

Supporting Information for

Development of a series of flurbiprofen and zaltoprofen platinum(IV) complexes with anti-metastasis competence targeting COX-2, PD-L1 and DNA

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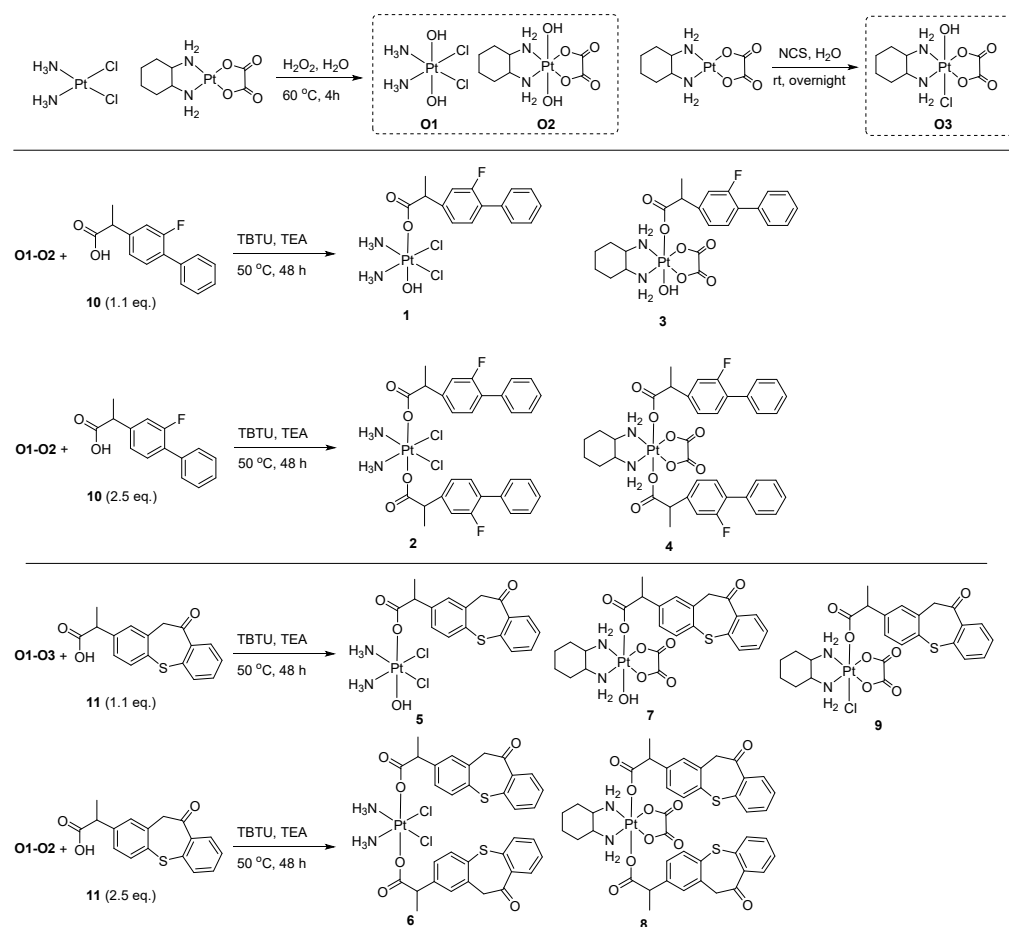
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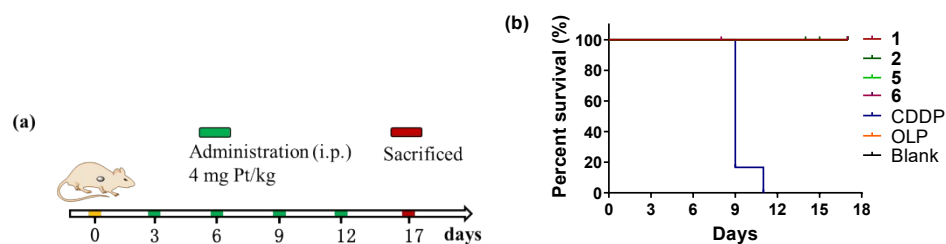
1. The synthetic route of flurbiprofen and zaltoprofen platinum(IV) complexes



Scheme S1 Synthetic route for flurbiprofen and zaltoprofen platinum(IV) complexes 1-9.

2. The antitumor activities *in vivo*

To compare the antitumor activities of NSAIDs platinum(IV) complexes extensively, platinum(IV) complexes 1-10 with ketoprofen and loxoprofen ligands listed in our previous work^{S1} (signed as J1-J10 in this manuscript) and complexes 1-9 bearing flurbiprofen, zaltoprofen in this work were tested at the same time both *in vitro* and *in vivo* with same control groups. The MTT results were given in Table 1 with cisplatin and oxaliplatin as reference drugs. Meanwhile, the antitumor activities for complexes 1, 2, 5, 6, J1, J7, J9, cisplatin and oxaliplatin *in vivo* against CT26 tumors were shown in Figure S1 and Figure 3, the antitumor activities *in vivo* against 4T1 tumors were shown in Figure S2 and Figure 4, and the anti-metastasis properties against 4T1 breast carcinoma tumors *in vivo* were shown in Figure S6 and Figure 5.



