Supporting Information for

A series of ligustrazine platinum(IV) complexes with potent anti-proliferative and anti-metastatic properties by causing chemotherapeutic and immunotherapeutic effects

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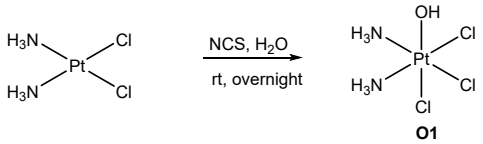
Contents

1. Supporting information for synthesis of the title compounds

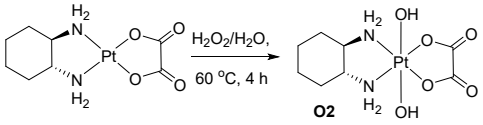
1.1 Preparation of compound **O1**



O1

Cisplatin (0.75 g, 2.5 mmol) was suspended in distilled water 150 mL and stirred at room temperature. *N*-chloro succinimide (NCS) solution (0.40 g, 3 mmol) in distilled water 150 mL was added to reaction system dropwise and the resultant mixture was stirred overnight at room temperature. After that, the solid was filtrated and the solution was concentrated under vacuum. The residue was washed with ethanol and ethylether. The product **O1** was obtained as a yellow solid (0.46 g, 52%).

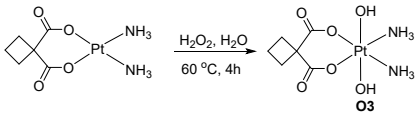
1.2 Preparation of compound **O2**



O2

A suspension of oxaliplatin (1.0 g, 2.5 mmol) in distilled water 30 mL was stirred at room temperature. Then H_2O_2 (30%) 50 mL was added drop wise. The mixture was kept stirring for 4 h at 60 °C. Then the resultant mixture was recrystallized at 4 °C. Crude product as yellow solid was obtained after filtration. Then recrystallization in water affords pure oxoplatin **O2** as white needles (0.9 g, 79%).

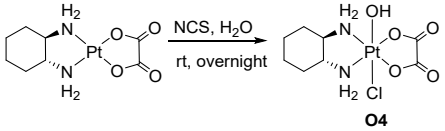
1.3 Preparation of compound **O3**



O3

A suspension of carboplatin (0.93 g, 2.5 mmol) in distilled water 30 mL was stirred at room temperature. Then H_2O_2 (30%) 50 mL was added drop wise. The mixture was kept stirring for 4 h at 60 °C. Then the resultant mixture was recrystallized at 4 °C. Crude product as yellow solid was obtained after filtration. Then recrystallization in water afford pure oxoplatin **O3** as white solid (0.84 g, 83%).

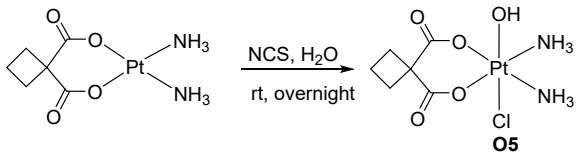
1.4 Preparation of compound **O4**



O4

Oxaliplatin (1.0 g, 2.5 mmol) was suspended in distilled water 150 mL and stirred at room temperature. *N*-chloro succinimide (NCS) solution (0.392 g, 2.94 mmol) in distilled water 150 mL was added to reaction system dropwise and the resultant mixture was stirred overnight at room temperature. After that, the solid was filtrated and the solution was concentrated under vacuum. The residue was washed with ethanol and ethylether. The product was obtained as a yellow solid (1.0 g, 84%).

1.5 Preparation of compound **O5**



O5

Carboplatin (0.93 g, 2.5 mmol) was suspended in distilled water 150 mL and stirred at room temperature. *N*-chloro succinimide (NCS) solution (0.400 g, 3.0 mmol) in distilled water 150 mL was added to reaction system dropwise and the resultant mixture was stirred overnight at room temperature. After that, the solid was filtrated and the solution was concentrated under vacuum. The residue was washed with ethanol and ethylether. The product **O5** was obtained as a yellow solid (0.58 g, 55%).

2. Supporting information for anti-metastasis activities *in vitro*

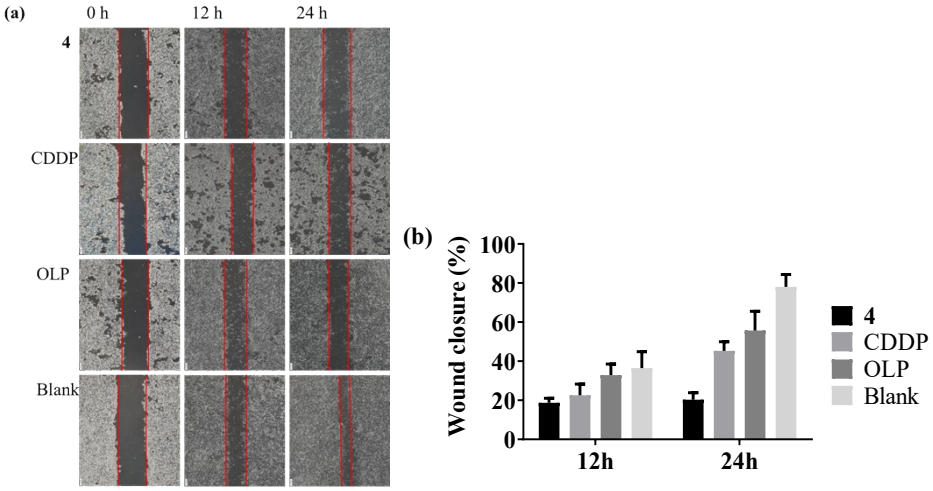


Figure S1 Migration inhibition of complex **4**, CDDP and OLP at 10 μM to 4T1 cells *in vitro*. The extent of wound healing was observed at 0 h, 12 h, and 24 h. (a) Representative images. (b) Analysis of wound closure.

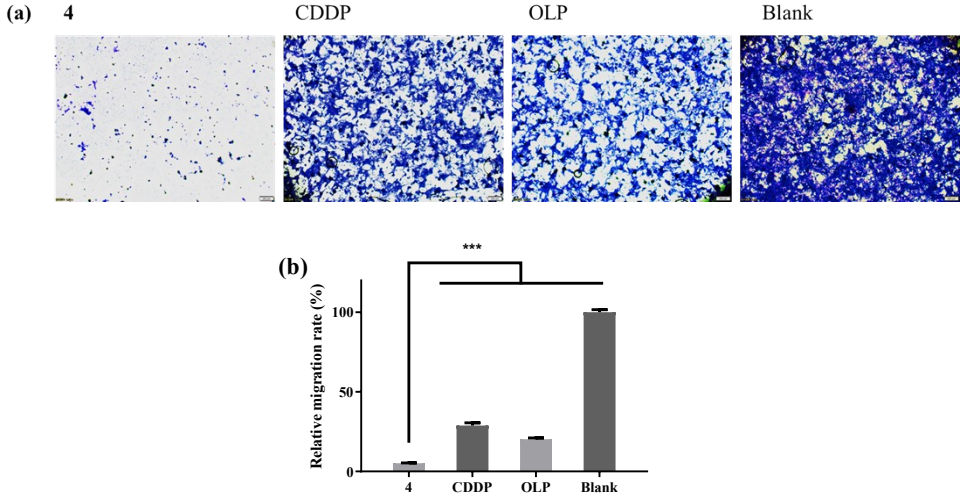


Figure S2 Evaluation of migration inhibition properties of complex **4**, CDDP and OLP (10 μM) to 4T1 cells using transwell assay *in vitro*. The tumor cells were treated with and without platinum complexes for 24 h. (a) Representative images. (b) Analysis of relative migration rate. ***$P < 0.001$.

3. Supporting information for reduction and DNA binding properties of compound 4

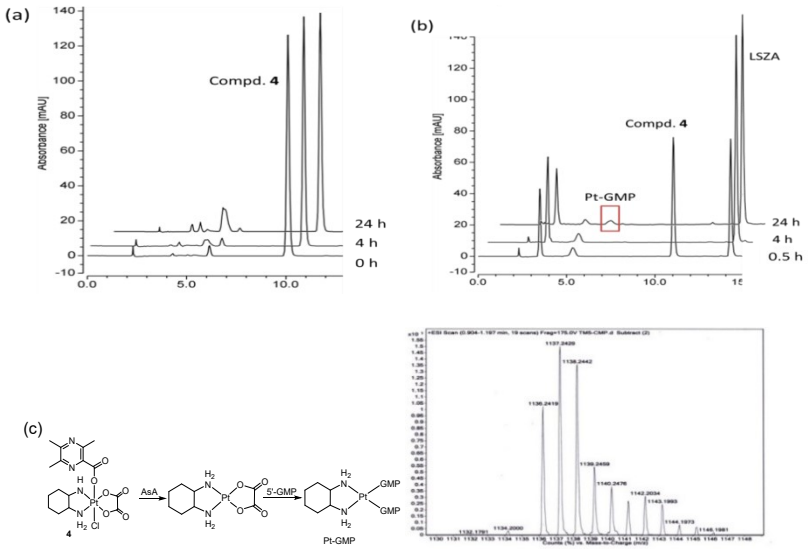


Figure S3 The reduction of compound **4** in the presence of AsA, and its interaction with DNA. (a) Compound **4** (0.5 mM) in PBS incubated for 24 h at 37 °C. (b) Solution of compound **4** (0.5 mM), AsA (1 mM) and GMP (3 mM) in PBS incubated for 24 h at 37 °C. (c) HRMS of Pt-GMP. Reprinted with permission from Wang et al., Dalton Trans. 2016, 45, 10366–10374. Copyright 2016 The Royal Society of Chemistry [S1]

4. Supporting information for ROS generation

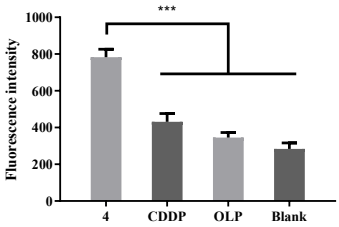


Figure S4 Staining of ROS with DCFH-DA in A549 cells treated by platinum complexes. A549 cells were incubated with and without compounds **4**, CDDP and OLP (10 μM) for 24 h at 37 ˚C, and stained by DCFH-DA. The ROS production was quantified by flow cytometry. ***$P < 0.001$.

