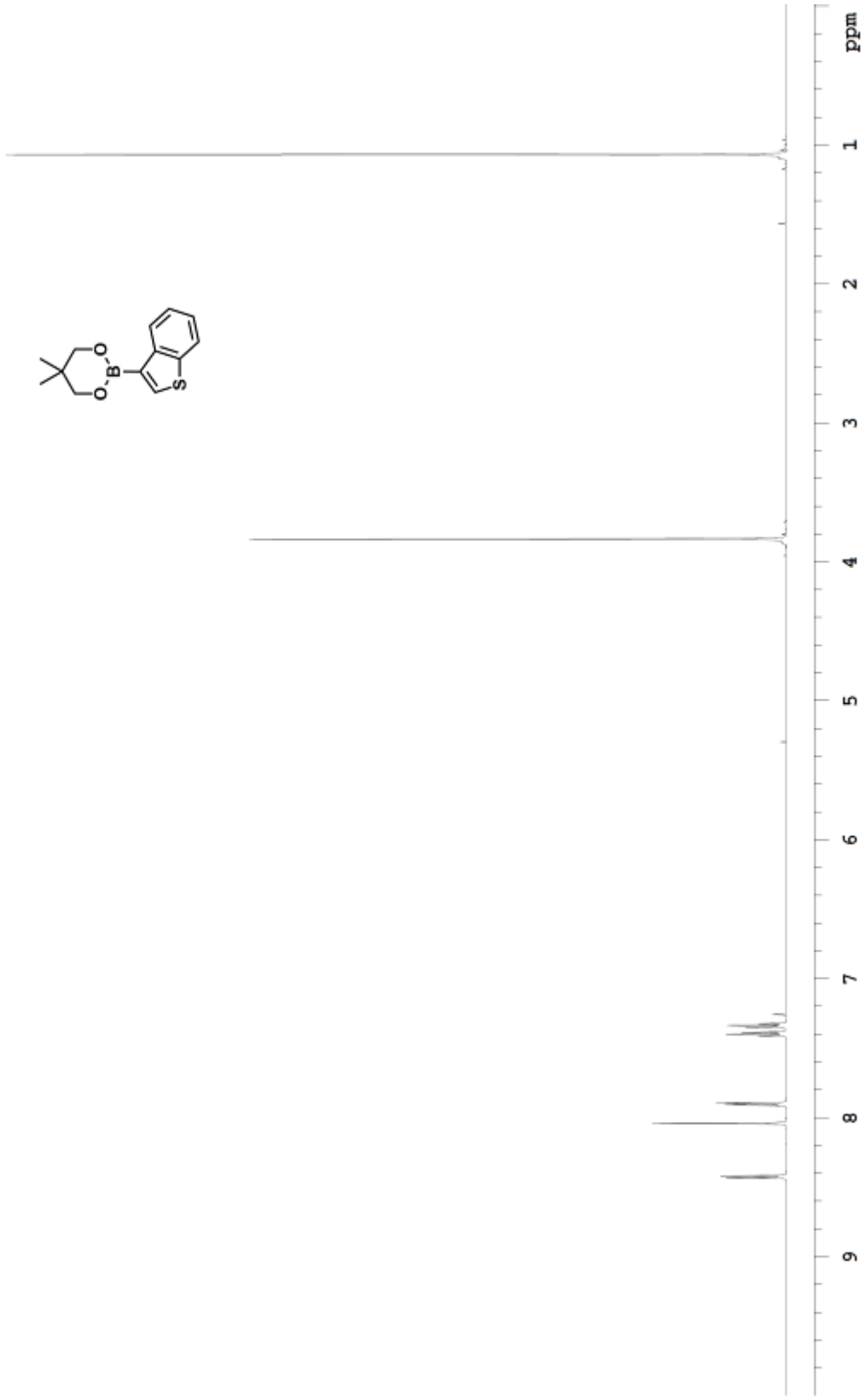
^{13}C NMR of **3a**, pyridine-d_5, 125 MHz, 23 ºC



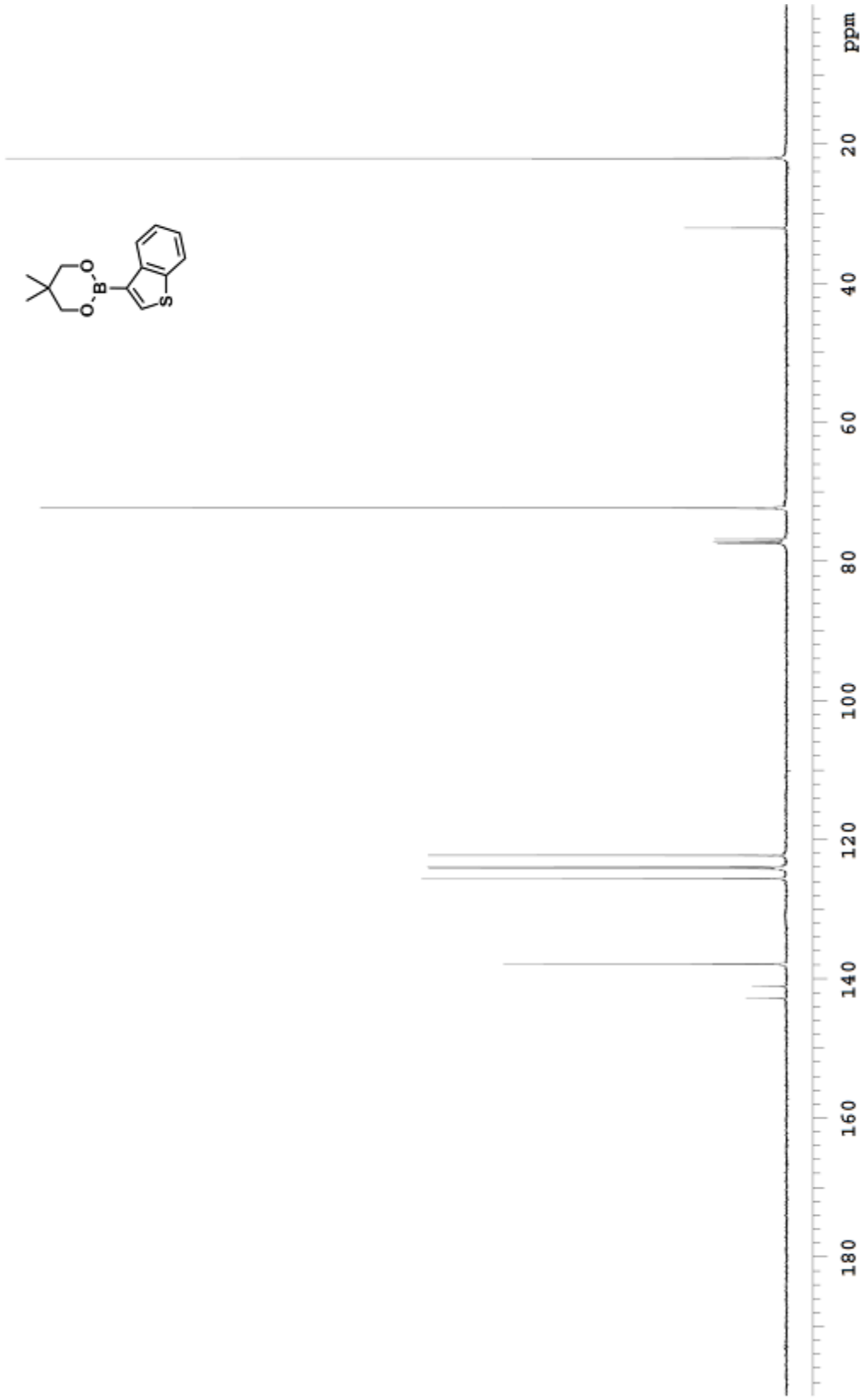
^{1}H NMR of **3b**, $CDCl_3$, 600 MHz, 23 ºC



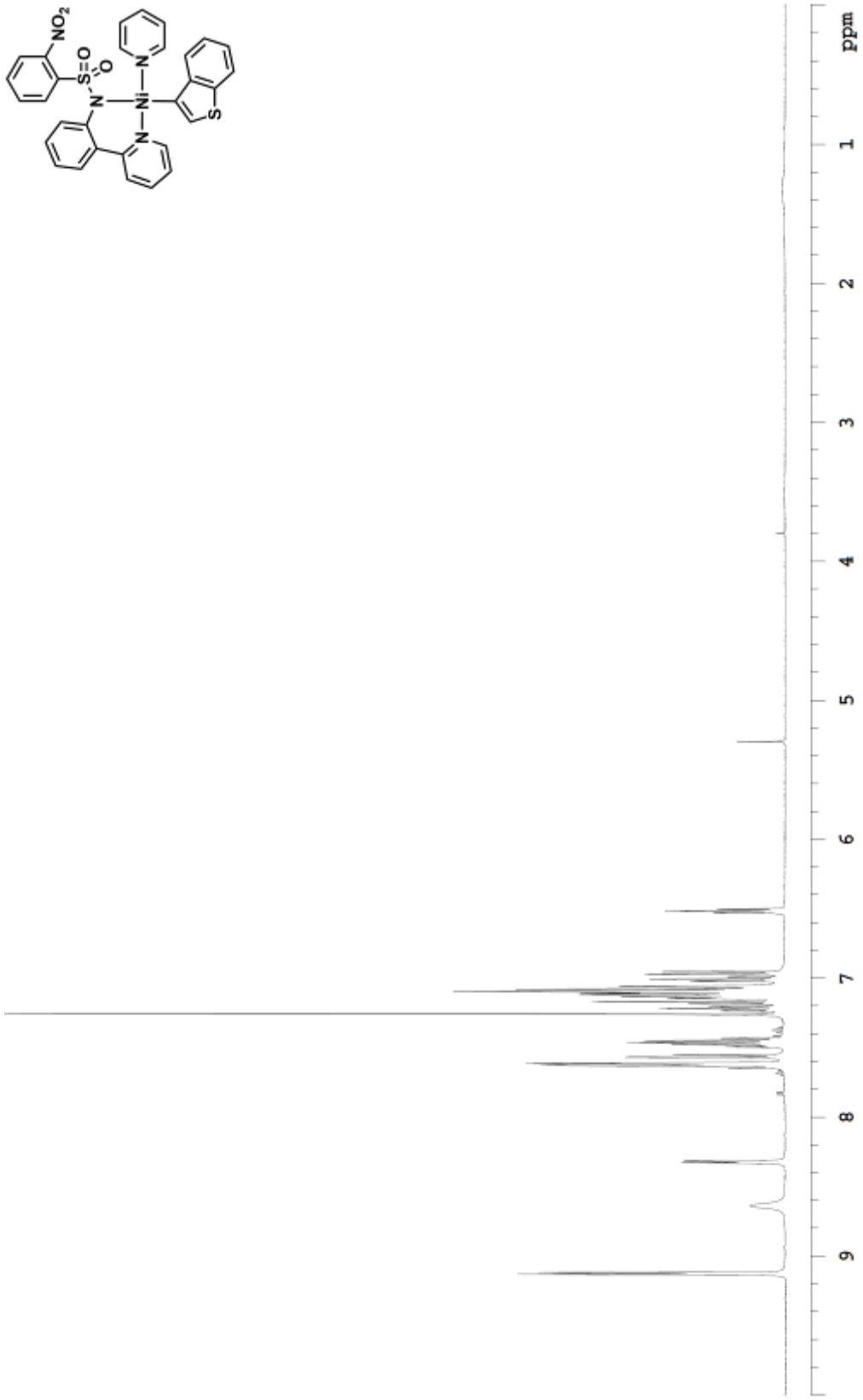
^{13}C NMR of **3b**, $CDCl_3$, 125 MHz, 23 ºC