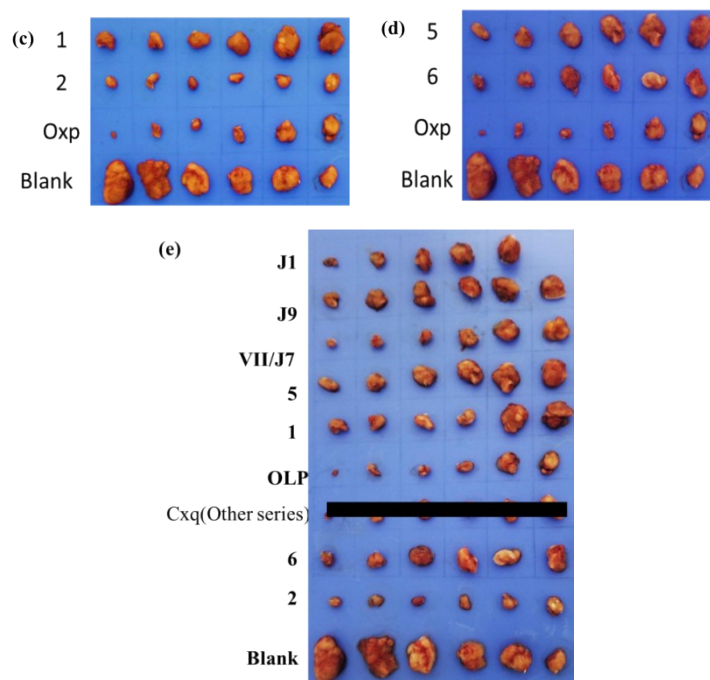


Figure S1 *In vivo* antitumor activities to CT-26 tumors in BALB/c mice ($n = 6$). (a) Schematic illustration of the experimental design. (b) Survival analysis of mice during the treatment of **1**, **2**, **5**, **6**, cisplatin and oxaliplatin. (c) Images of tumors treated by **1**, **2**, oxaliplatin and blank. (d) Images of tumors treated by **5**, **6**, oxaliplatin and blank. Figure 3d is a combination of images (c) and (d). (e) Full images of tumors treated by **1**, **2**, **5**, **6**, **J1**, **J7 (VII)**, **J9** and oxaliplatin and blank. Reprinted with permission from Li et al., *J. Med. Chem.*, 2021, **64**, 17920. Copyright 2021 American Chemical Society ^{S1}.

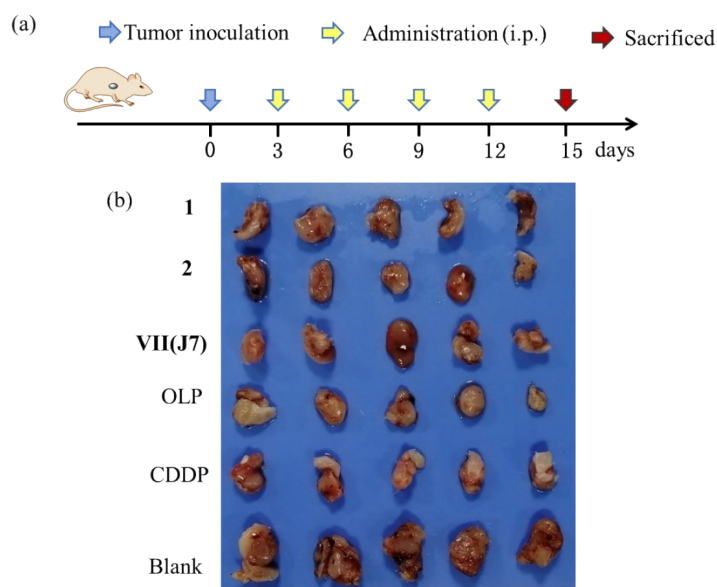


Figure S2 *In vivo* antitumor activities to 4T1 tumors in BALB/c mice ($n = 5$). (a) Schematic illustration of the experimental design. (b) Full images of tumors treated by drugs **1**, **2**, **J7 (VII)**, cisplatin and oxaliplatin and blank. Reprinted with permission from Li et al., *J. Med. Chem.*, 2021, **64**, 17920. Copyright 2021 American Chemical Society ^{S1}.

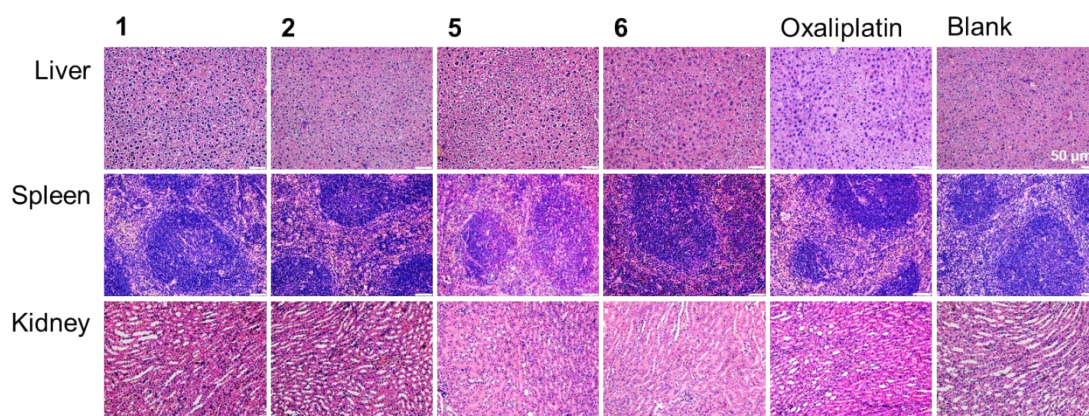


Figure S3 H&E staining of liver, spleen and kidney from mice treated by complexes **1**, **2**, **5**, **6** and oxaliplatin and saline.