5. ^{1}H NMR, ^{13}C NMR, MS and HRMS spectra

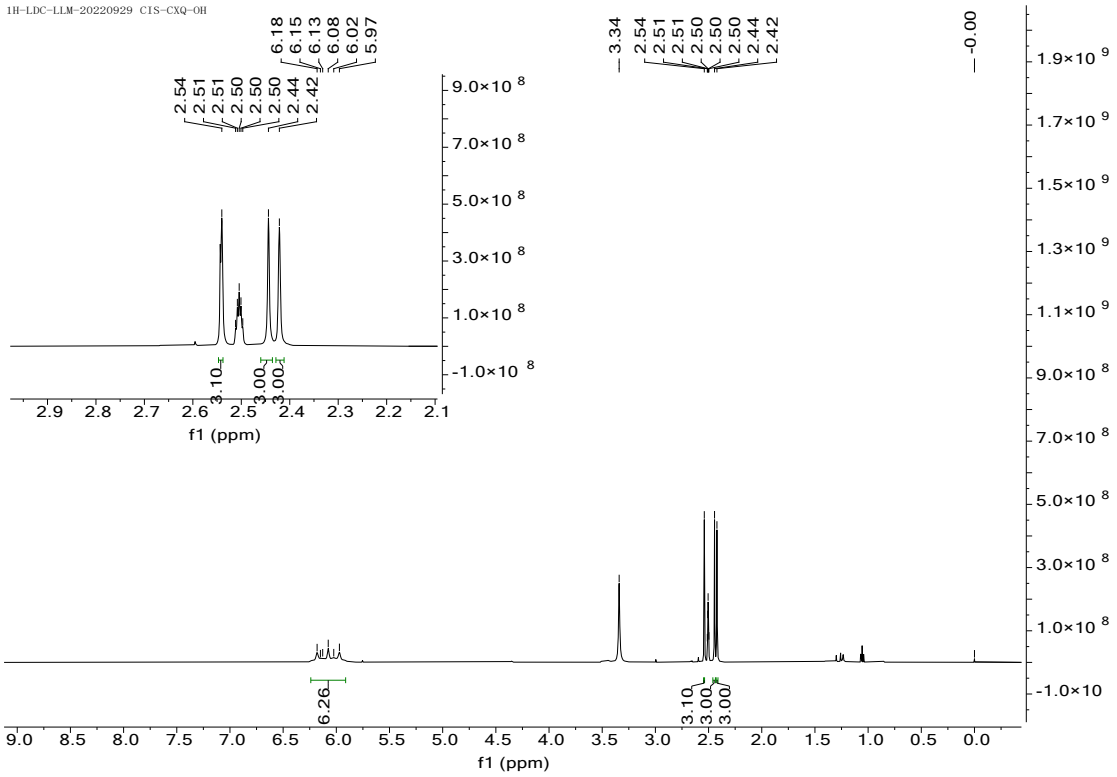


Figure S6 ^{1}H NMR spectrum for complex **1**.

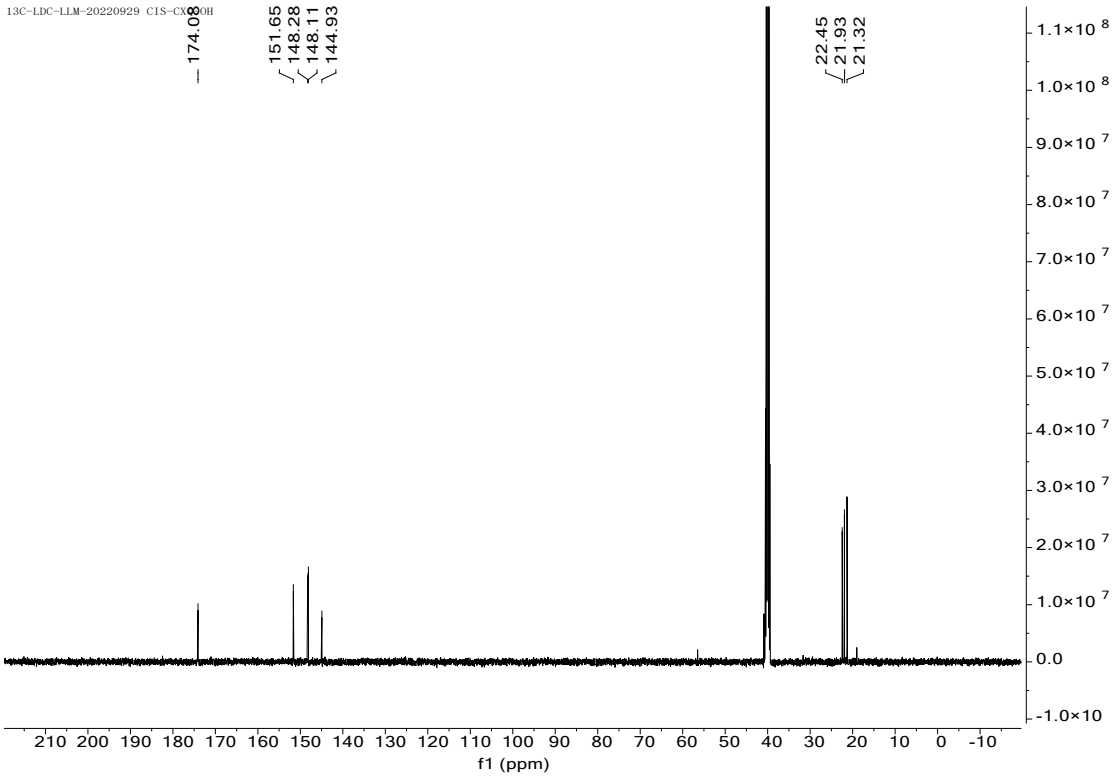


Figure S7 ^{13}C NMR spectrum for complex **1**.

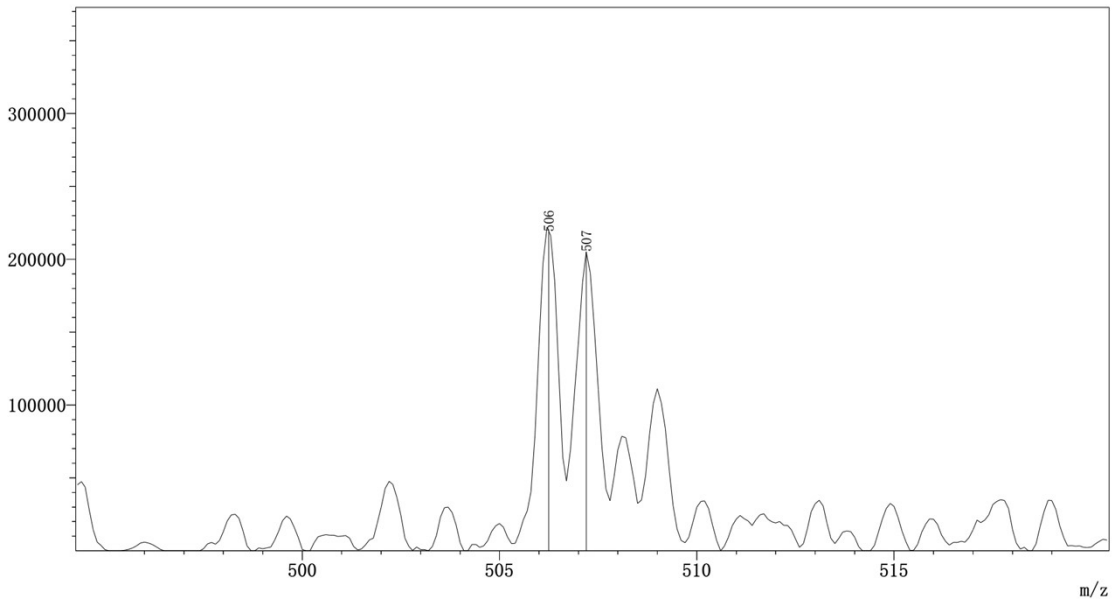


Figure S8 ESI-MS spectrum for complex **1**.

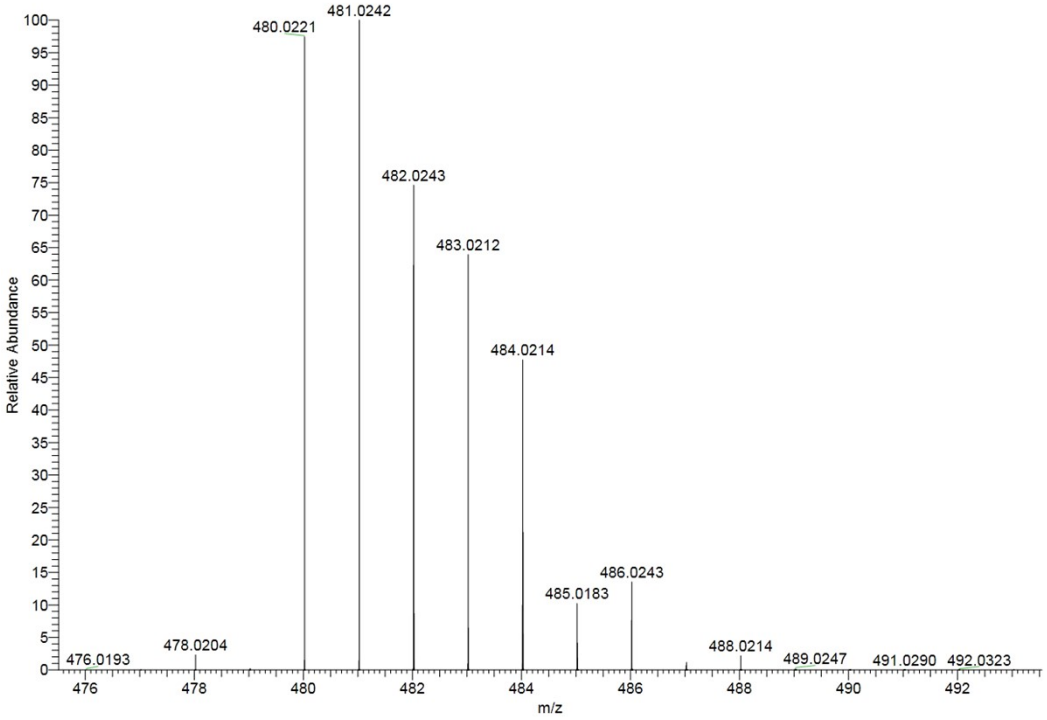


Figure S9 HRMS spectrum for complex **1**.

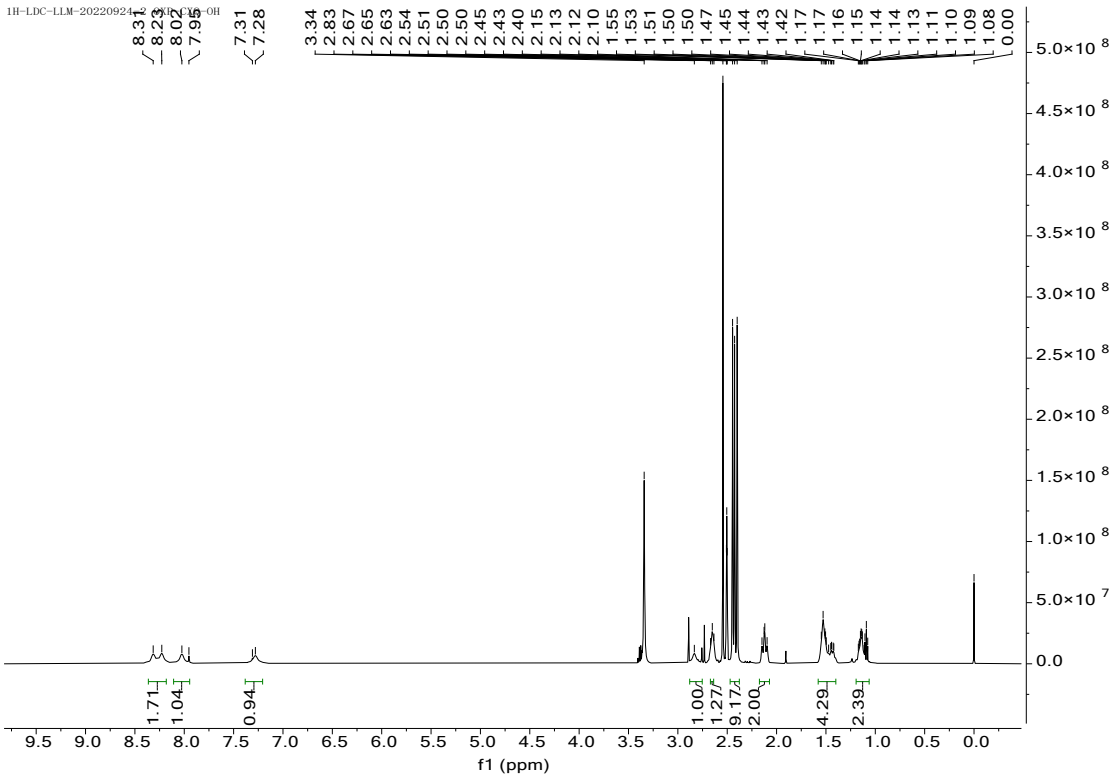


Figure S10 ^{1}H NMR spectrum for complex **2**.