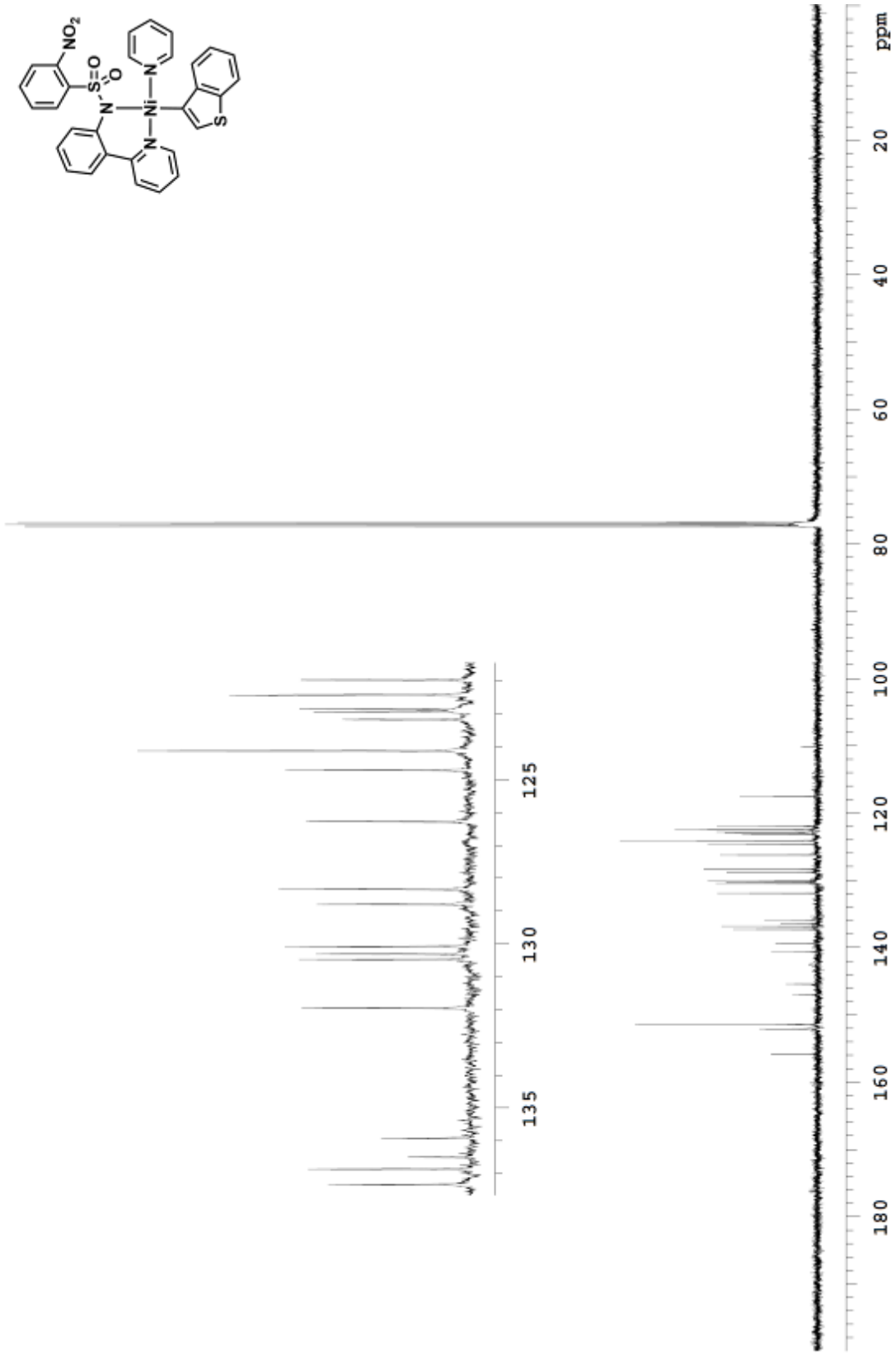
^{1}H NMR of **S1**, $CDCl_3$, 600 MHz, 23 ºC



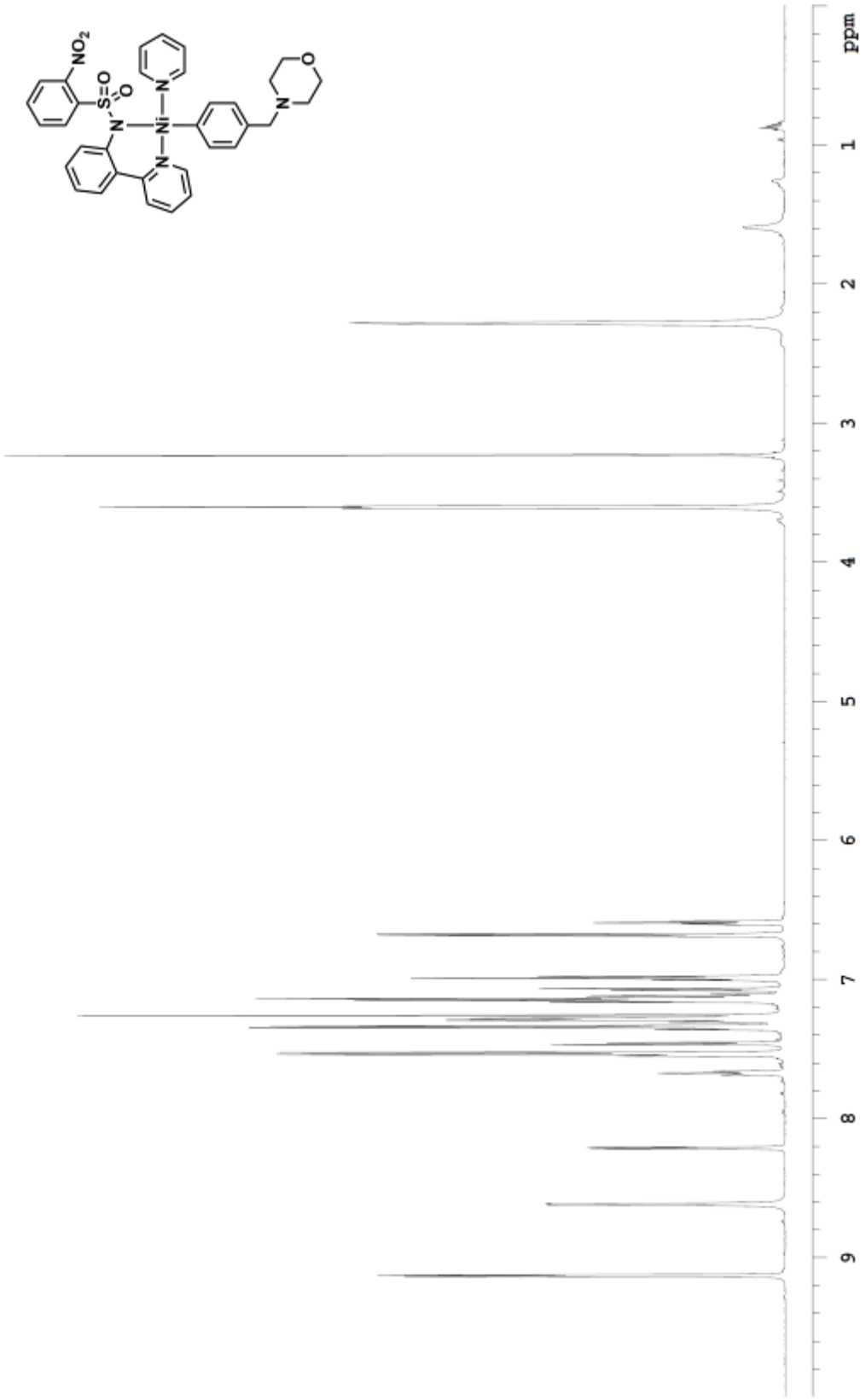
^{13}C NMR of **S1**, $CDCl_3$, 100 MHz, 23 ºC



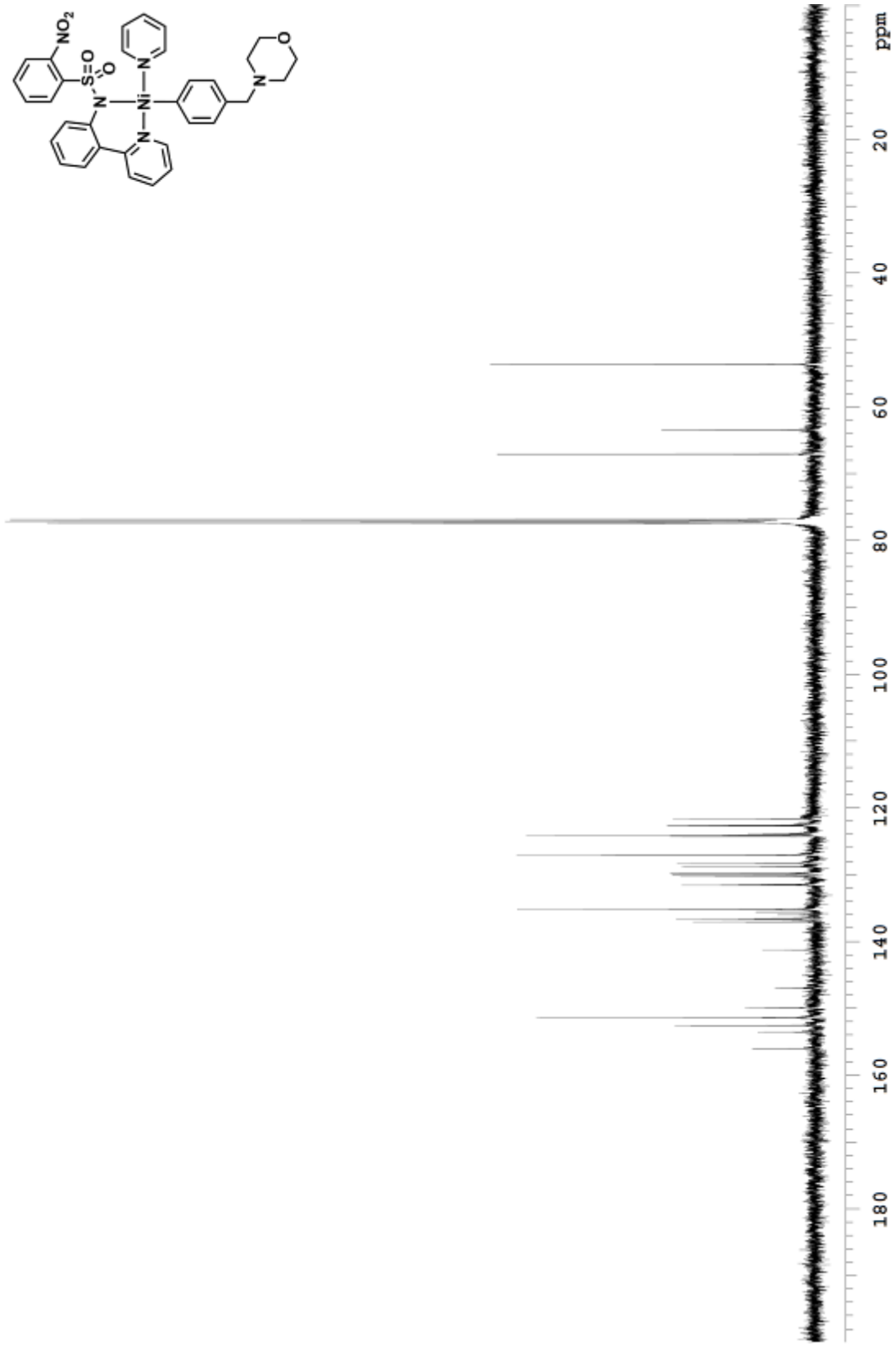
^{1}H NMR of **3c**, pyridine-d_5, 500 MHz, 23 ºC