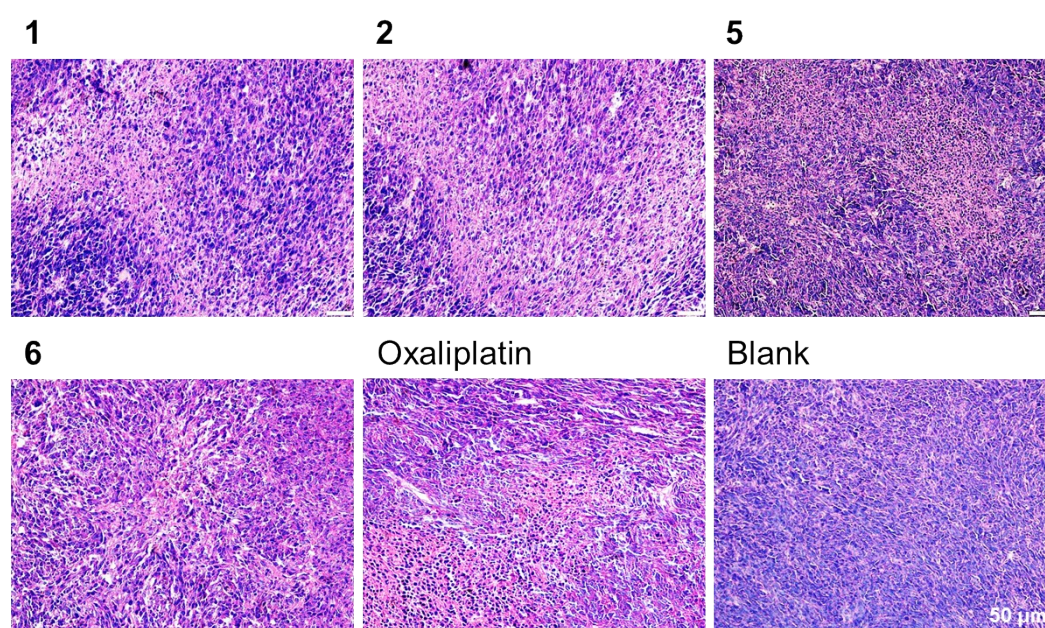


Figure S4 H&E staining of tumor tissues from mice treated by complexes **1**, **2**, **5**, **6** and oxaliplatin and saline.

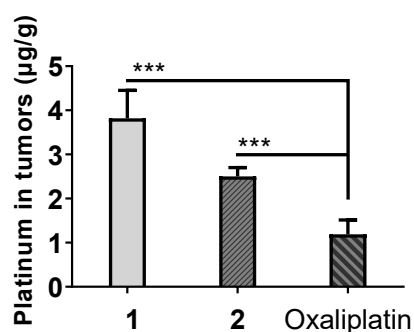


Figure S5 Platinum accumulation in tumors tissues treated by complexes **1**, **2** and oxaliplatin.

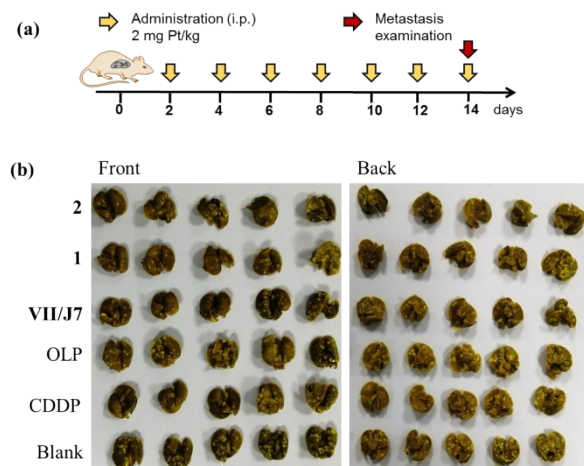


Figure S6 Pulmonary metastasis inhibition of compounds **1**, **2**, **VII(J7)** cisplatin and oxaliplatin against 4T1 breast carcinoma tumors *in vivo* (n = 5). (a) Schematic illustration of the experimental design. (b) Full images of lungs from each group at the end of the experiment. Reprinted with permission from Li et al., *J. Med. Chem.*, 2021, **64**, 17920. Copyright 2021 American Chemical Society^{S1}.