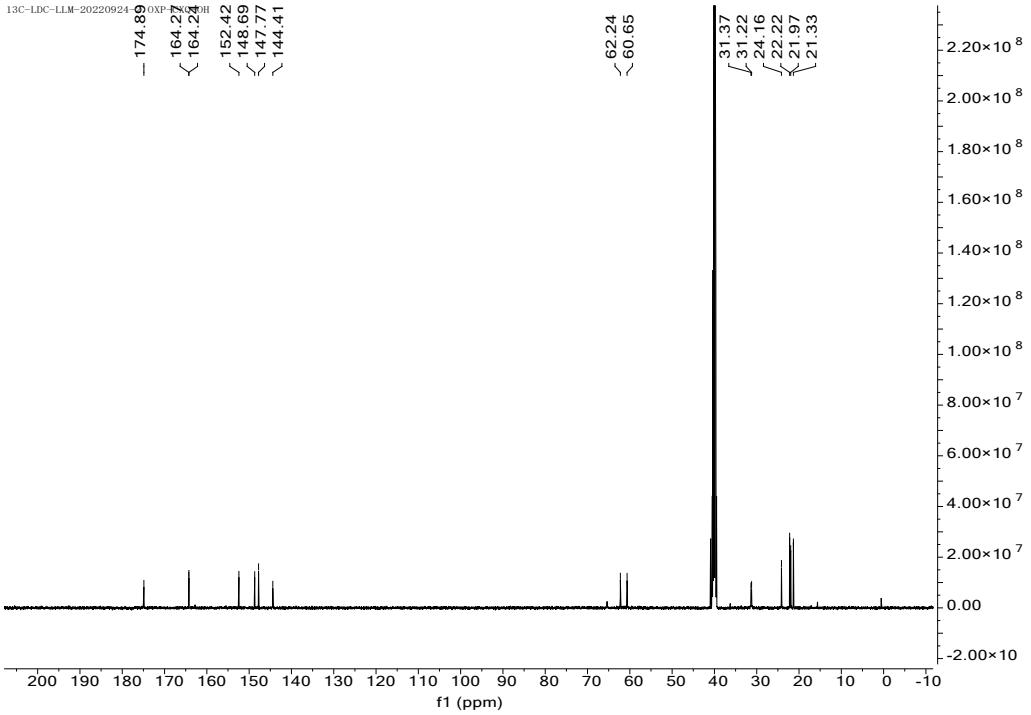


Figure S11 ^{13}C NMR spectrum for complex **2**.

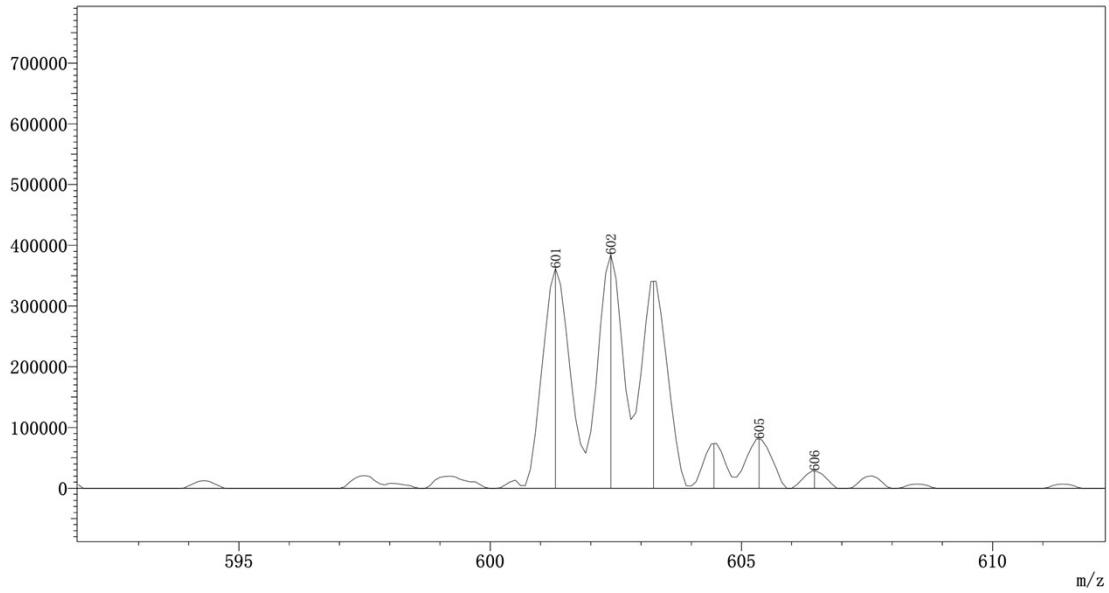


Figure S12 ESI-MS spectrum for complex **2**.

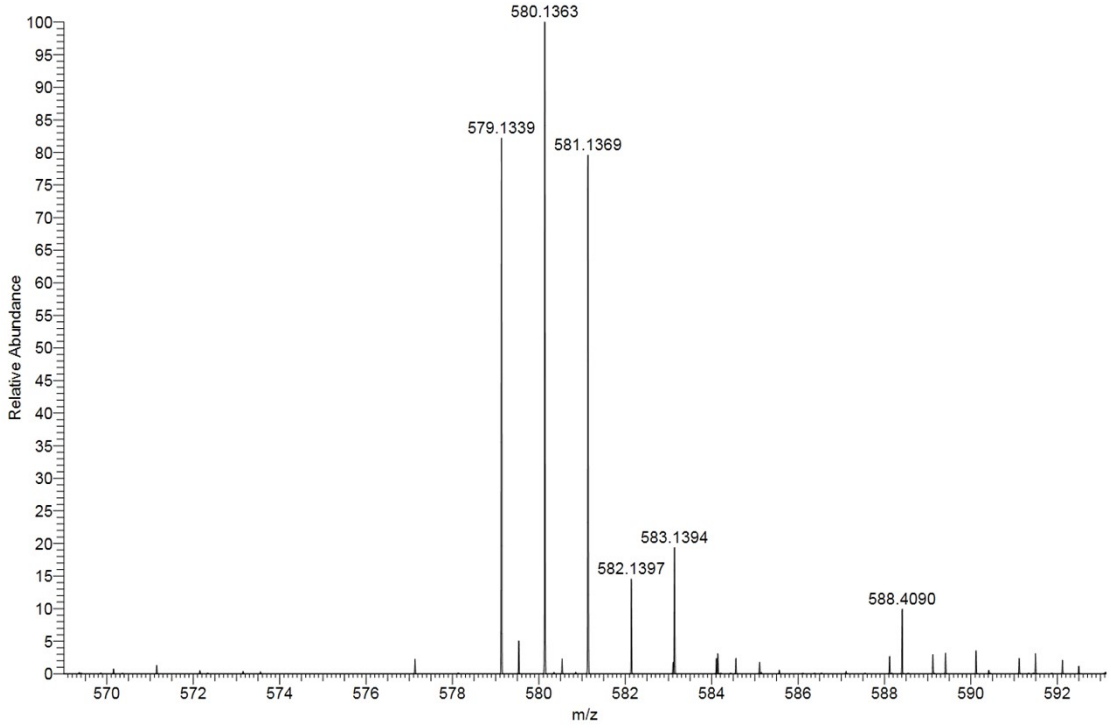


Figure S13 HRMS spectrum for complex **2**.

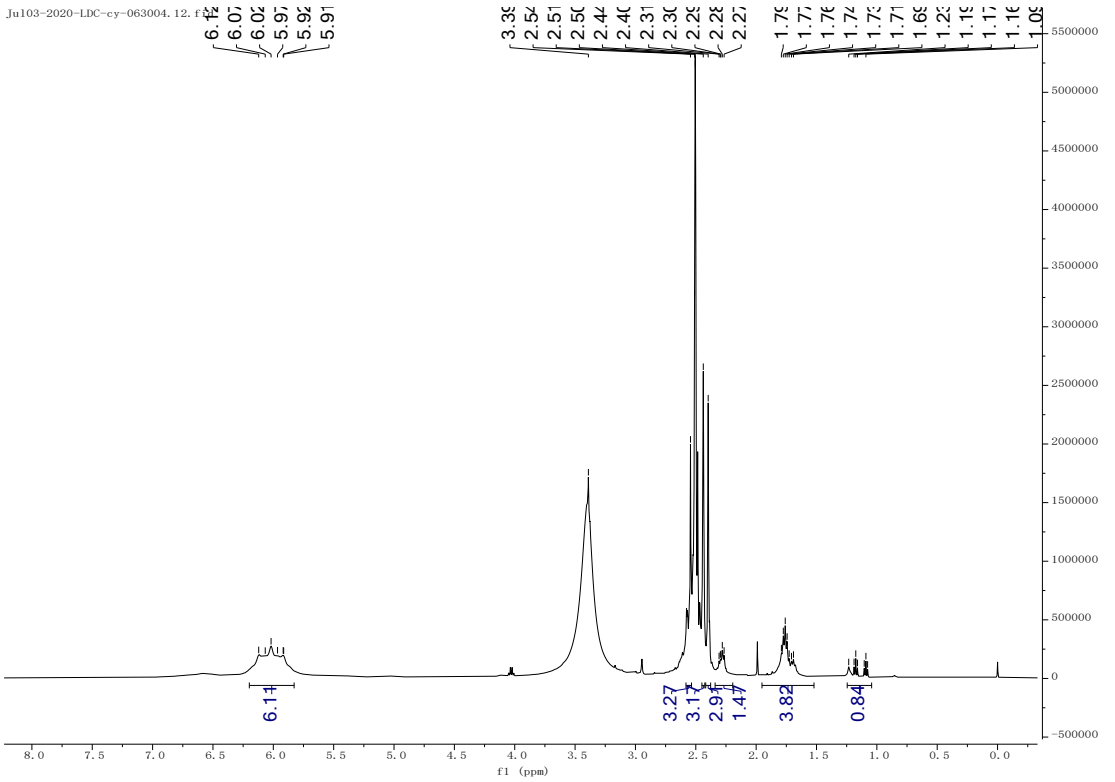


Figure S14 ^{1}H NMR spectrum for complex **3**.