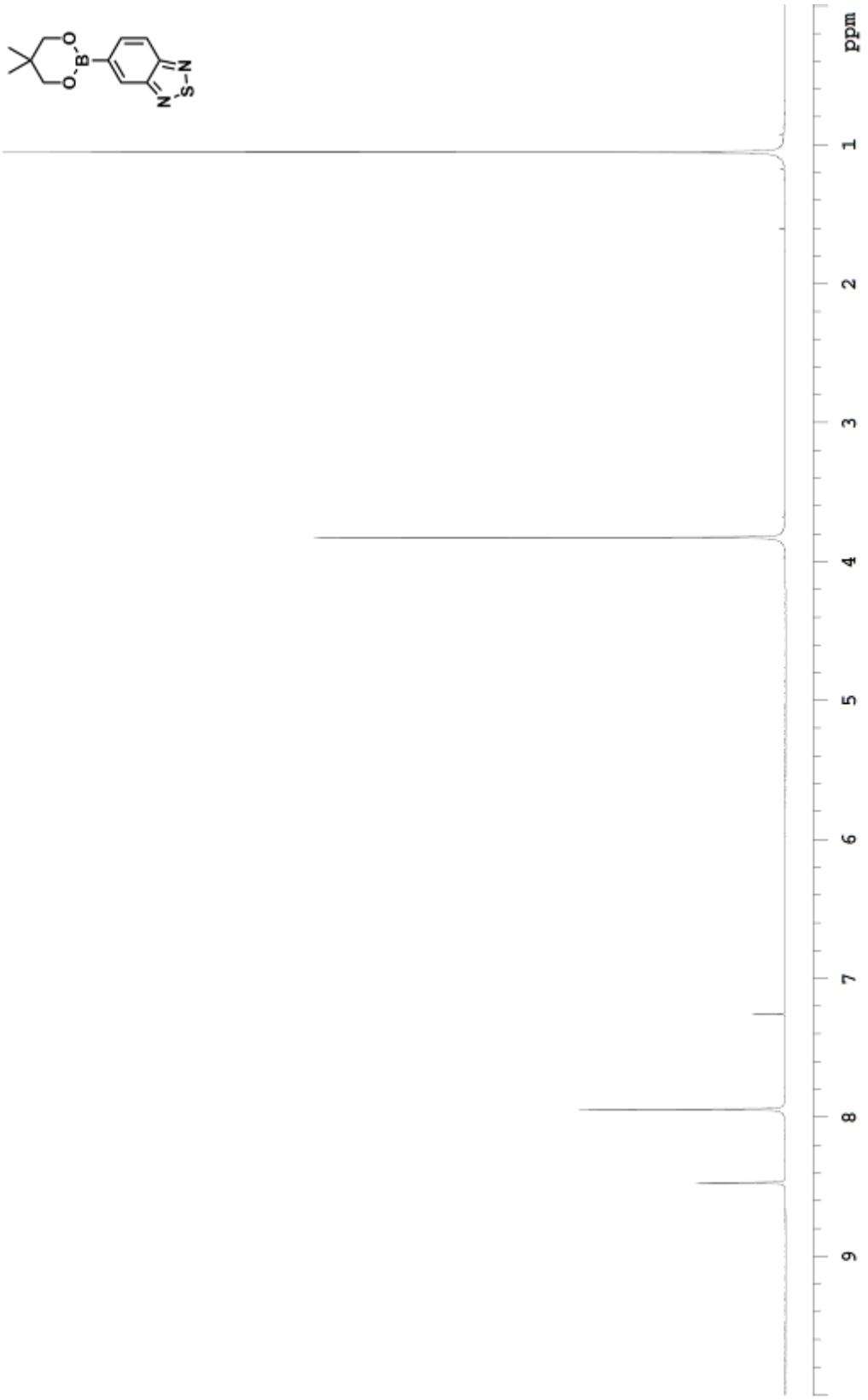
^{13}C NMR of **3c**, pyridine-d_5, 125 MHz, 23 ºC



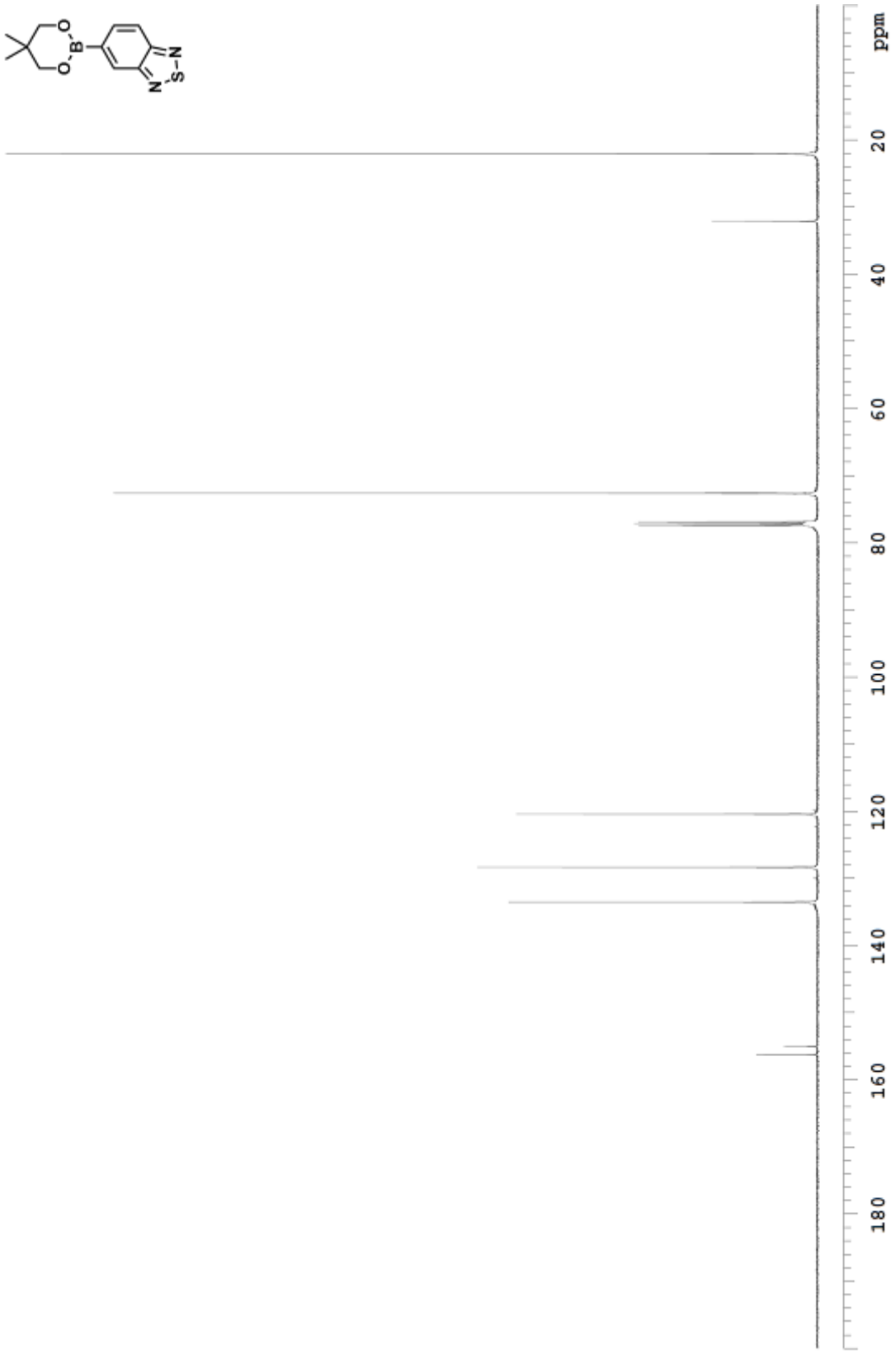
^{1}H NMR of **3d**, $CDCl_3$, 600 MHz, 23 ºC



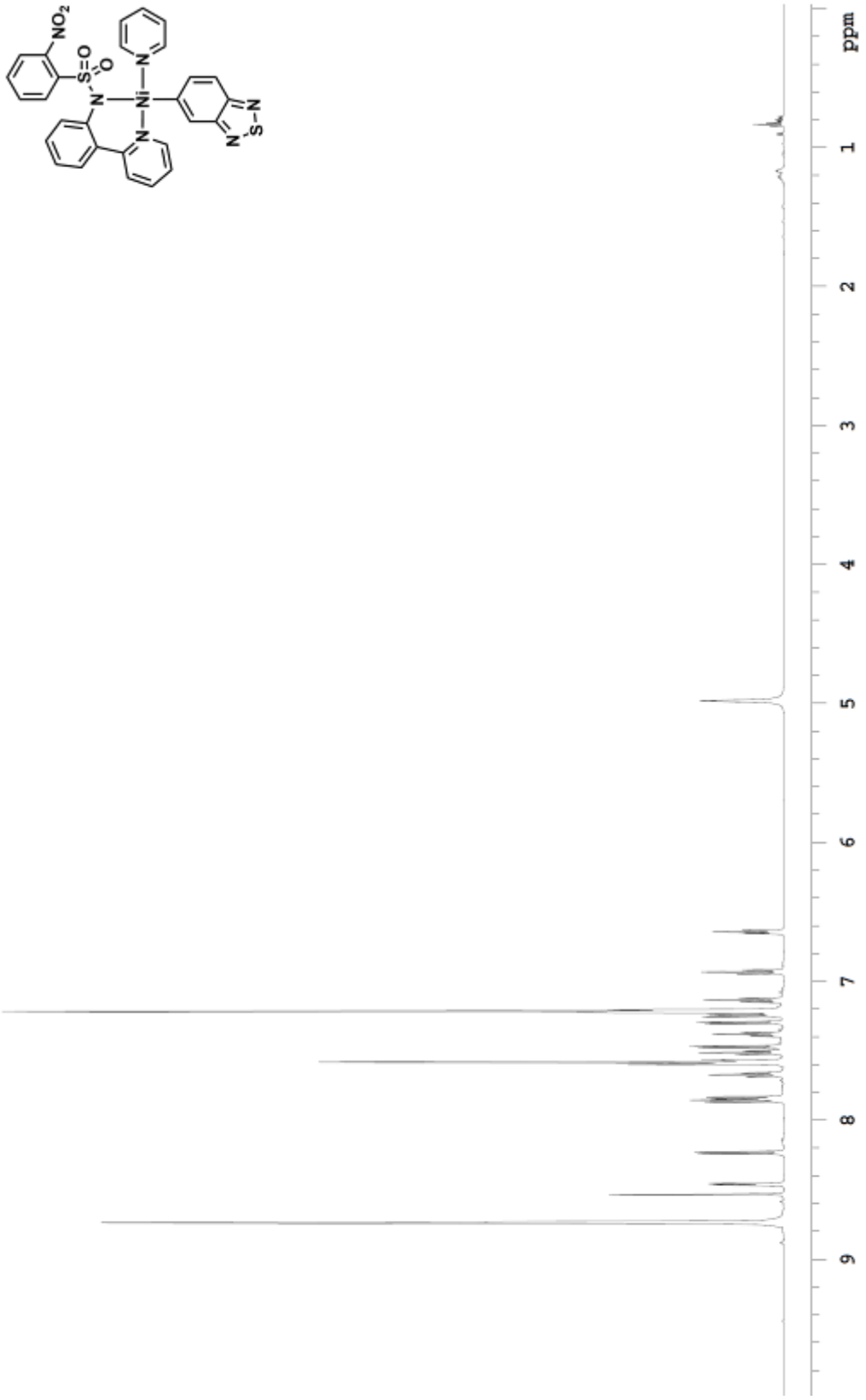
^{13}C NMR of **3d**, $CDCl_3$, 125 MHz, 23 ºC