3. Metastasis inhibitory activities *in vitro*

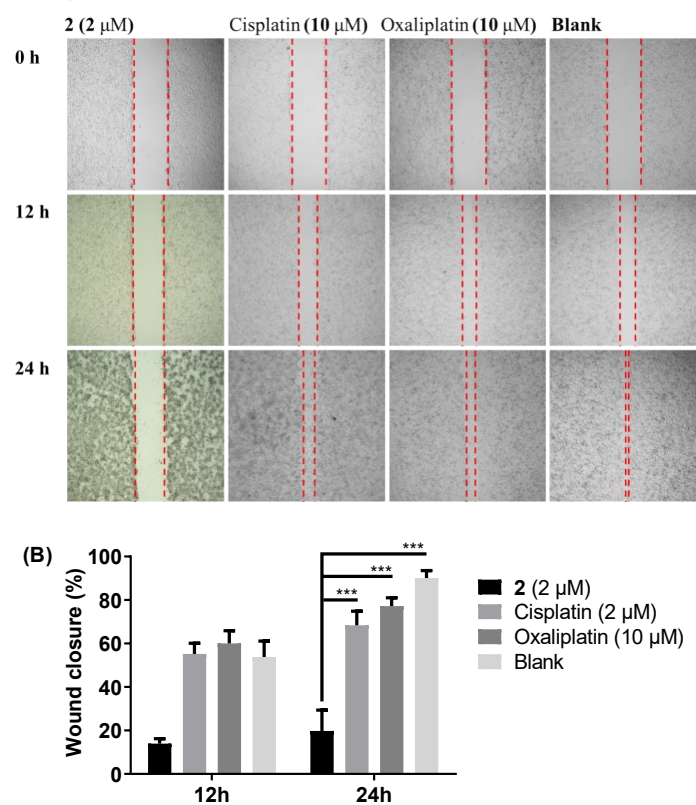


Figure S7 Migration inhibition to 4T1 cells of complex **2** (2 μM), cisplatin (10 μM) and oxaliplatin (10 μM) *in vitro*. The untreated group was set as blank. The extent of wound healing was observed at 0, 12, and 24 h. (a) Representative images; (b) Analysis of wound closure.

4. The reduction potential and DNA binding properties

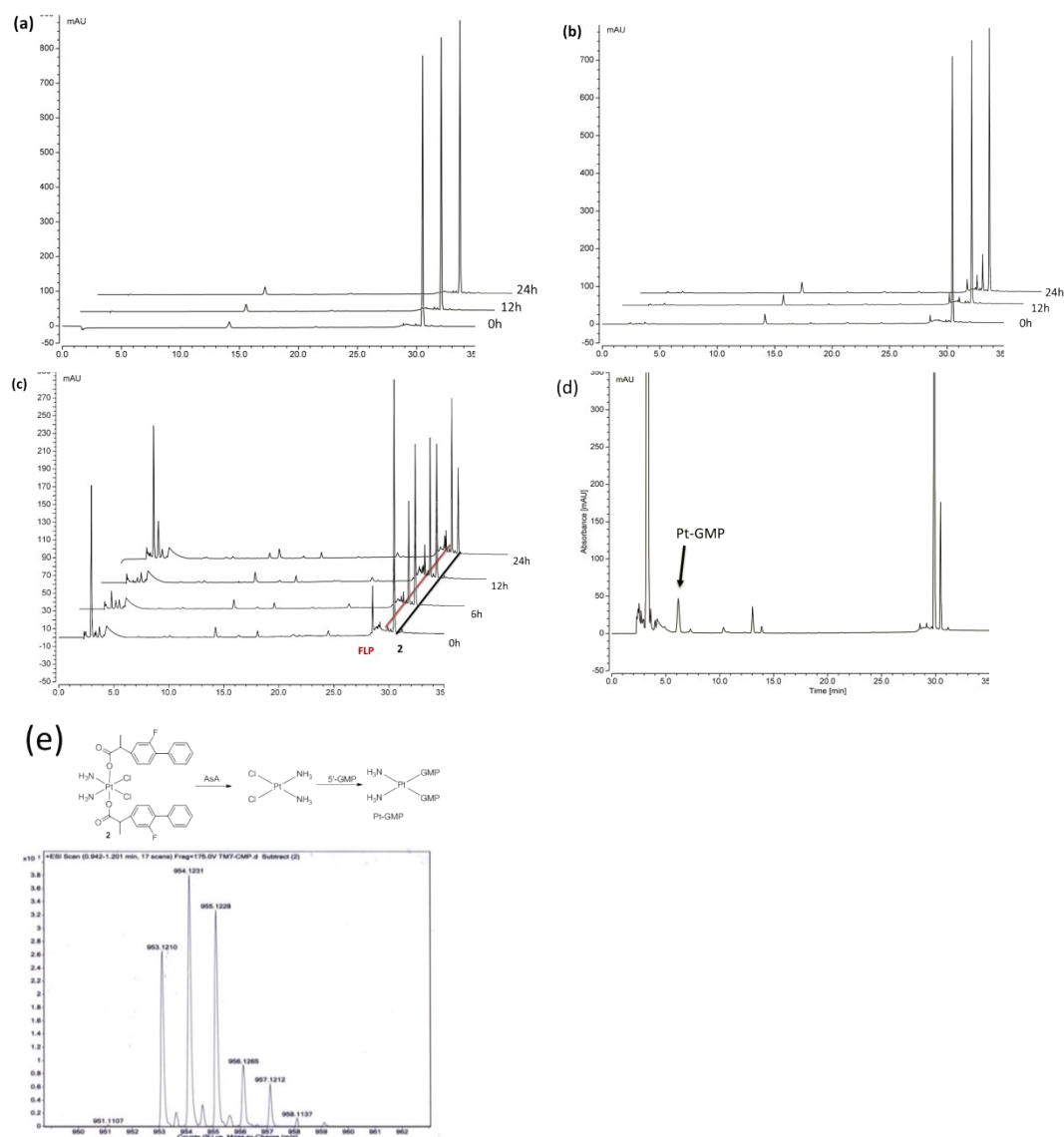


Figure S8 HPLC spectra of compound **2** in different media incubated at 37 °C. (a) Compound **2** (0.5 mM) in PBS; (b) Compound **2** (0.5 mM) in RPMI 1640; (c) Compound **2** (0.25 mM) in RPMI 1640 with AsA (1 mM); (d) Solution of compound **2** (0.25 mM) in RPMI 1640 with AsA (1 mM) and 5'-GMP (3 mM) incubated for 24 h; (e) The formation of platinated GMP (Pt-GMP) ^{S2}.