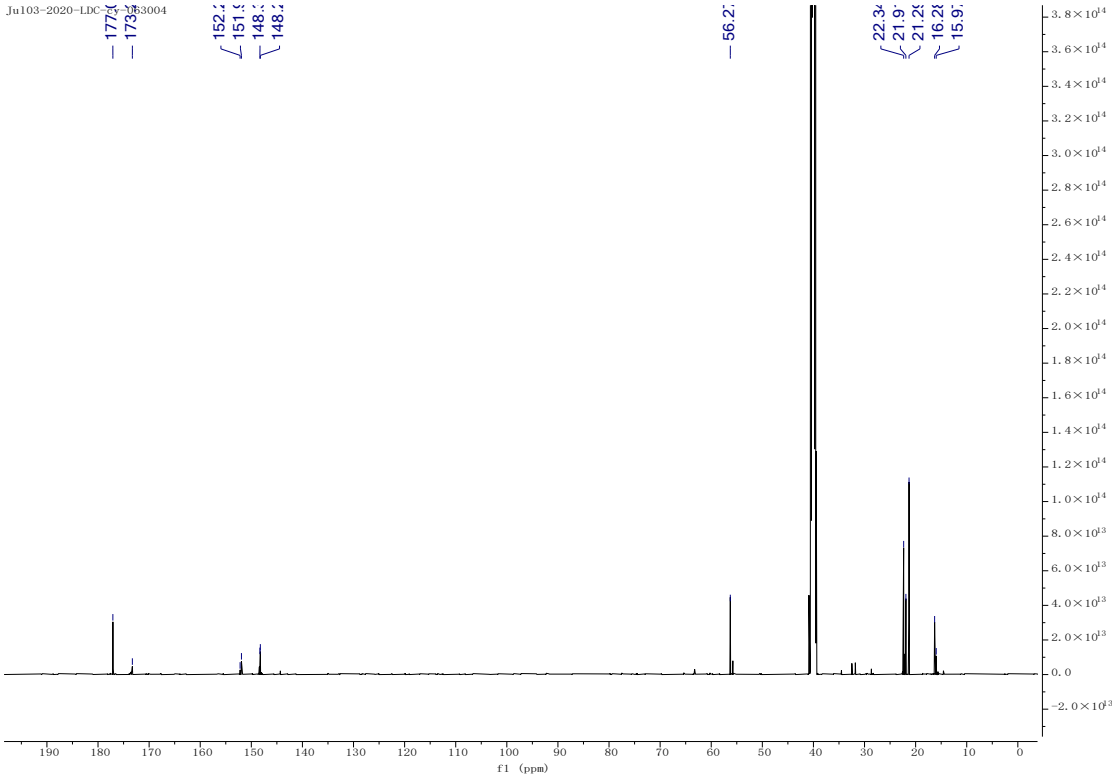


Figure S15 ^{13}C NMR spectrum for complex **3**.

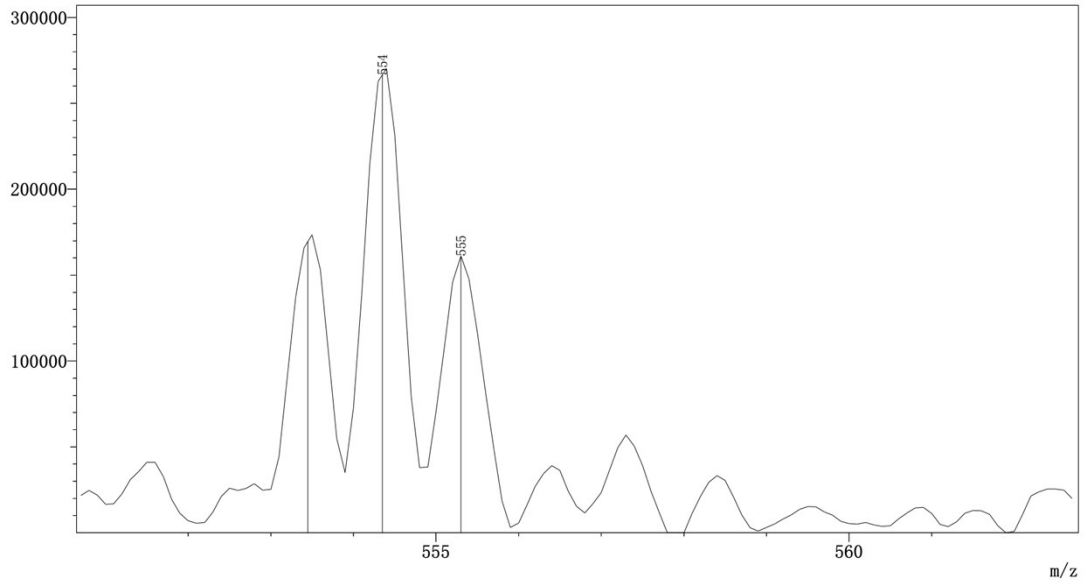


Figure S16 ESI-MS spectrum for complex **3**.

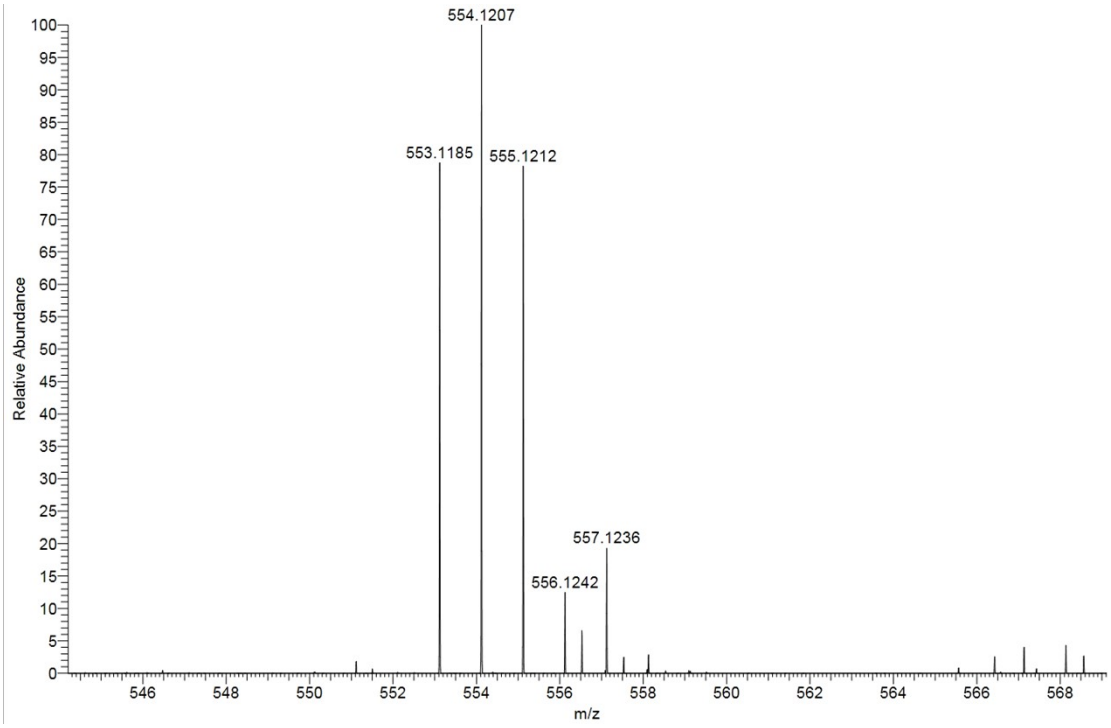


Figure S17 HRMS spectrum for complex **3**.

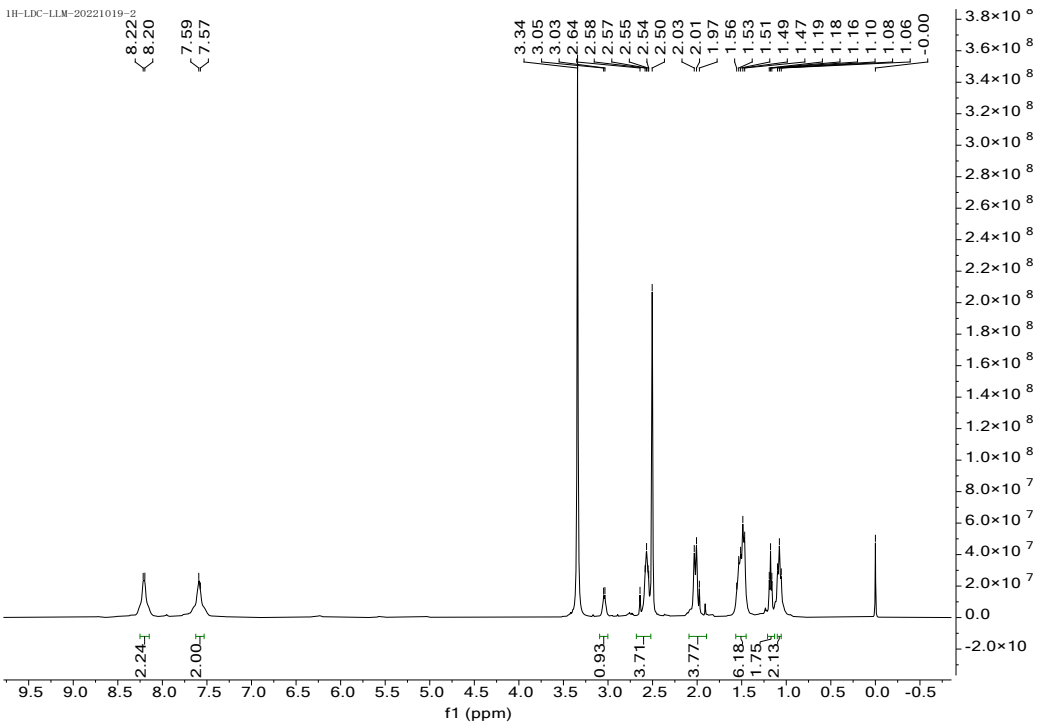


Figure S18 ^{1}H NMR spectrum for complex **4**.

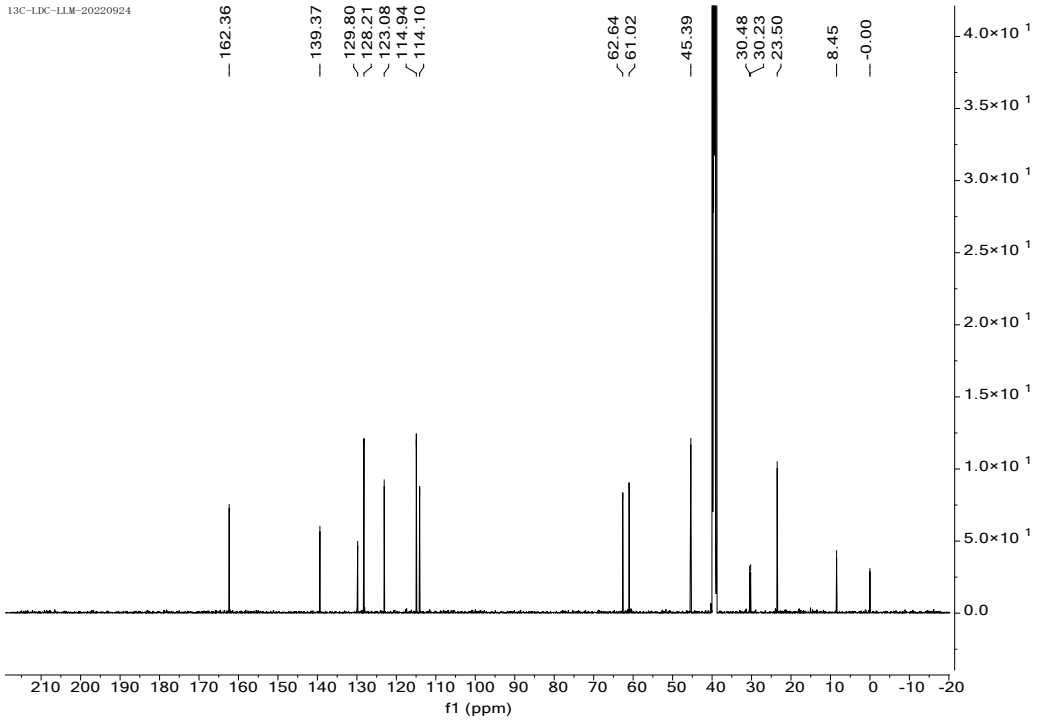


Figure S19 ^{13}C NMR spectrum for complex **4**.