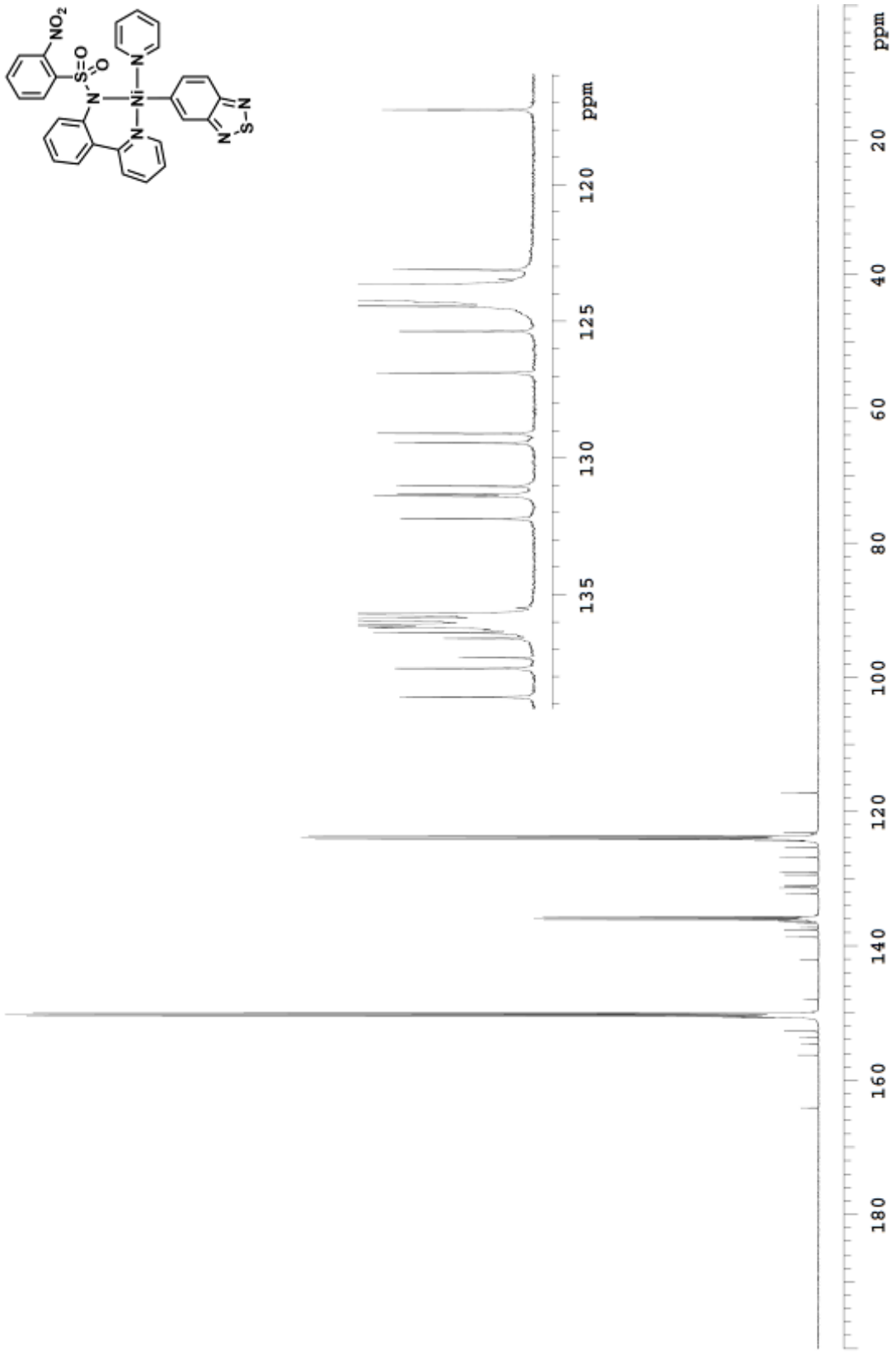
^{1}H NMR of **S2**, $CDCl_3$, 500 MHz, 23 ºC



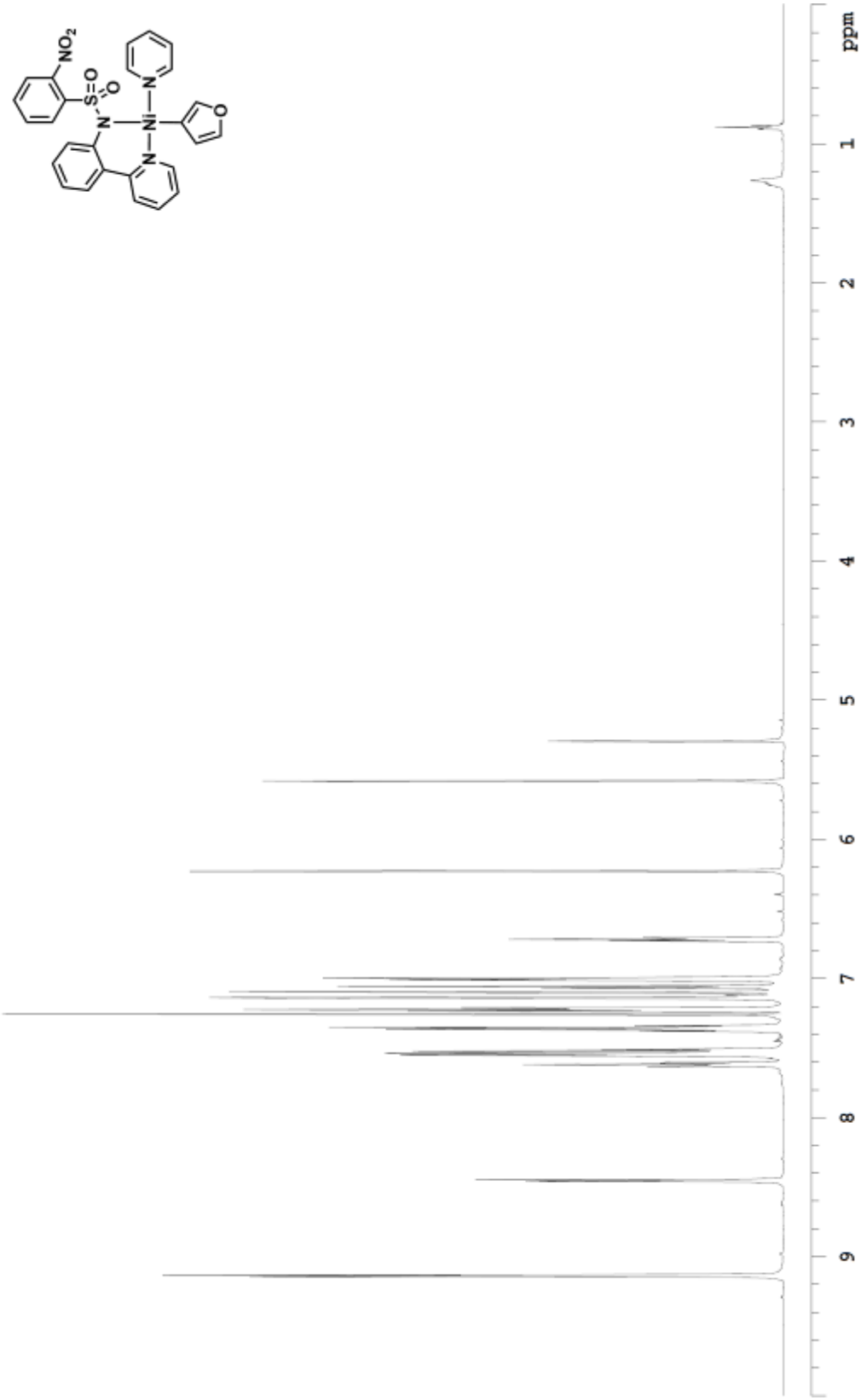
^{13}C NMR of **S2**, $CDCl_3$, 125 MHz, 23 ºC



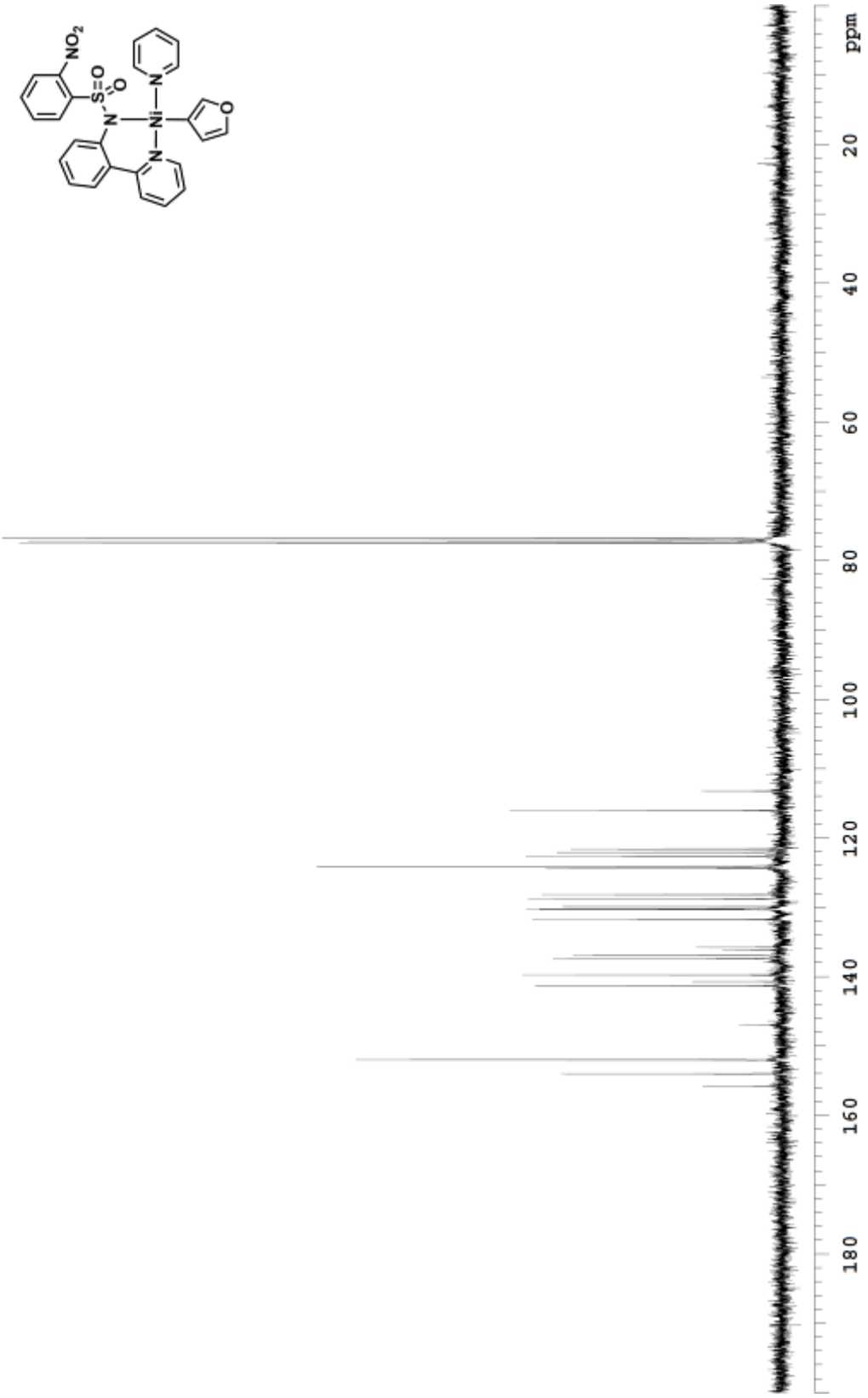
^{1}H NMR of **3e**, pyridine-d_5, 600 MHz, 23 ºC