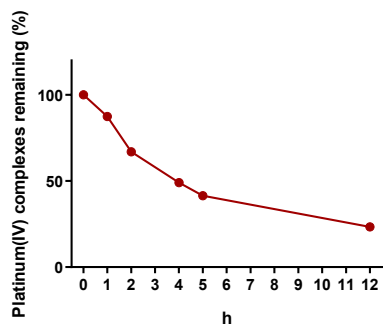
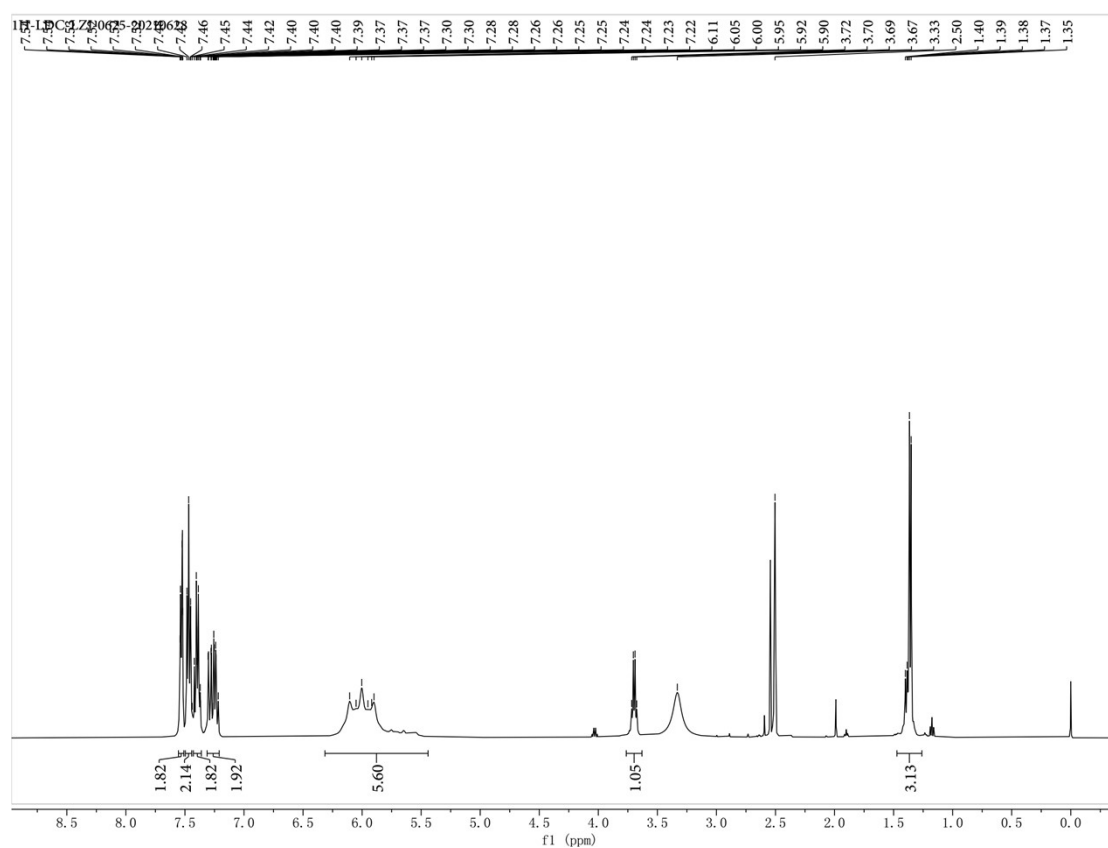
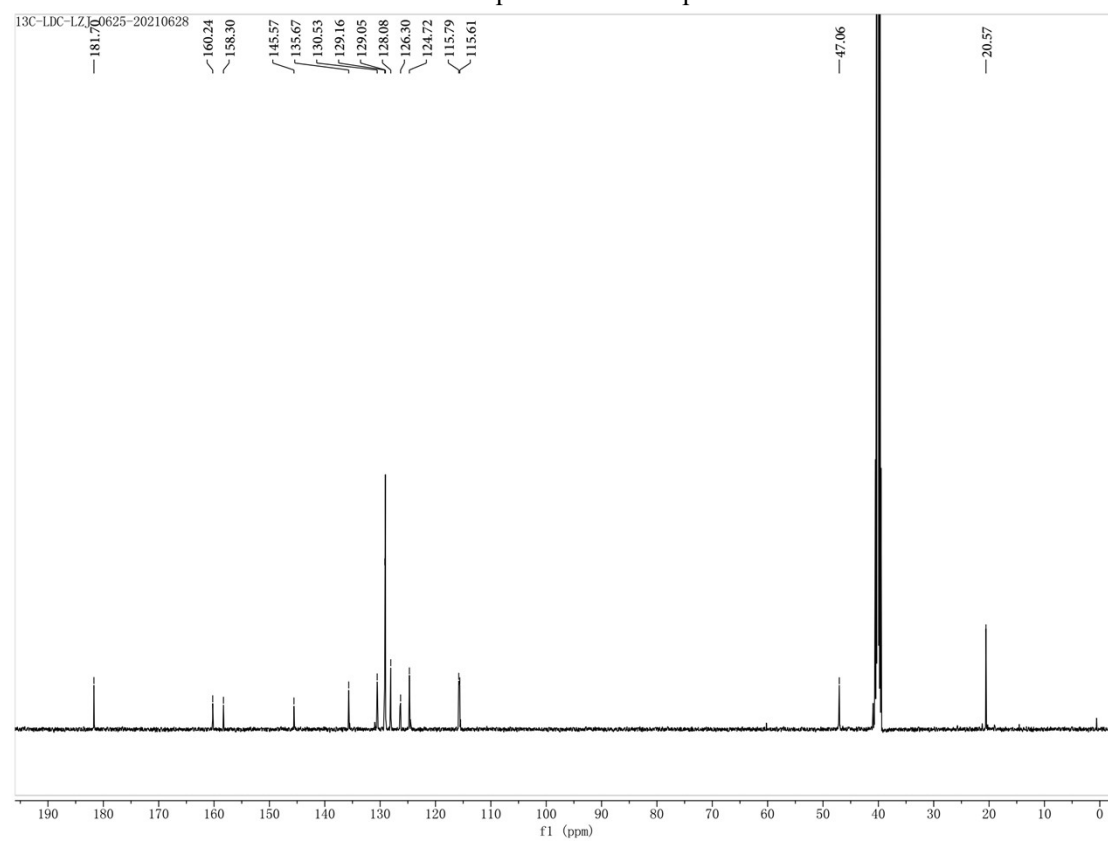