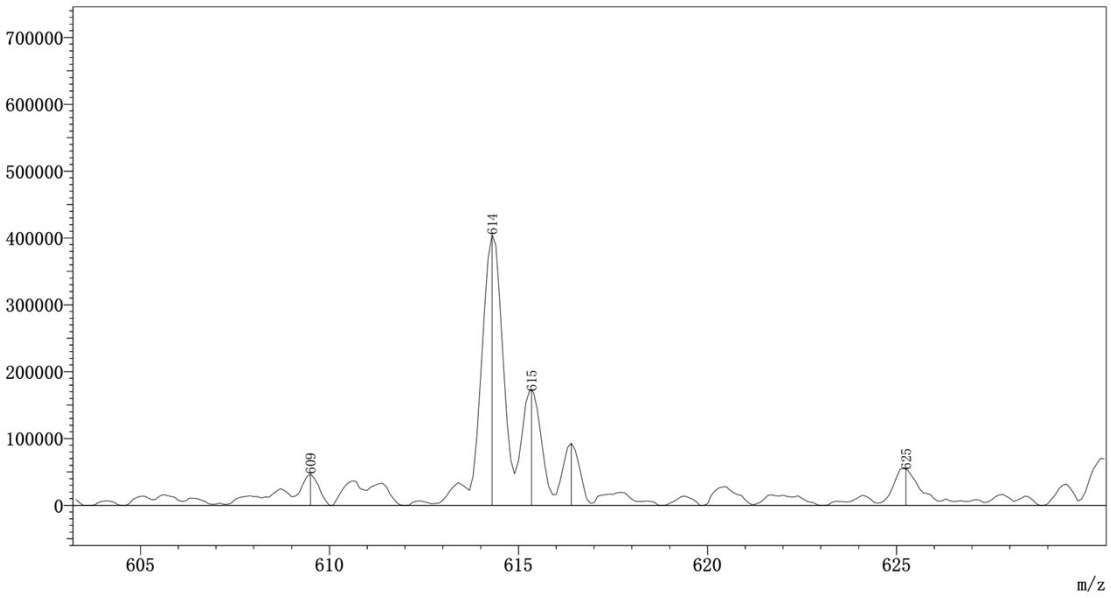


Figure S20 ESI-MS spectrum for complex **4**.

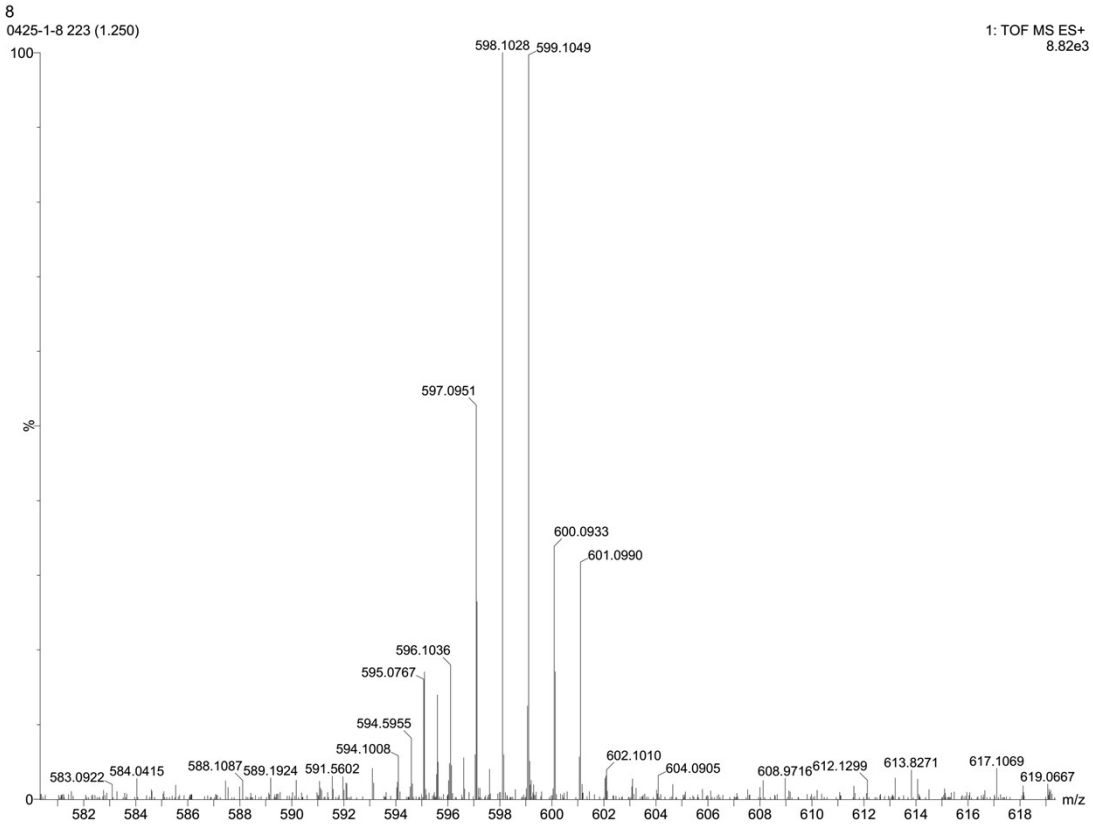


Figure S21 HRMS spectrum for complex **4**.

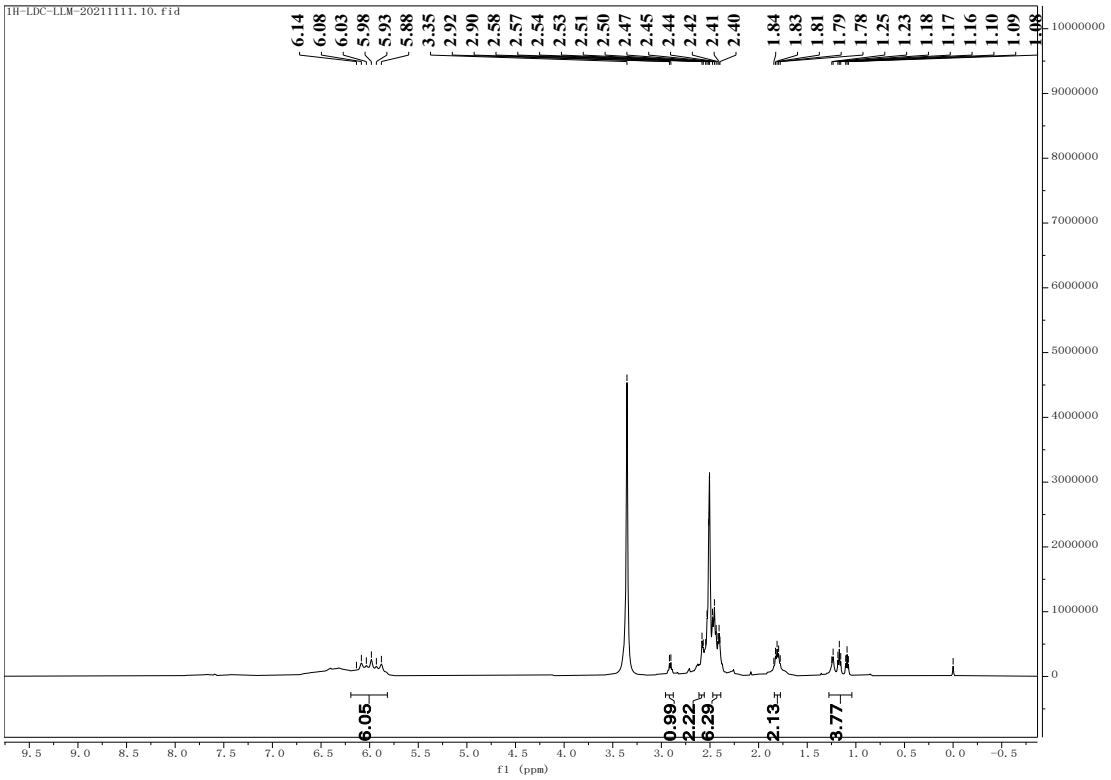


Figure S22 ^{1}H NMR spectrum for complex **5**.

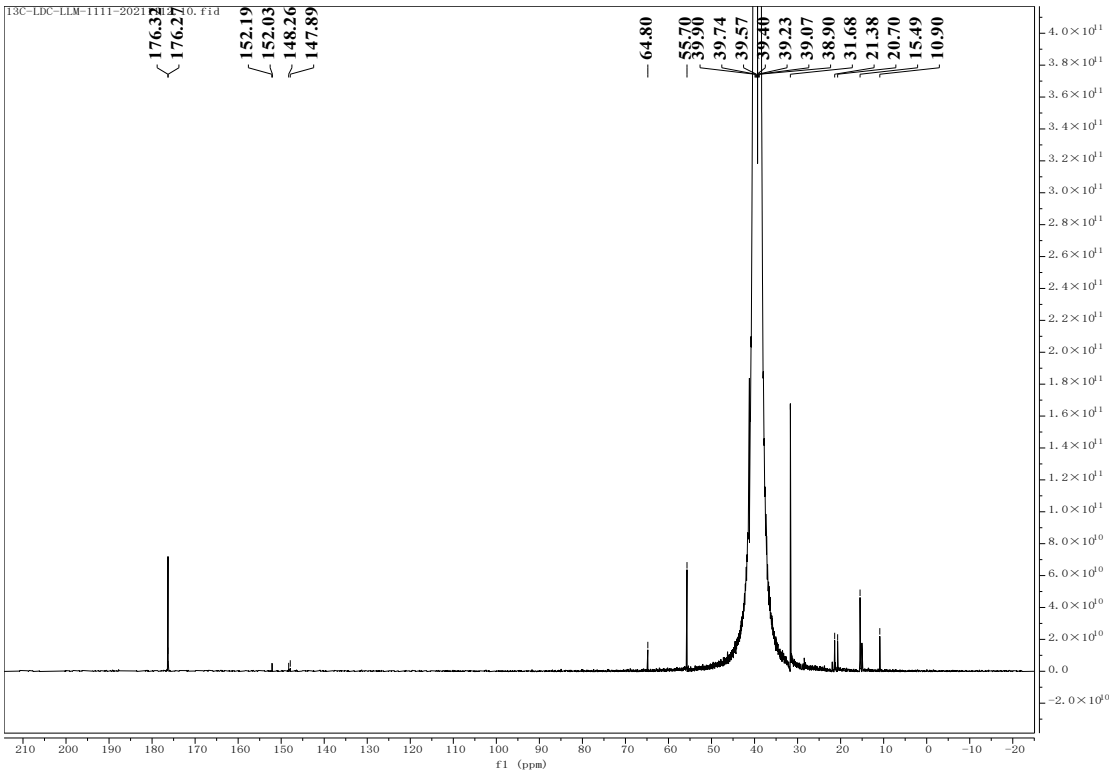


Figure S23 ^{13}C NMR spectrum for complex **5**.