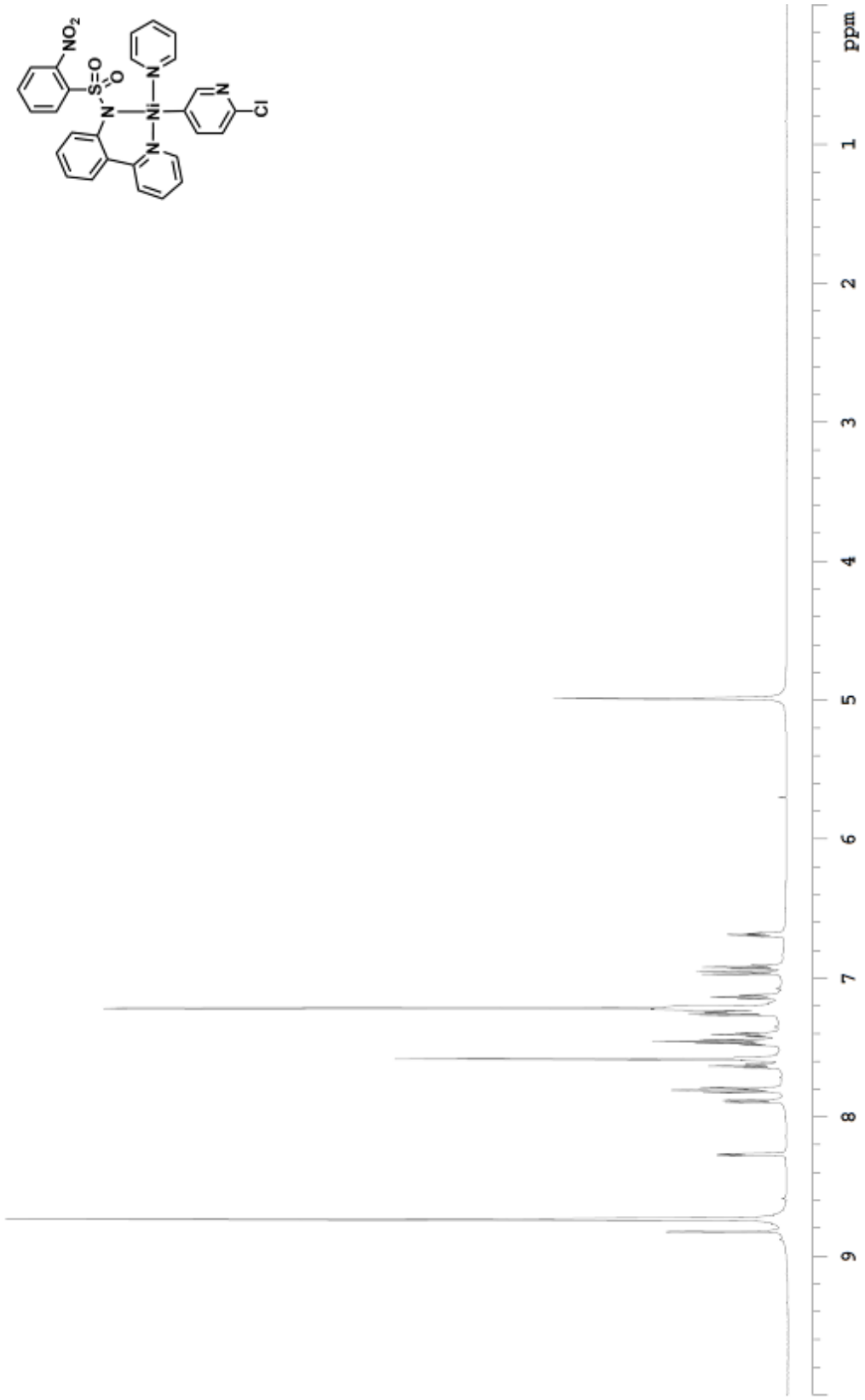
^{13}C NMR of **3e**, pyridine-d_5, 125 MHz, 23 ºC



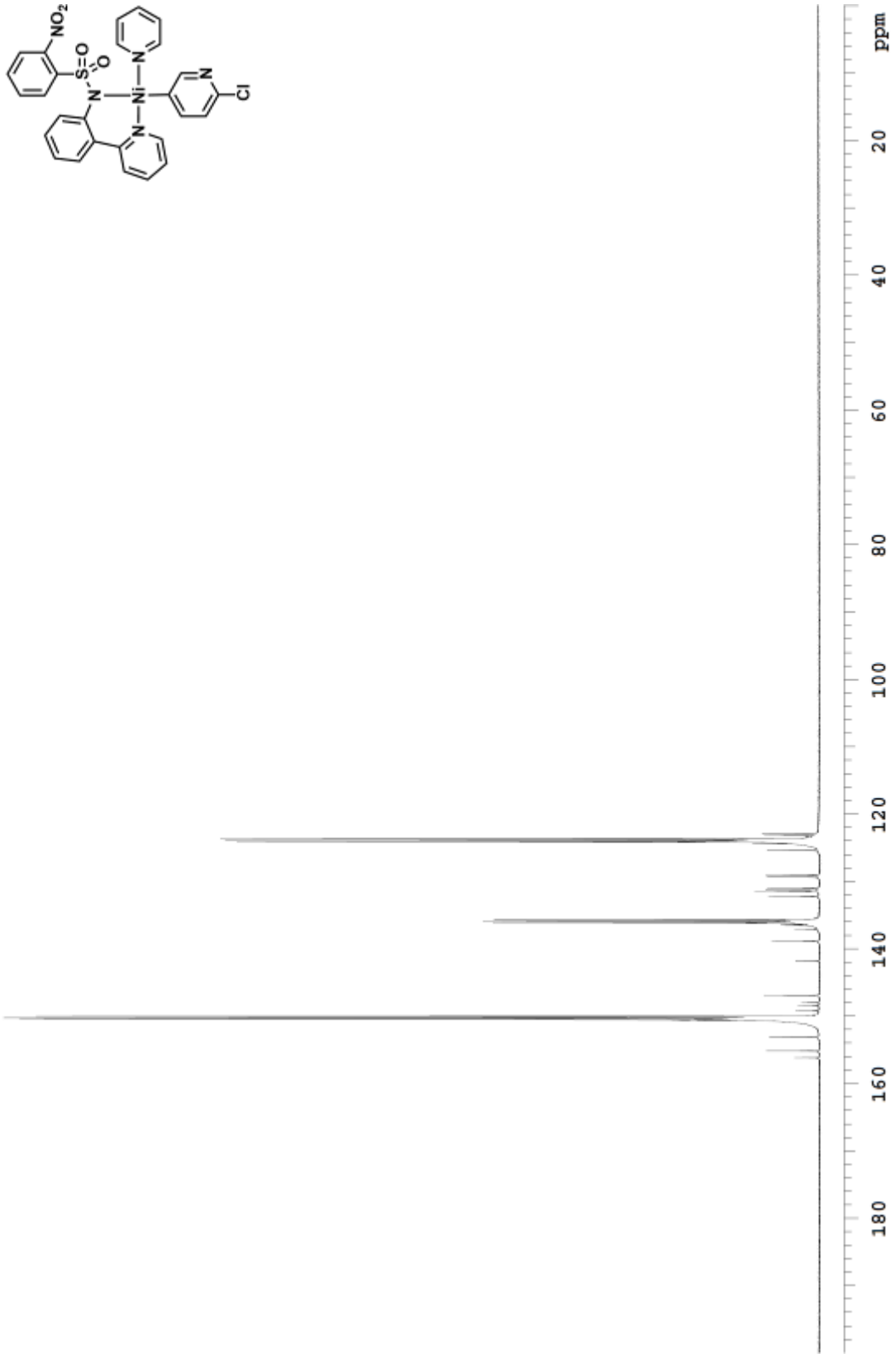
^{1}H NMR of **3f**, $CDCl_3$, 600 MHz, 23 ºC



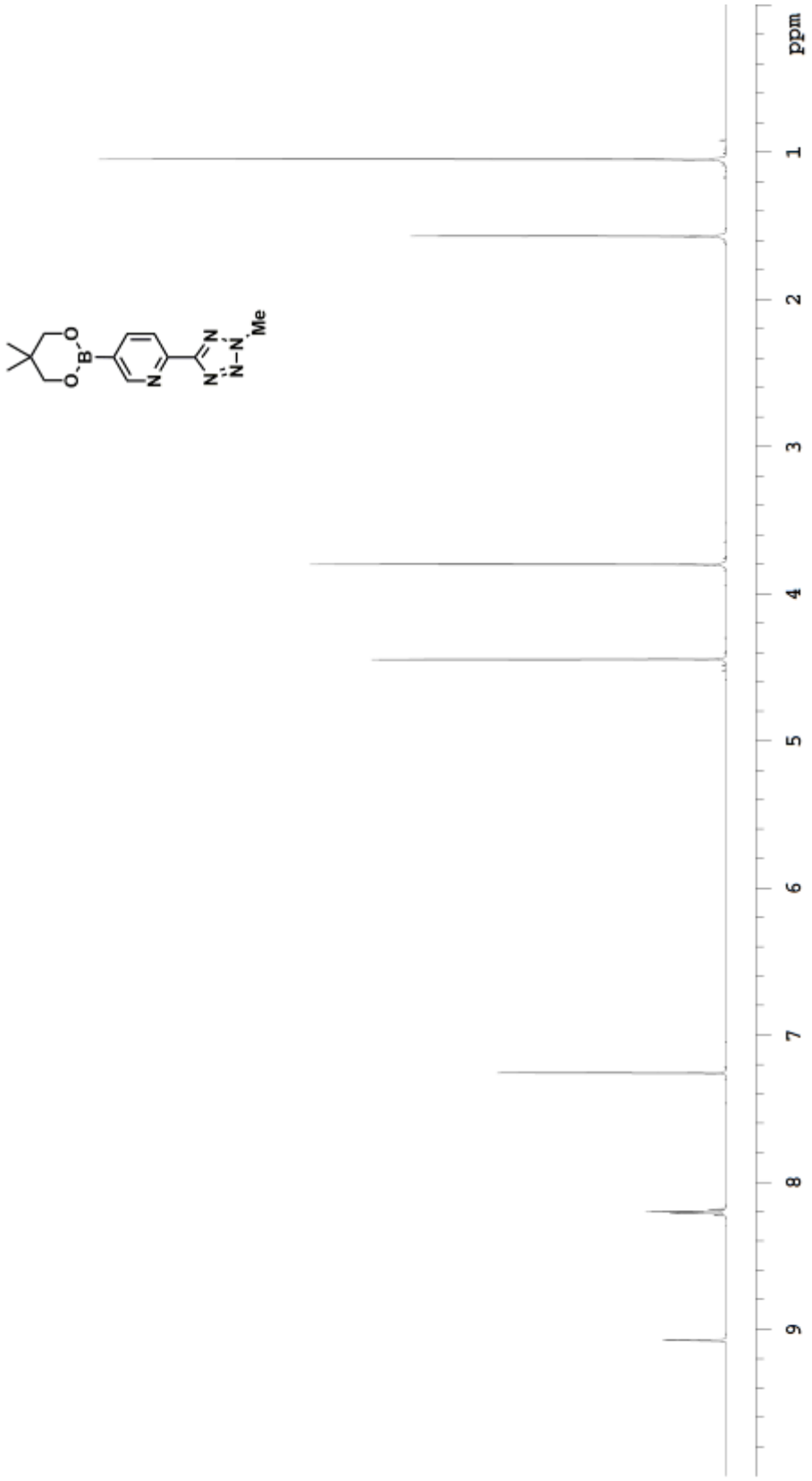
^{13}C NMR of **3f**, $CDCl_3$, 100 MHz, 23 ºC