Figure S9 Stability of complex **2** in whole blood incubated at 37 °C.

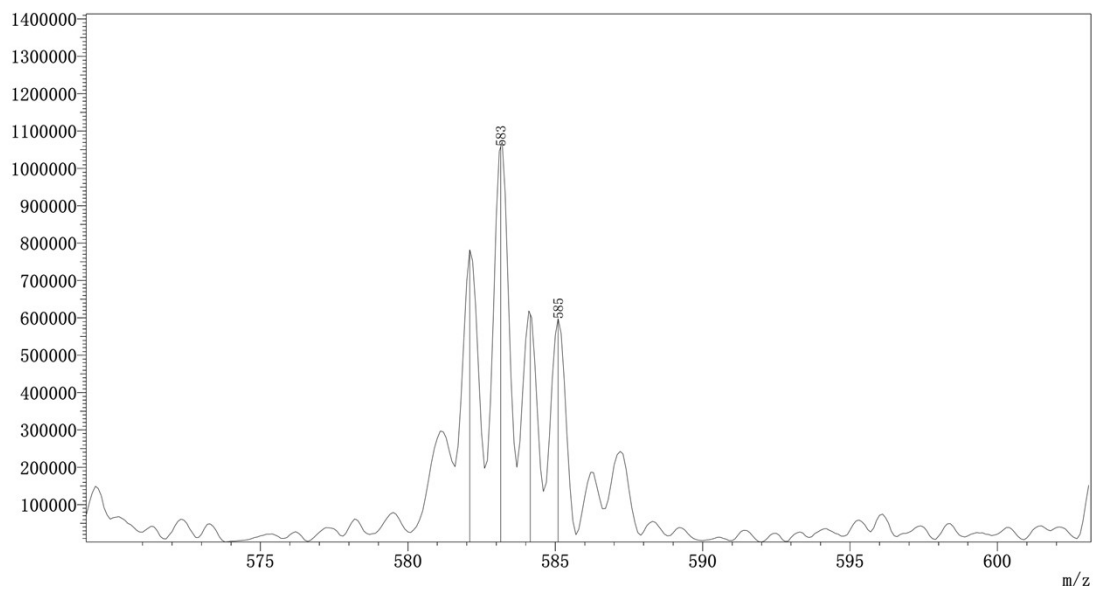
5. ^1H NMR, ^{13}C NMR and MS spectra