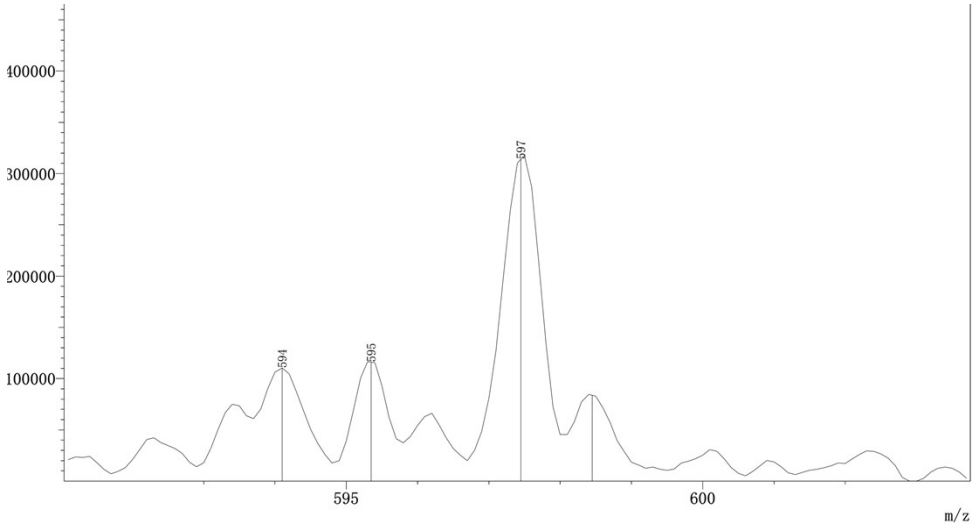


Figure S24 ESI-MS spectrum for complex **5**.

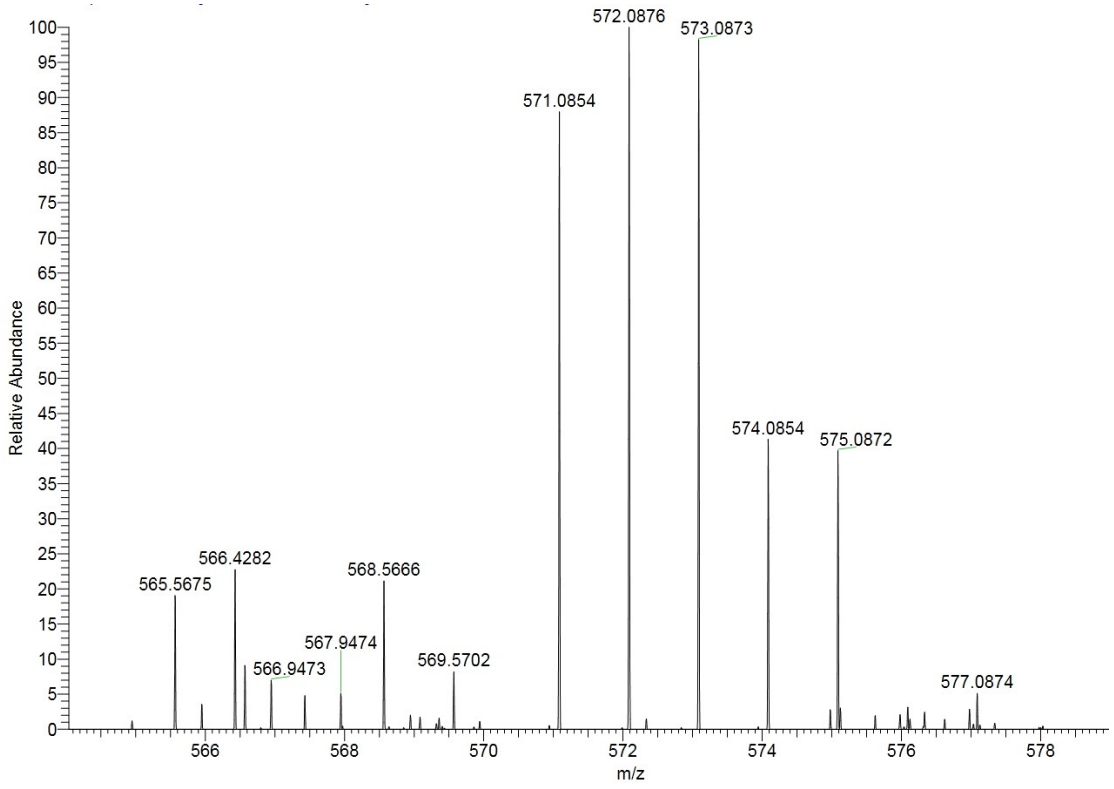
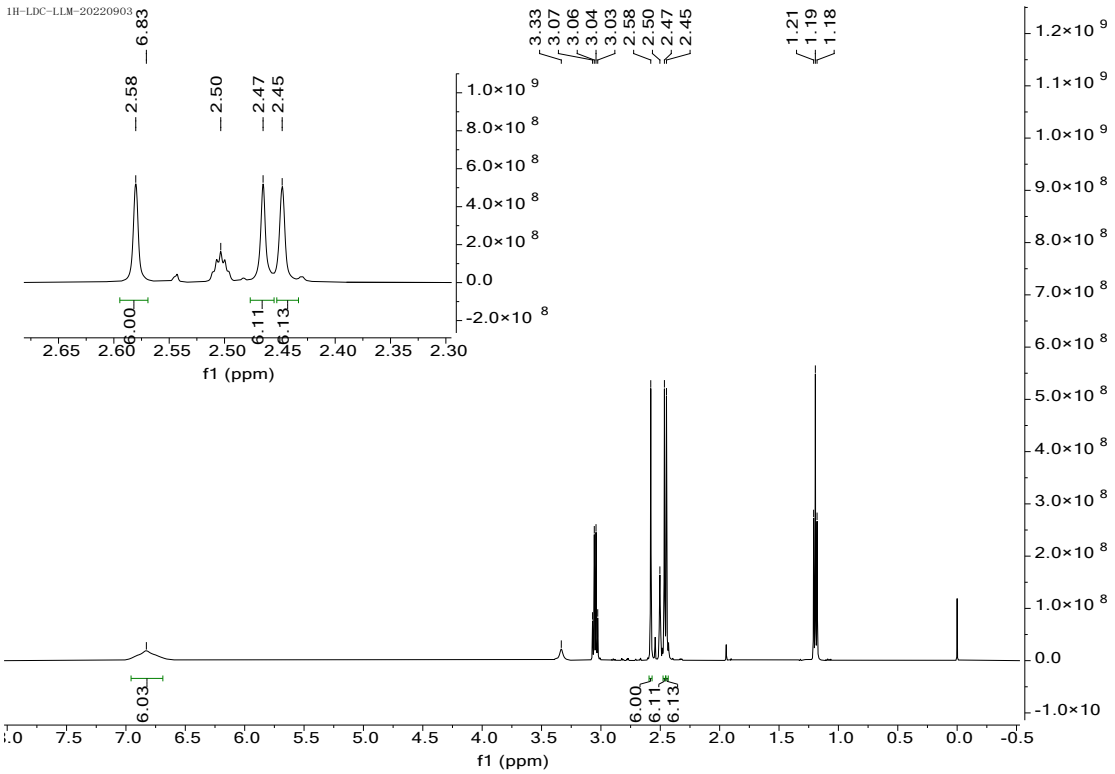
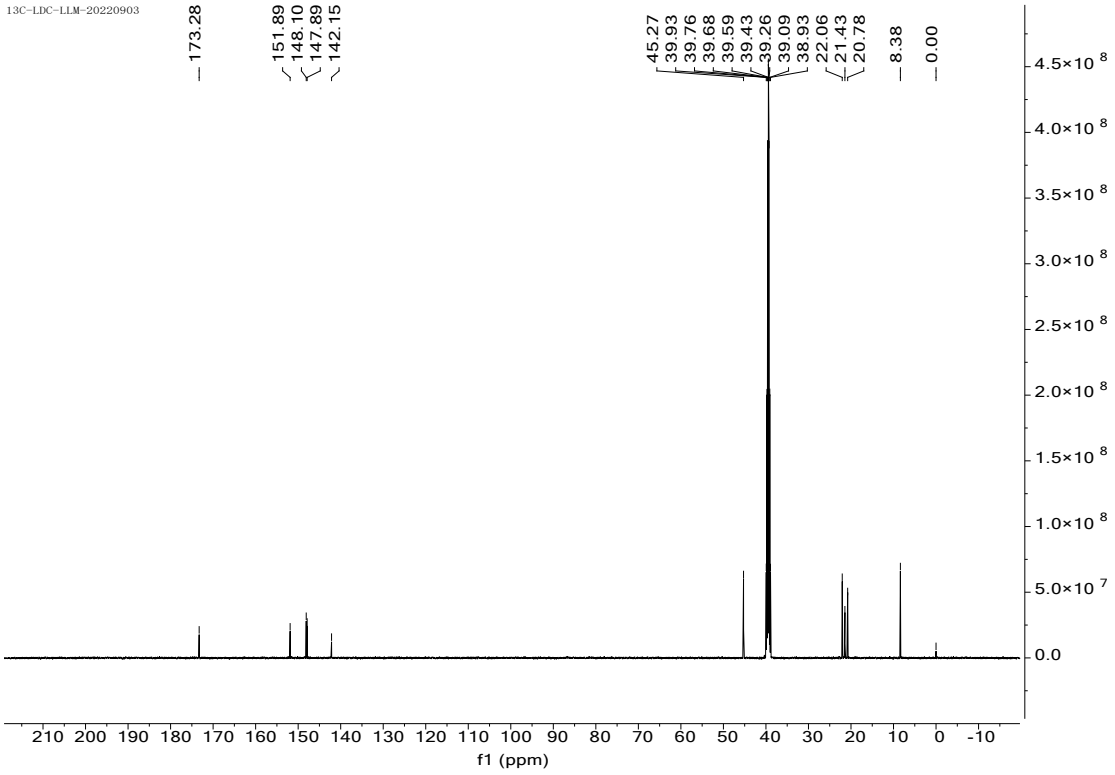


Figure S25 HRMS spectrum for complex **5**.



Peaks at 1.19 ppm (t) and 3.05 ppm (q) were ascribed to Et_2O.

Figure S26 ^{1}H NMR spectrum for complex **6**.



Peaks at 8.4 and 45.3 ppm were ascribed to Et_2O.

Figure S27 ^{13}C NMR spectrum for complex **6**.

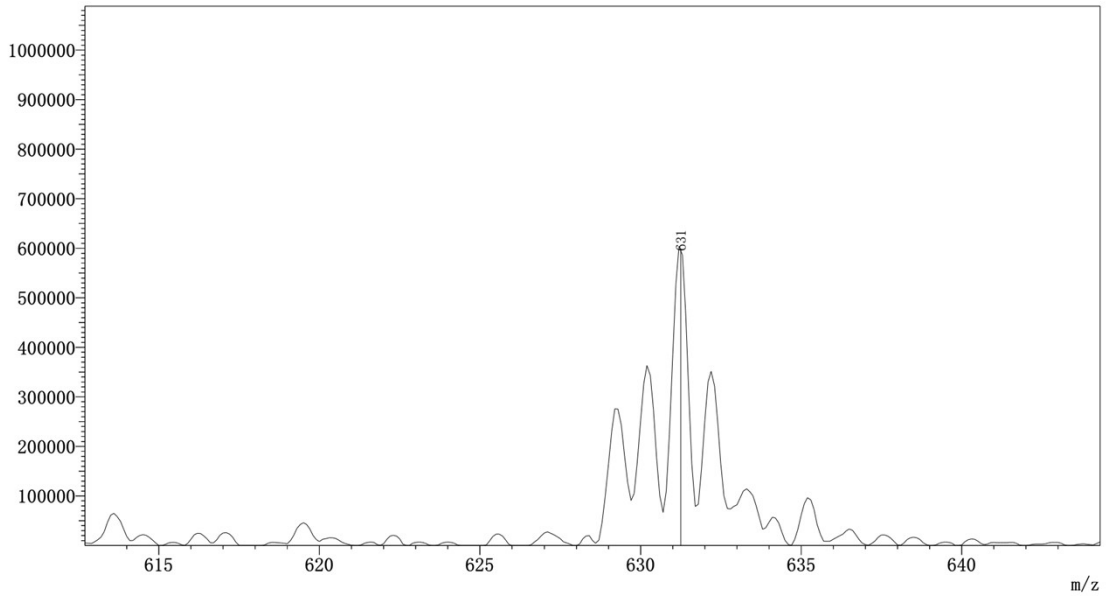


Figure S28 ESI-MS spectrum for complex **6**.