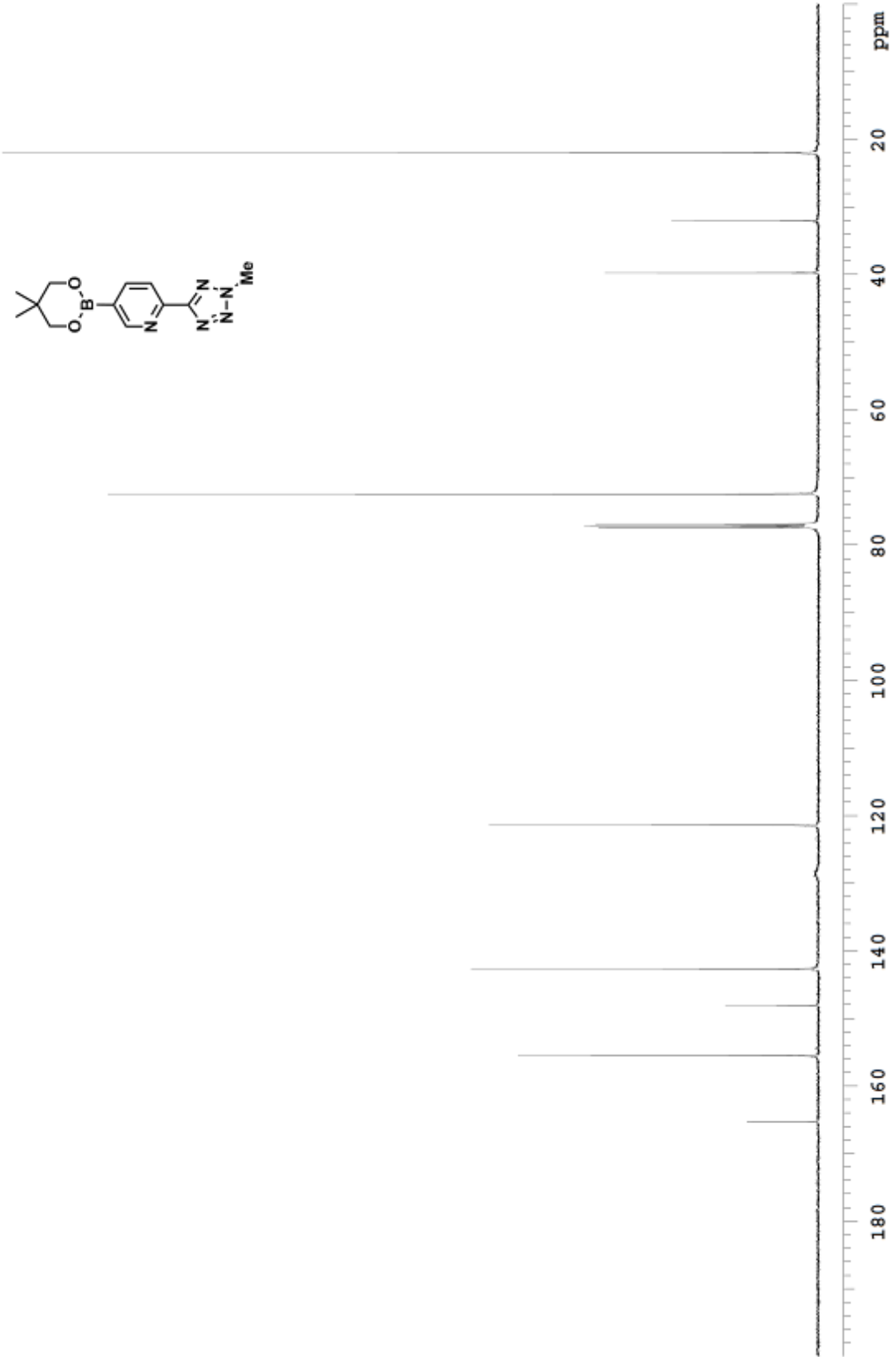
^{1}H NMR of **3g**, pyridine-d_5, 600 MHz, 23 ºC



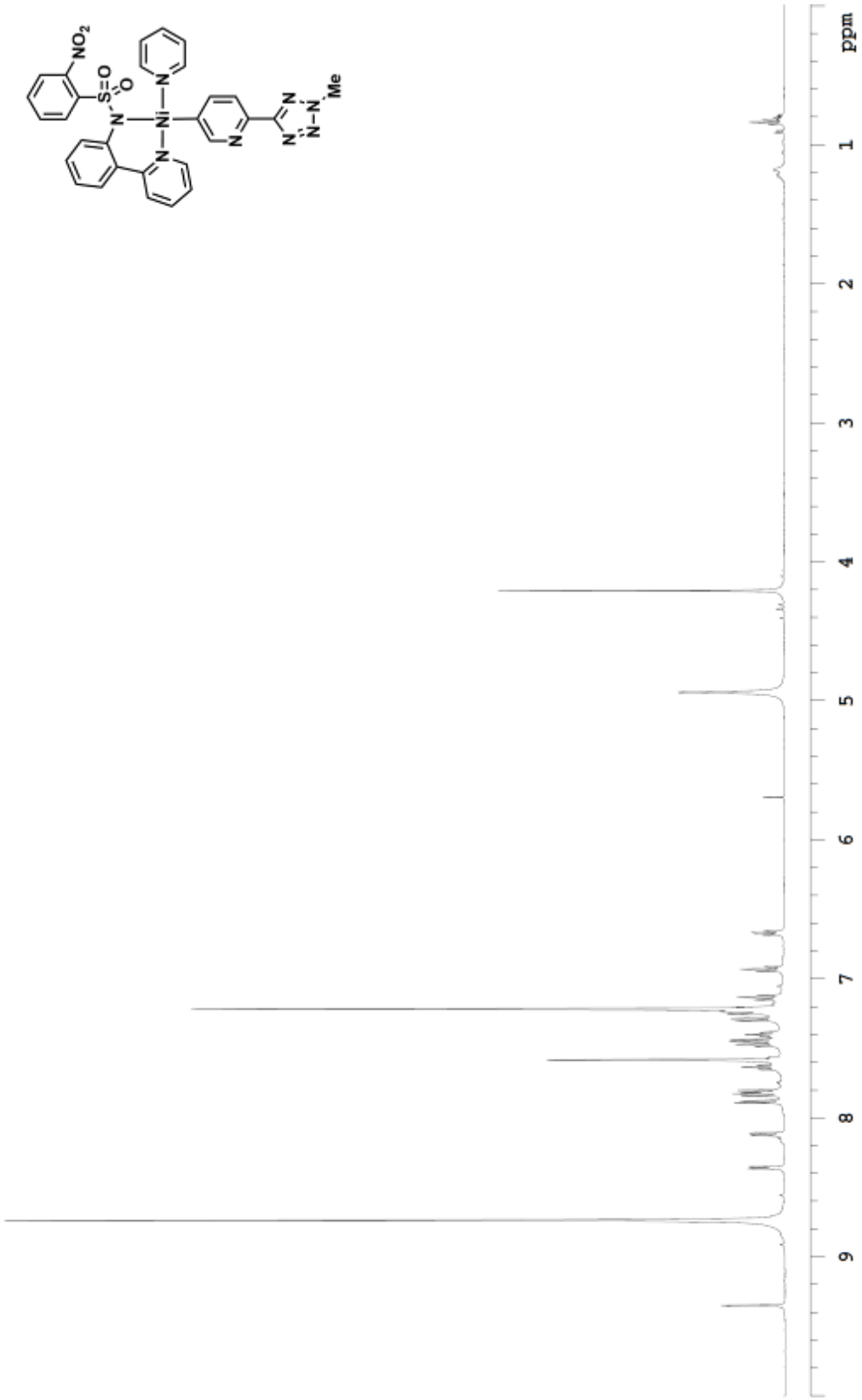
^{13}C NMR of **3g**, pyridine-d_5, 125 MHz, 23 ºC



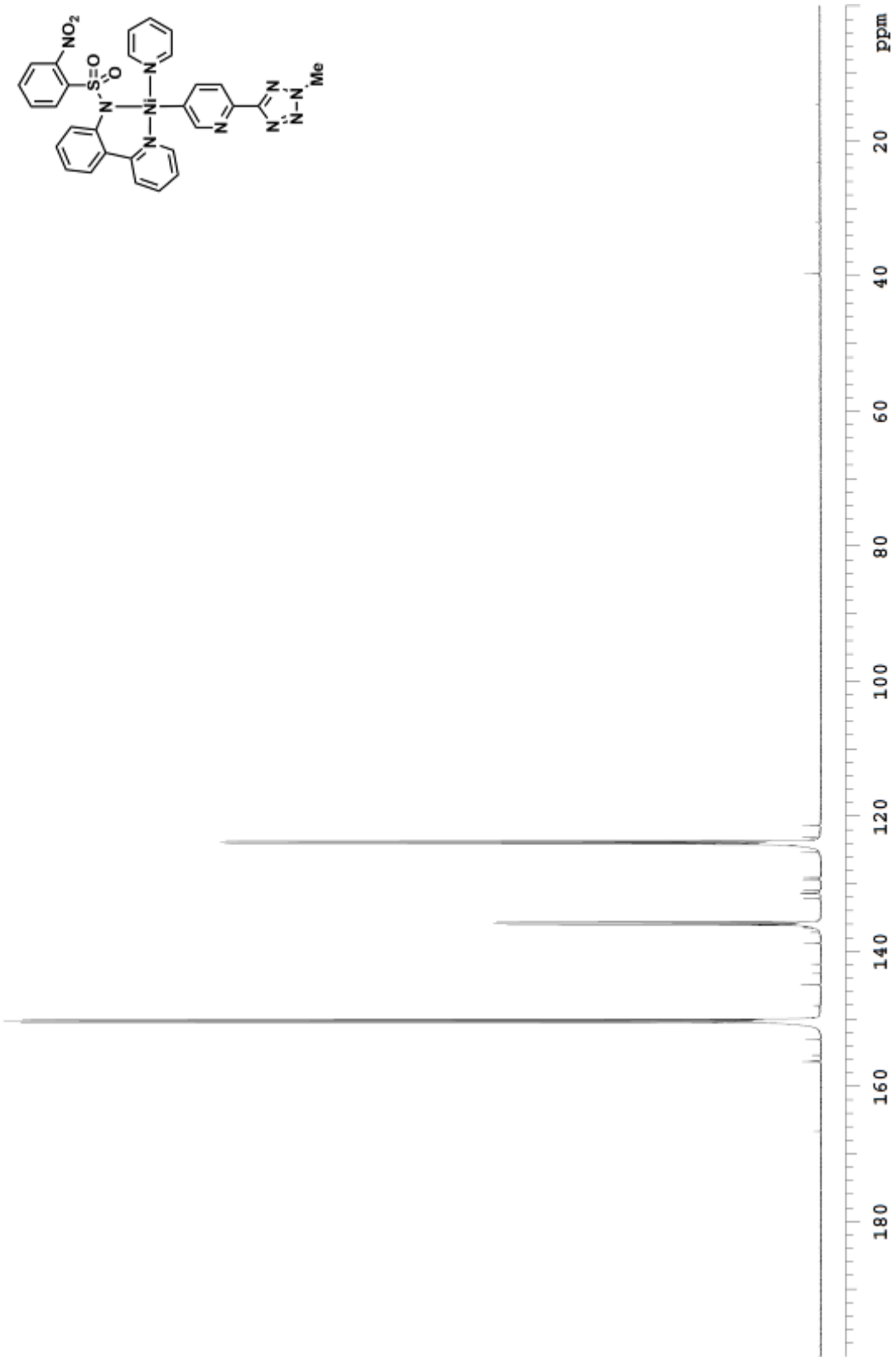
^{1}H NMR of **S3**, $CDCl_3$, 500 MHz, 23 ºC