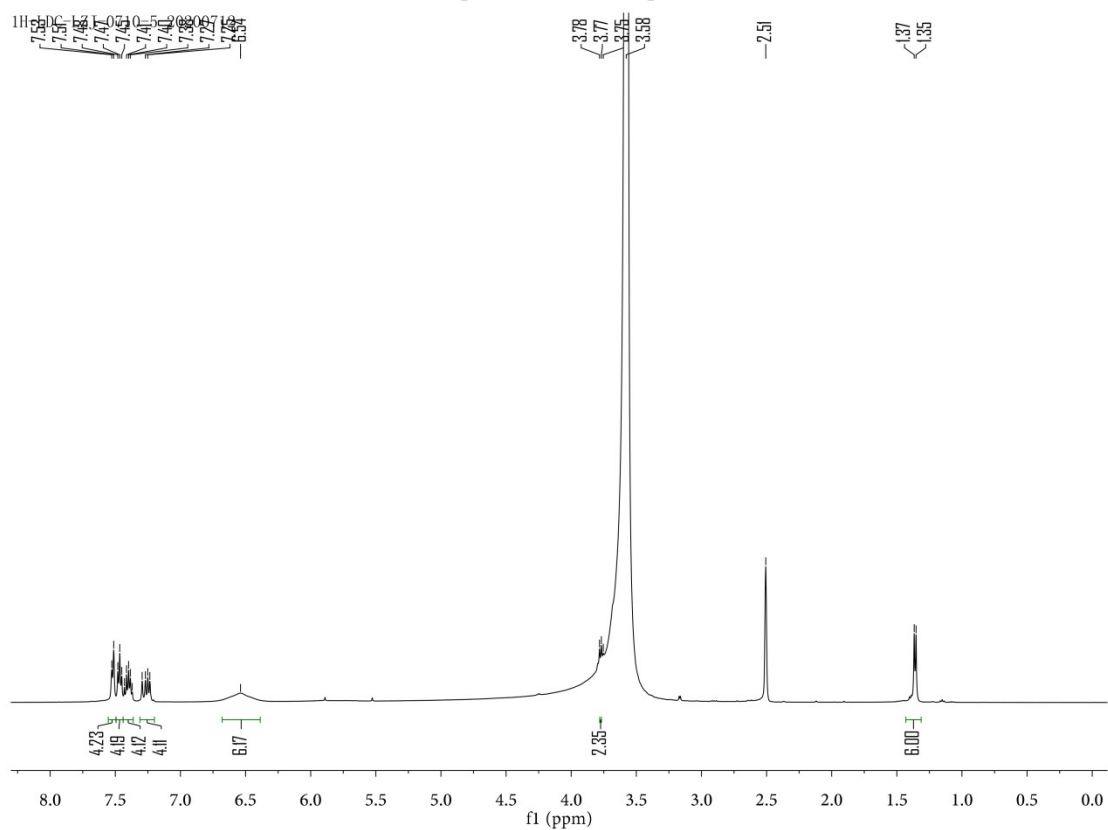
^1H NMR spectrum for complex 1



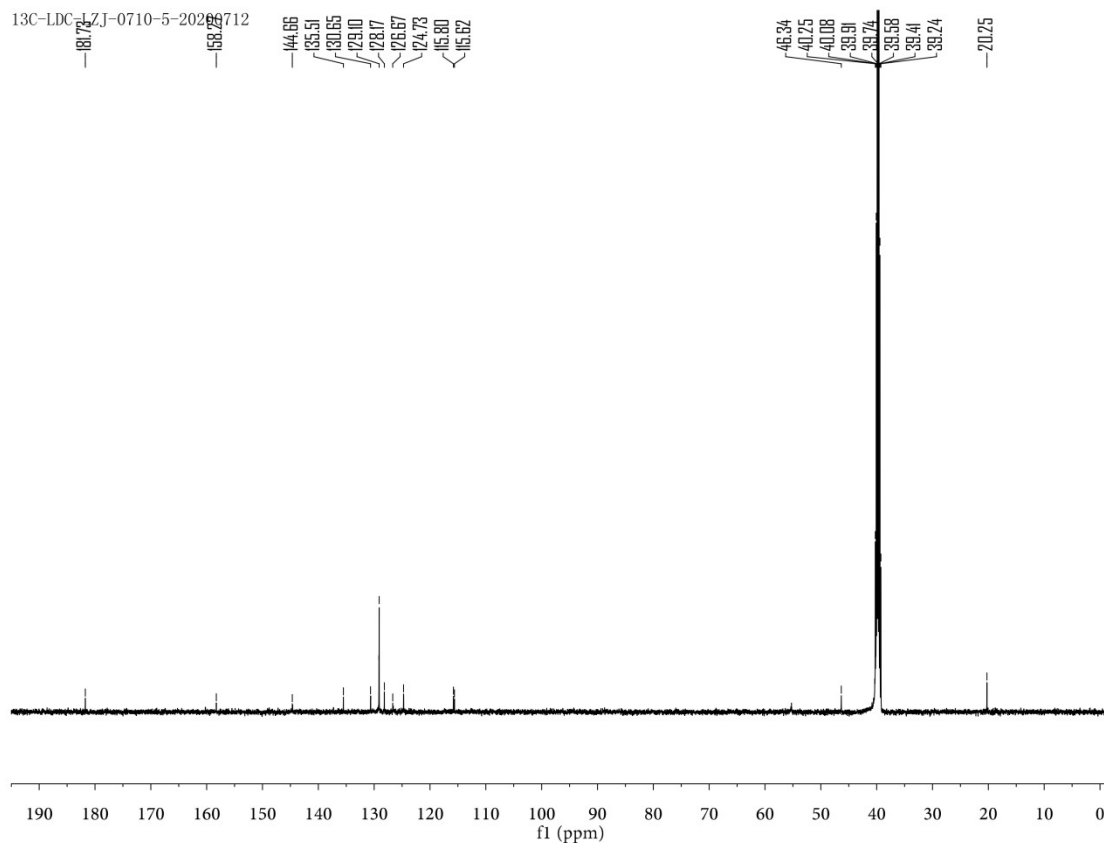
^{13}C NMR spectrum for complex 1