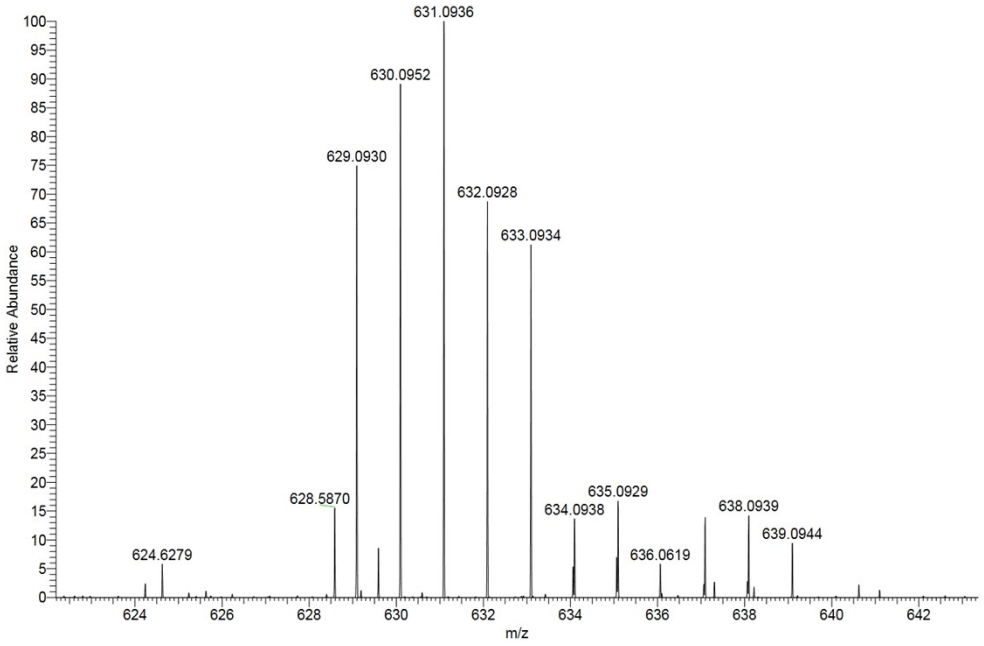


Figure S29 HRMS spectrum for complex **6**.

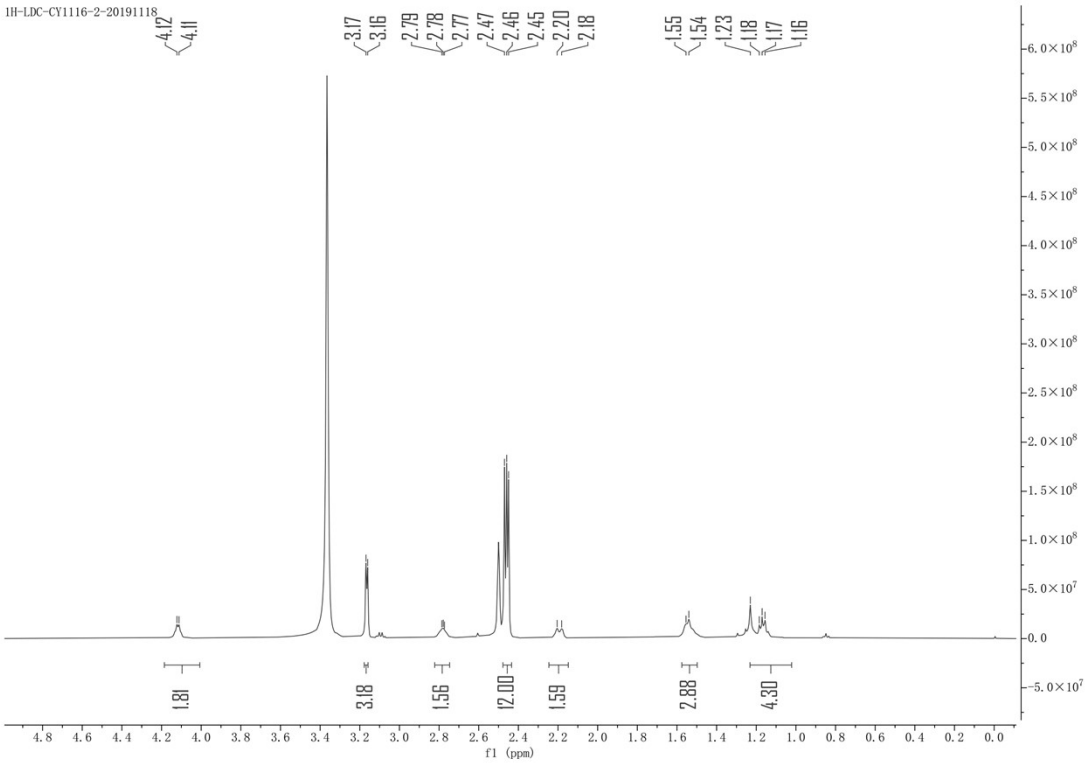


Figure S30 ^{1}H NMR spectrum for complex **7**.

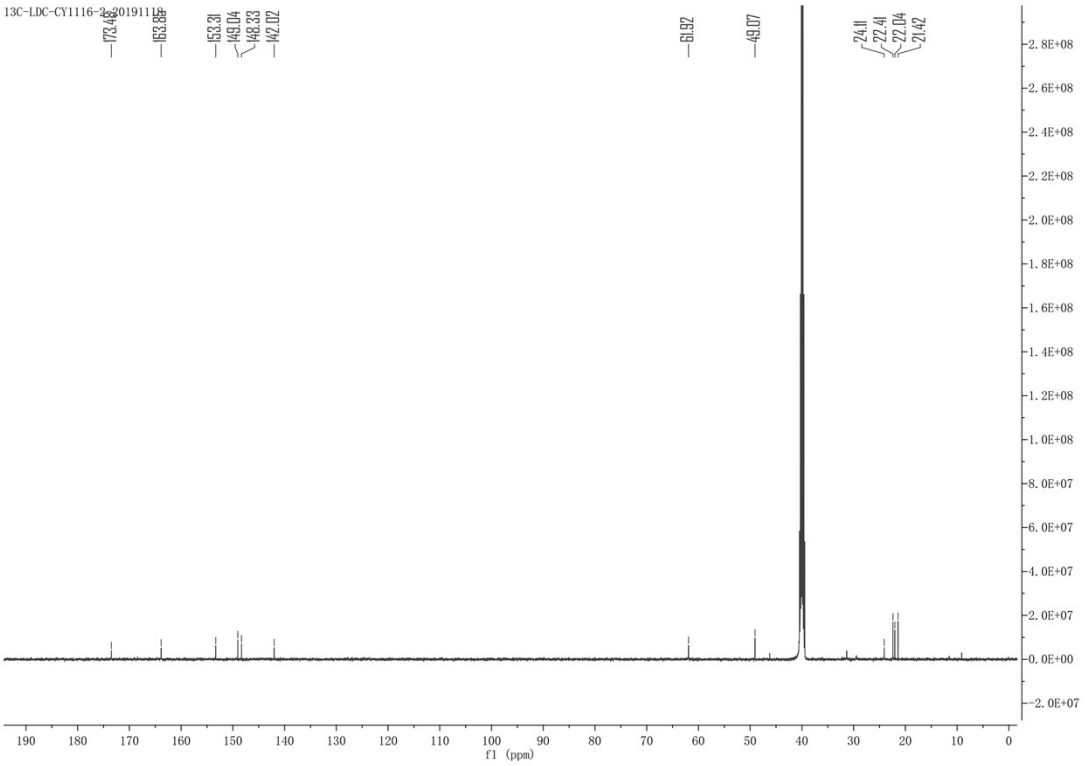


Figure S31 ^{13}C NMR spectrum for complex **7**.

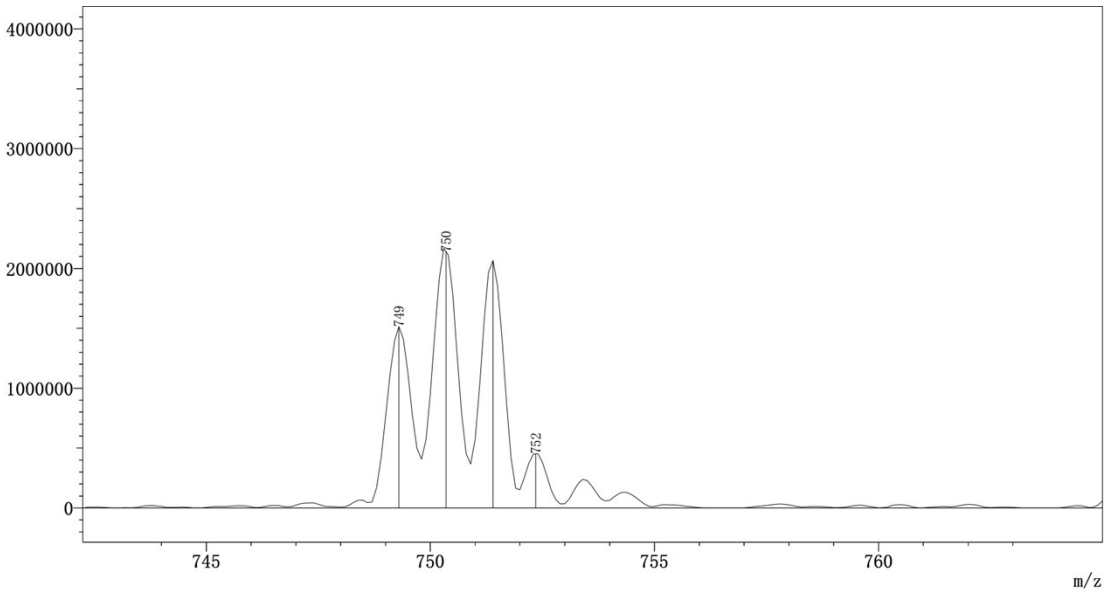


Figure S32 ESI-MS spectrum for complex **7**.