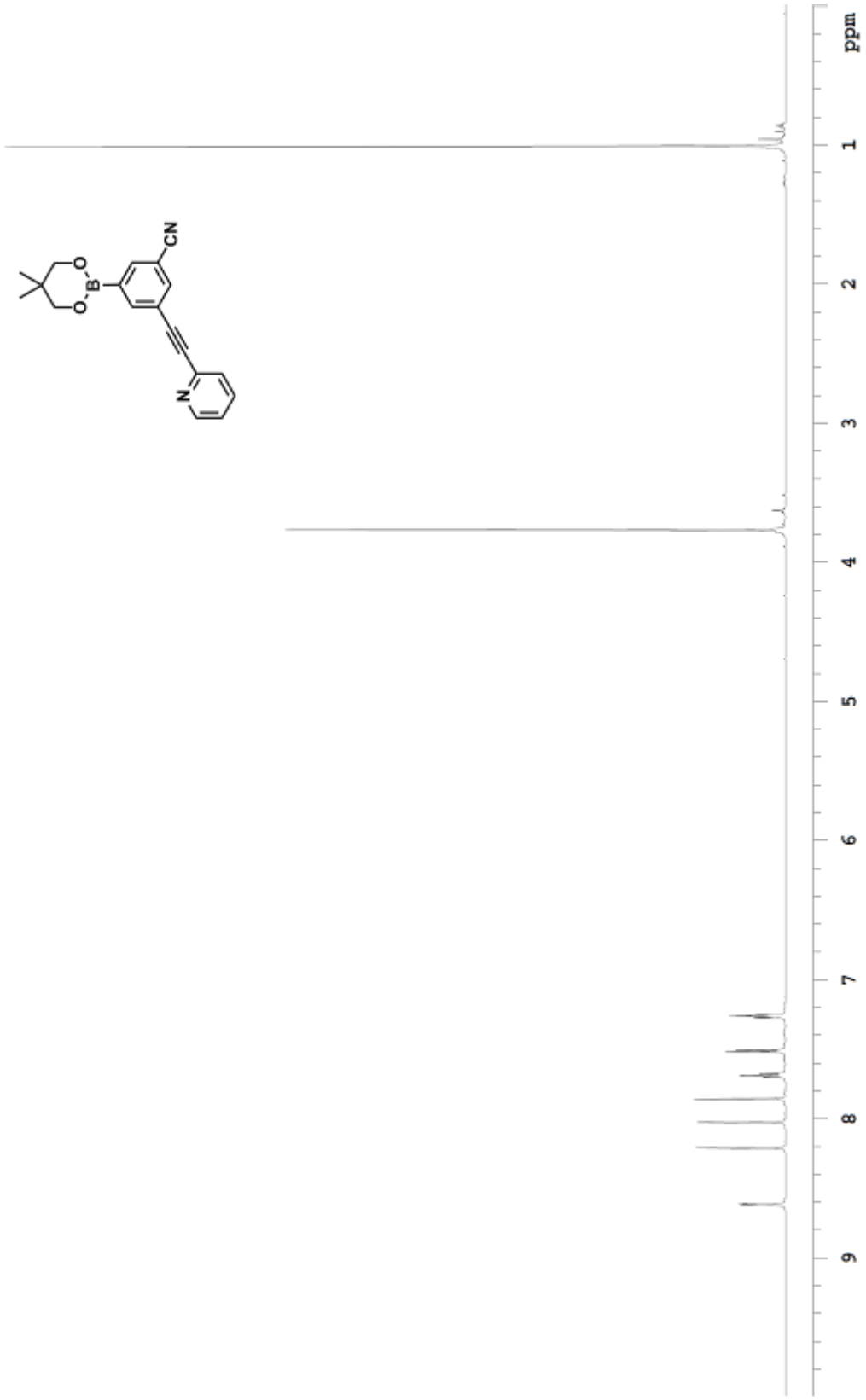
^{13}C NMR of **S3**, $CDCl_3$, 125 MHz, 23 ºC



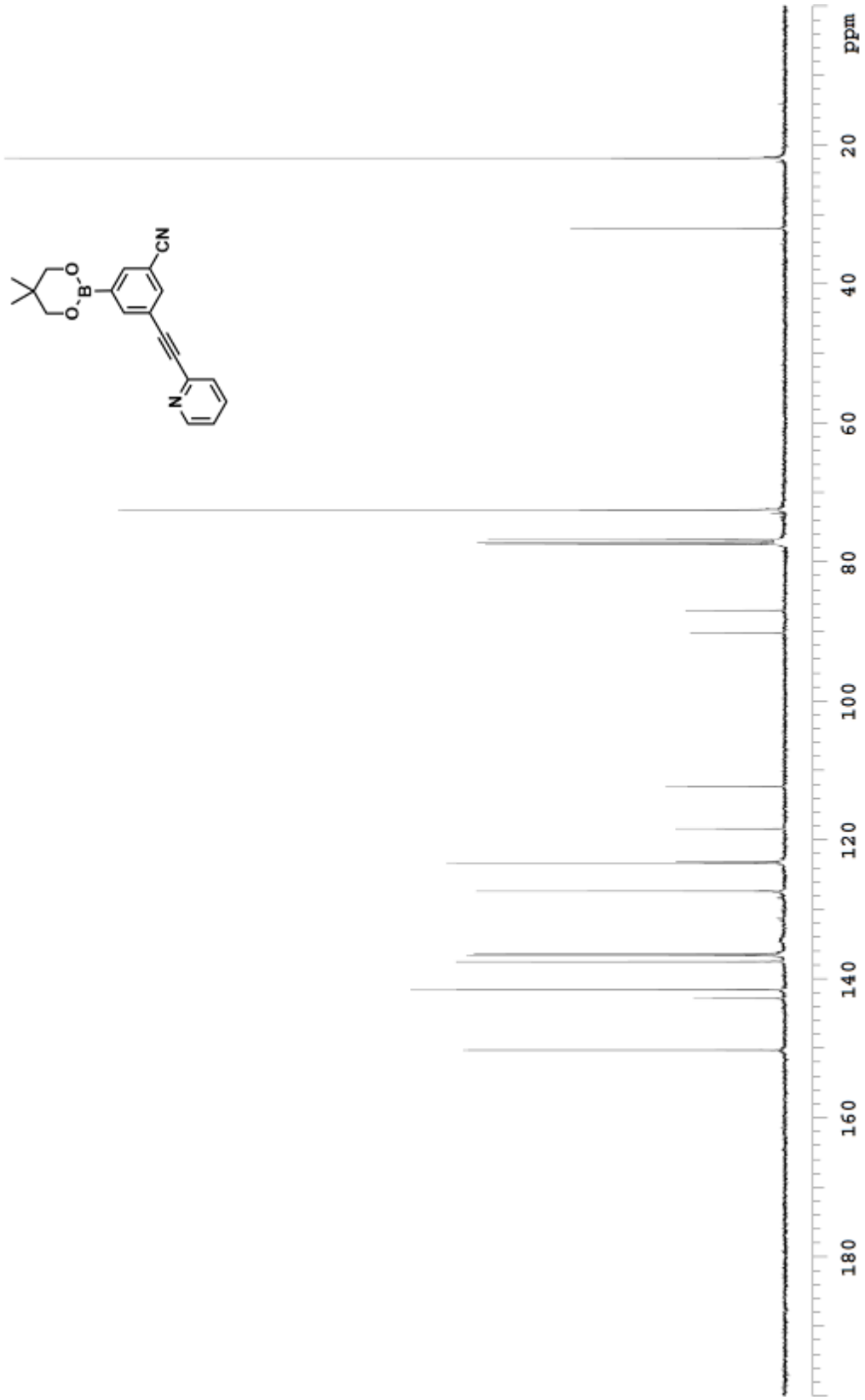
^{1}H NMR of **3h**, pyridine-d_5, 500 MHz, 23 ºC



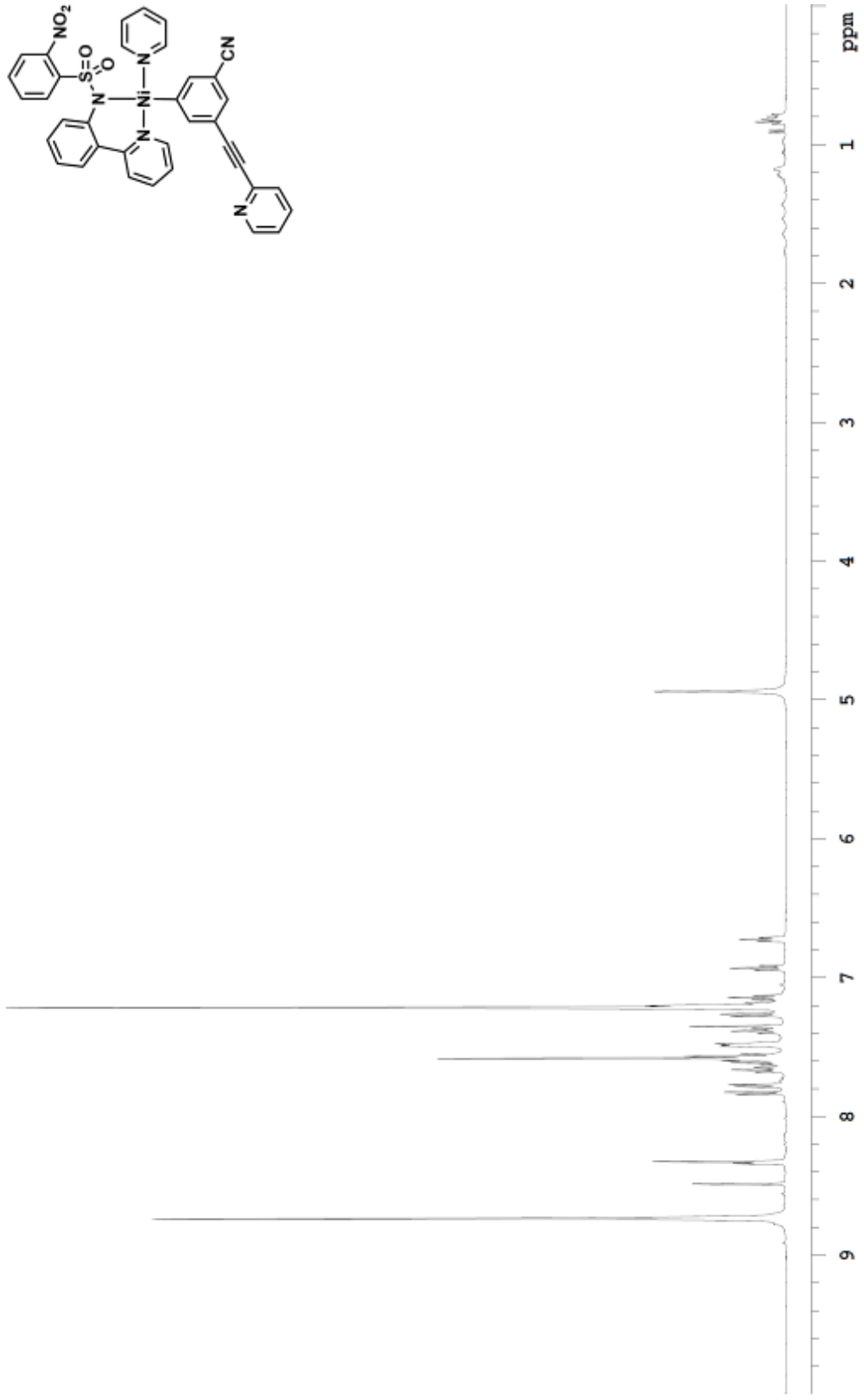
^{13}C NMR of **3h**, pyridine-d_5, 125 MHz, 23 ºC