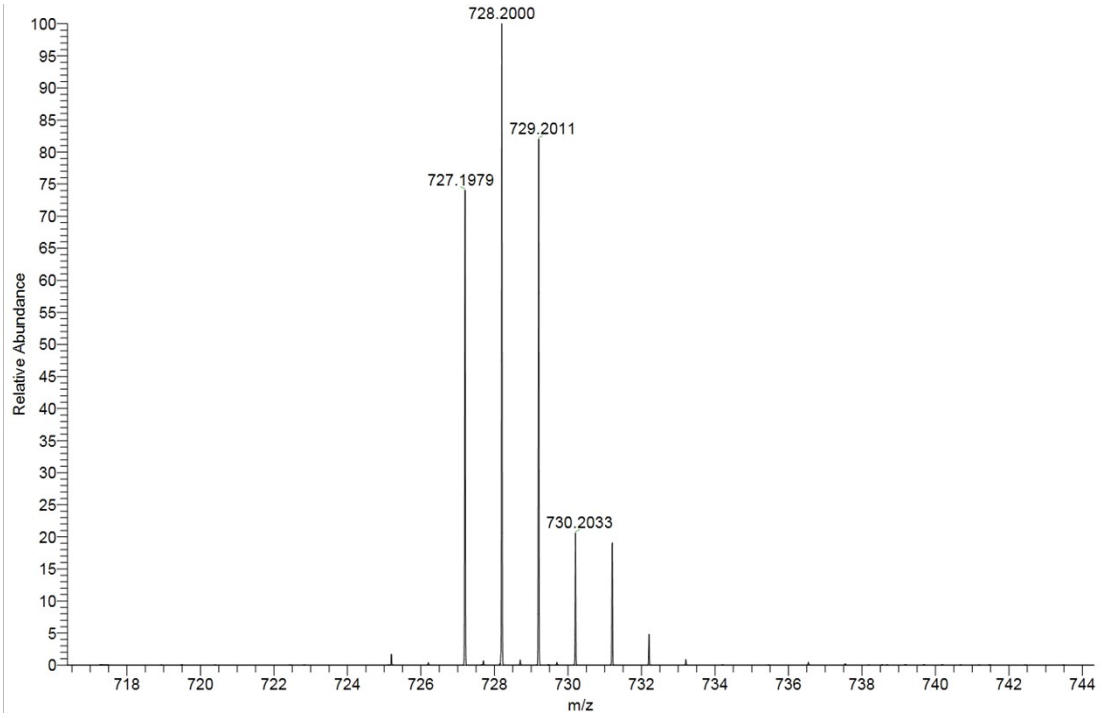
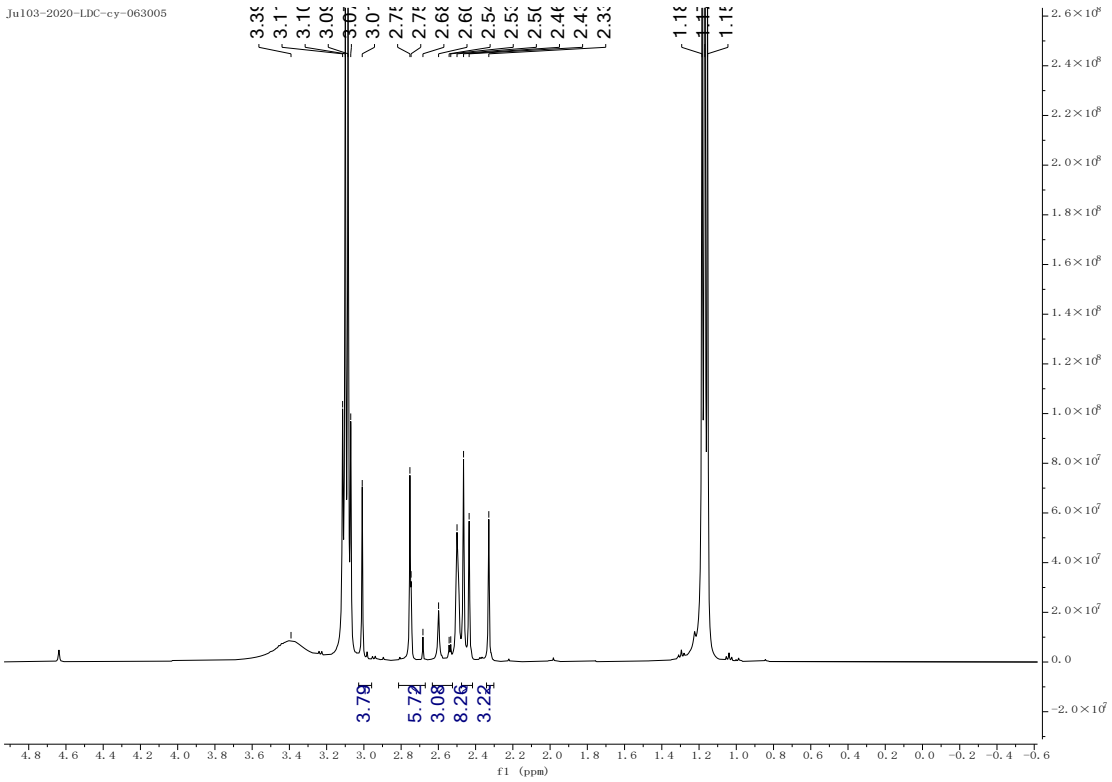
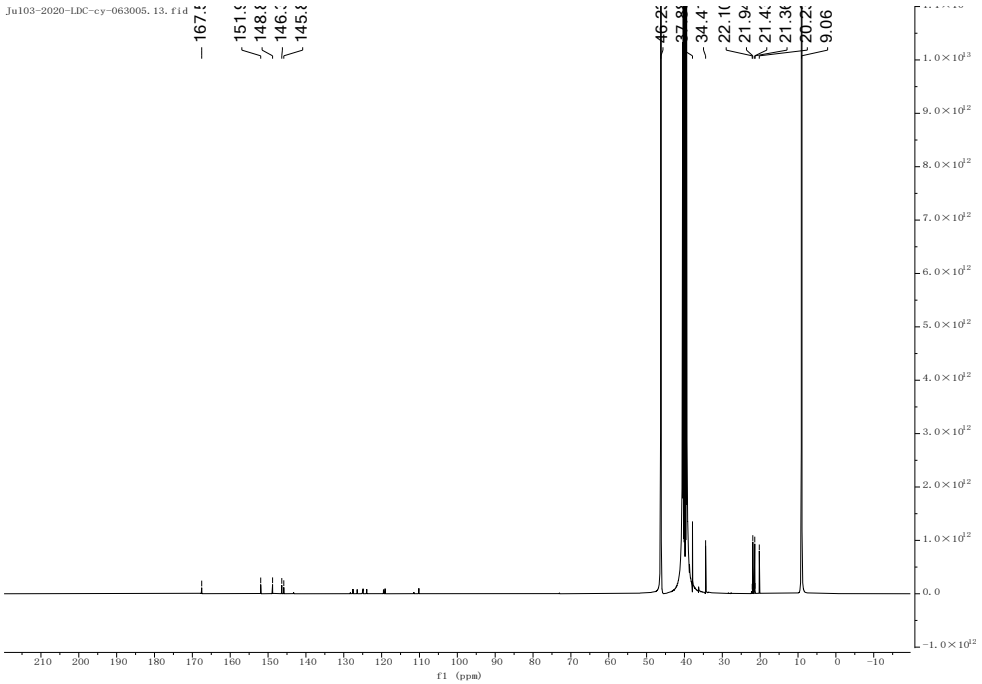


Figure S33 HRMS spectrum for complex **7**.



Peaks at 3.10 ppm (q) and 1.17 ppm (t) are ascribed to Et_2O.

Figure S34 ^{1}H NMR spectrum for complex **8**.



Peaks at 9.1 ppm and 46.2 ppm are ascribed to Et_2O.

Figure S35 ^{13}C NMR spectrum for complex **8**.

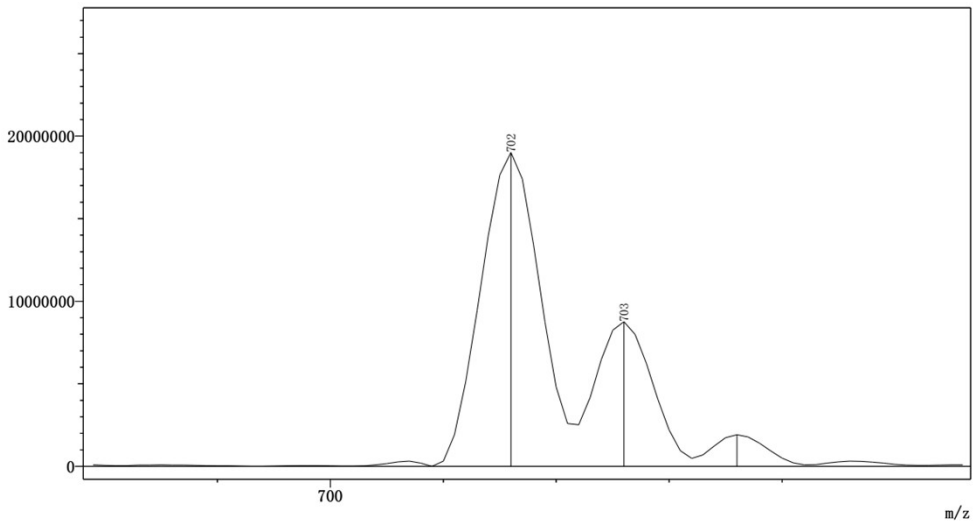


Figure S36 ESI-MS spectrum for complex **8**.

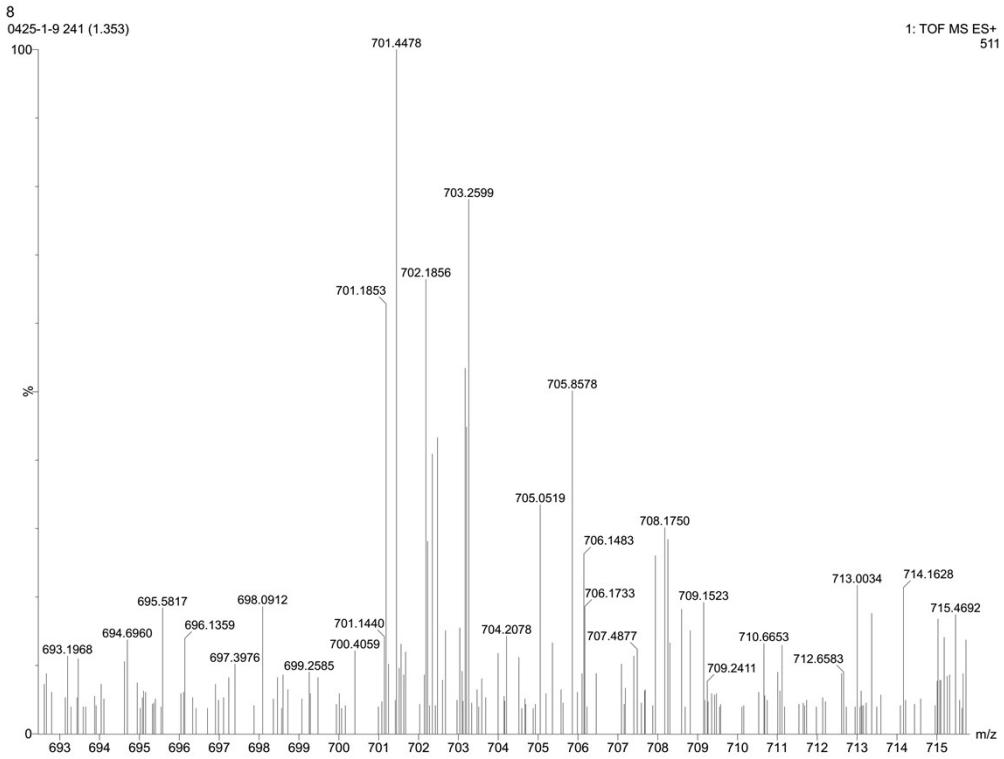


Figure S37 HRMS spectrum for complex **8**.

Reference

[S1] Q.P. Wang, Z.L. Huang, J. Ma, X.L. Lu, L. Zhang, X. Wang, P. G. Wang, *Dalton Trans*., 2016, 45, 10366–10374.