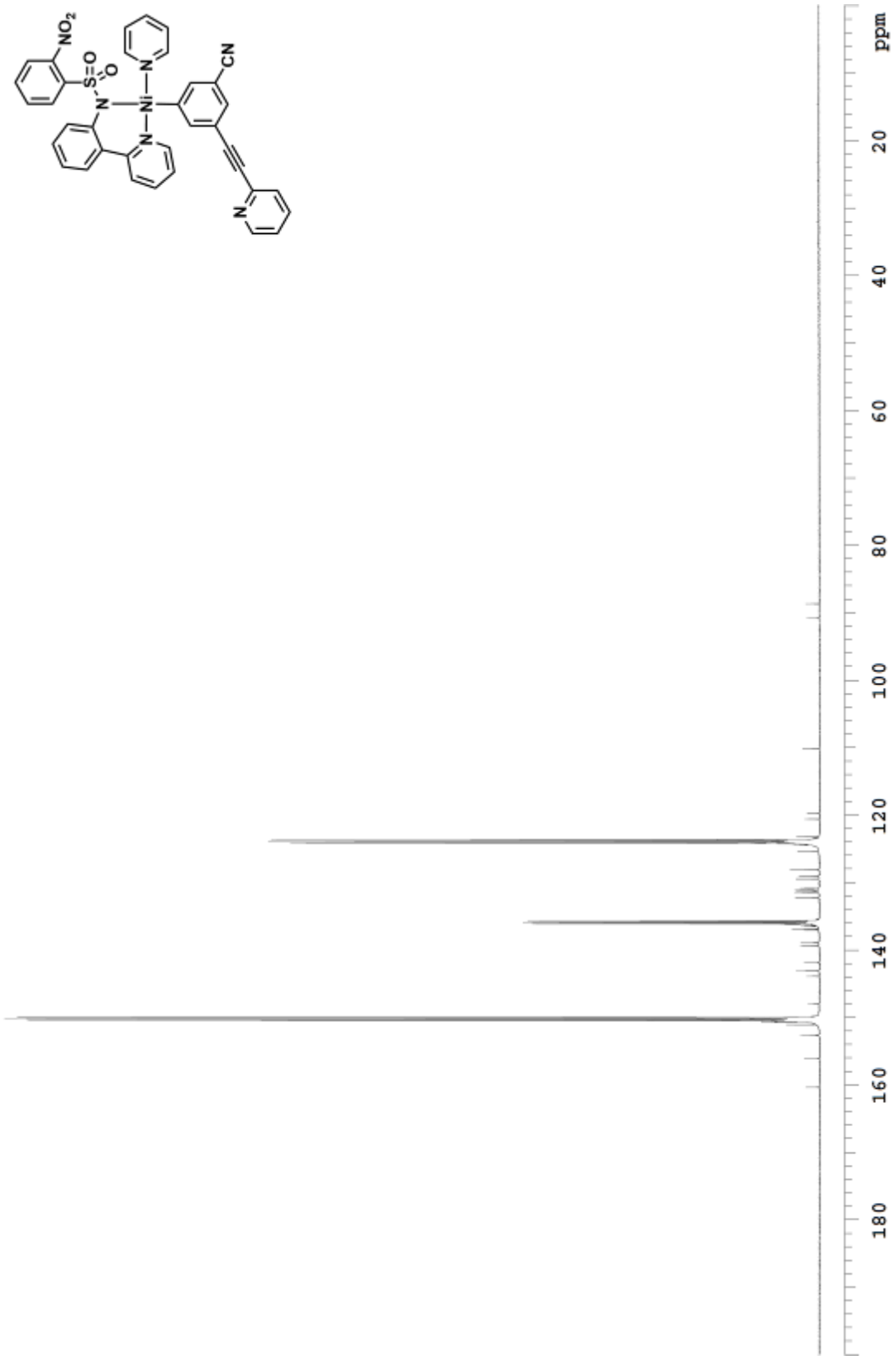
^{1}H NMR of **S4**, $CDCl_3$, 600 MHz, 23 ºC



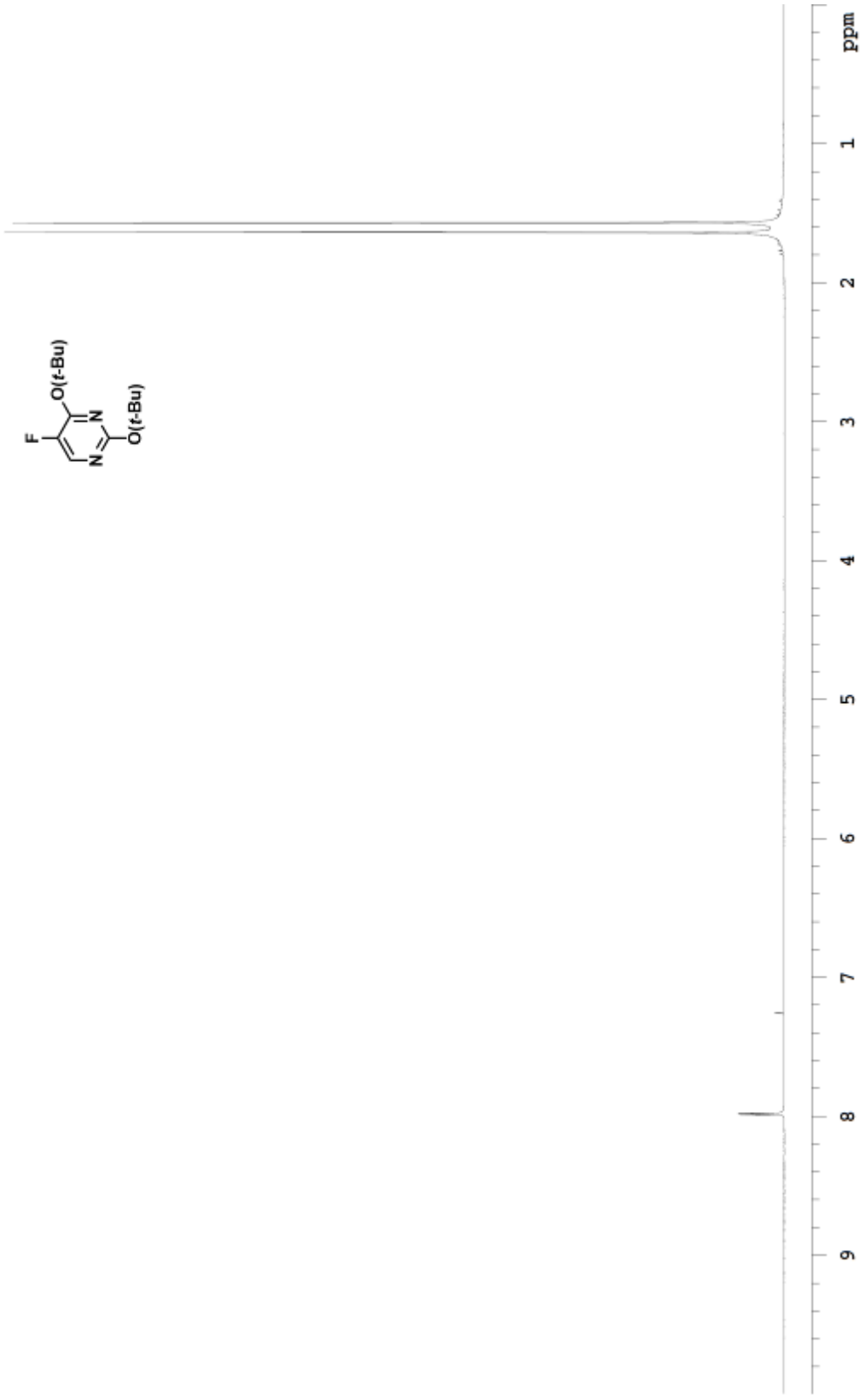
^{13}C NMR of **S4**, $CDCl_3$, 100 MHz, 23 ºC



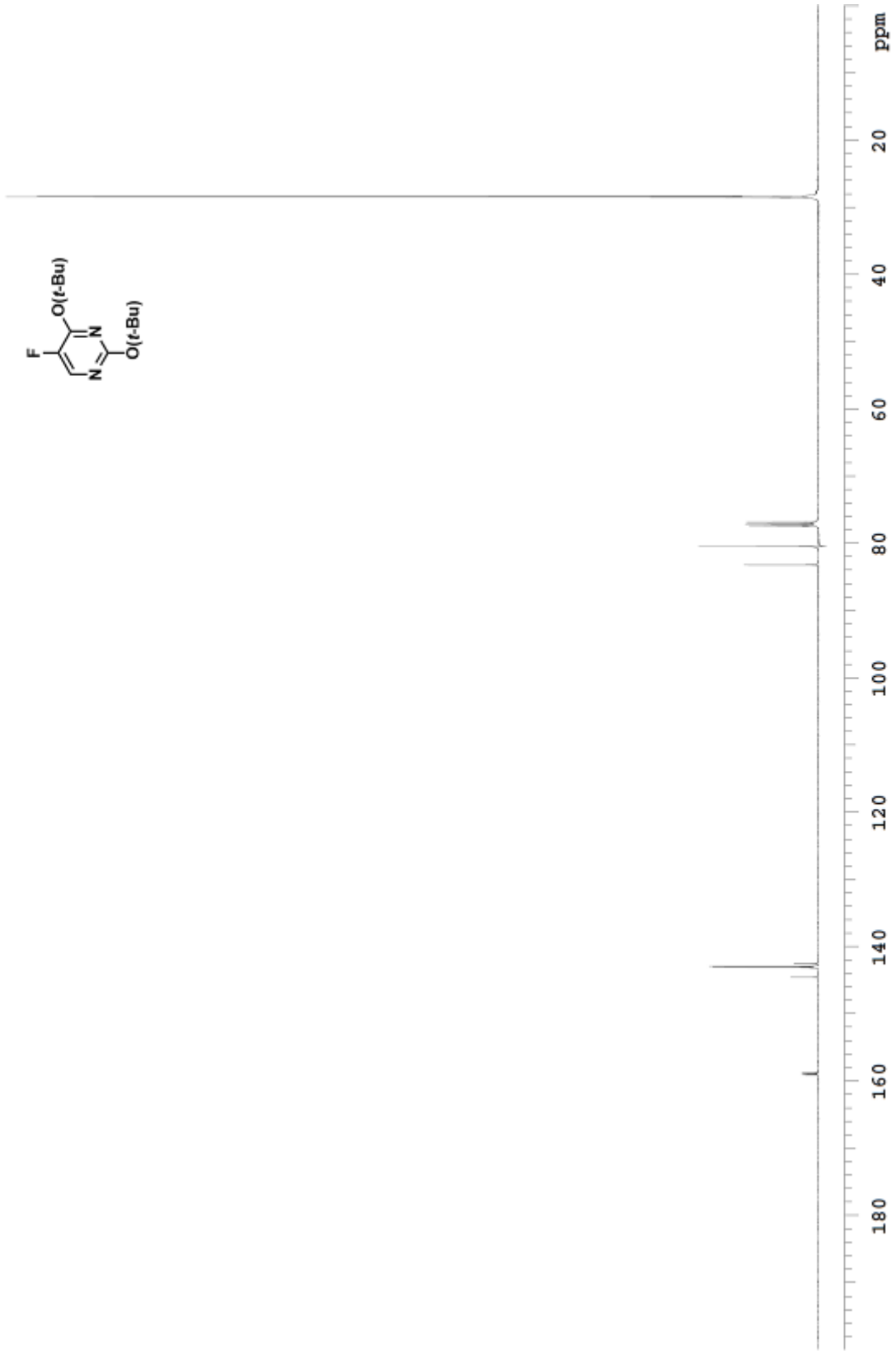
^{1}H NMR of **3i**, pyridine-d_5, 500 MHz, 23 ºC



^{13}C NMR of **3i**, pyridine-d_5, 125 MHz, 23 ºC



^{1}H NMR of **4a**, $CDCl_3$, 400 MHz, 23 ºC



^{13}C NMR of **4a**, $CDCl_3$, 125 MHz, 23 ºC

^{19}F NMR of **4a**, $CDCl_3$, 125 MHz, 23 ºC

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