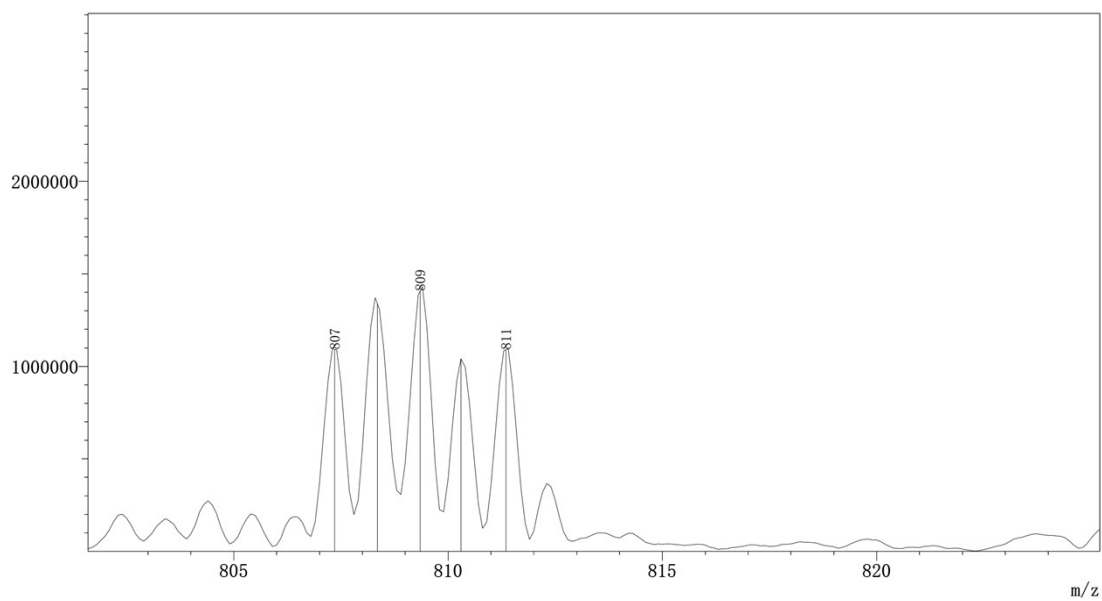
MS spectrum for complex 1



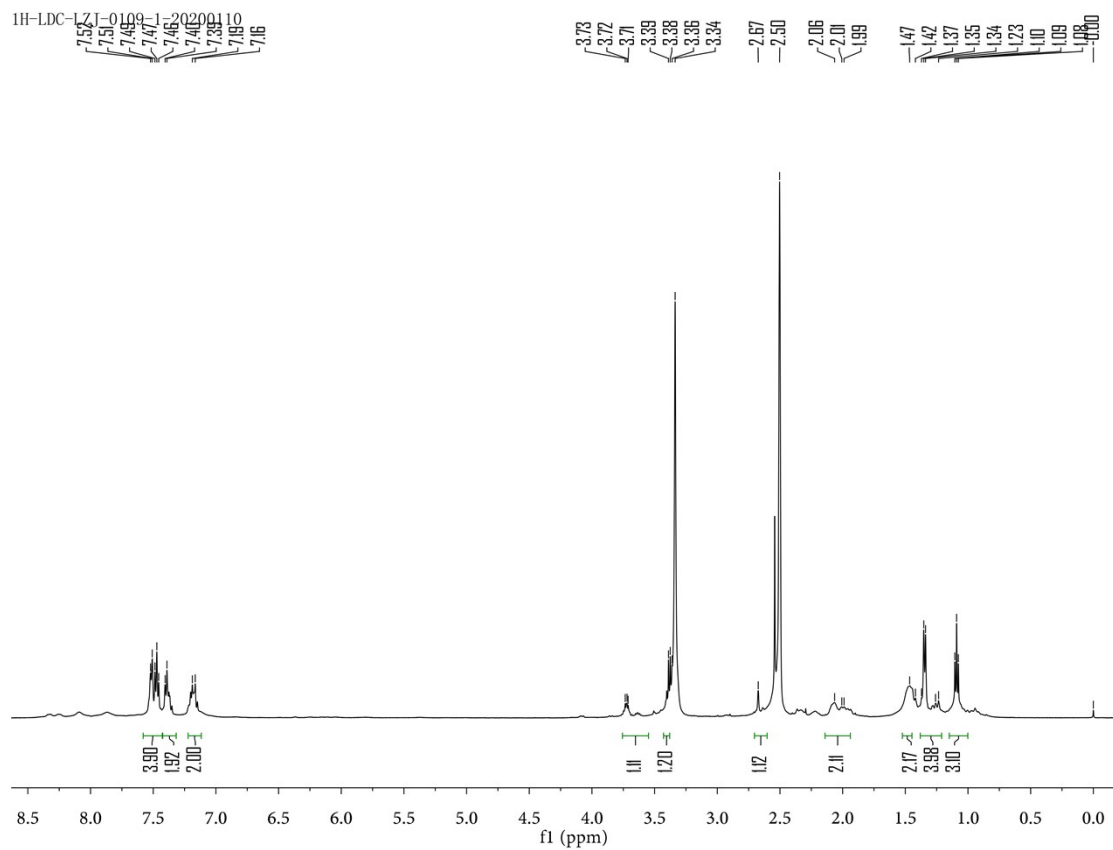
¹H NMR spectrum for complex 2



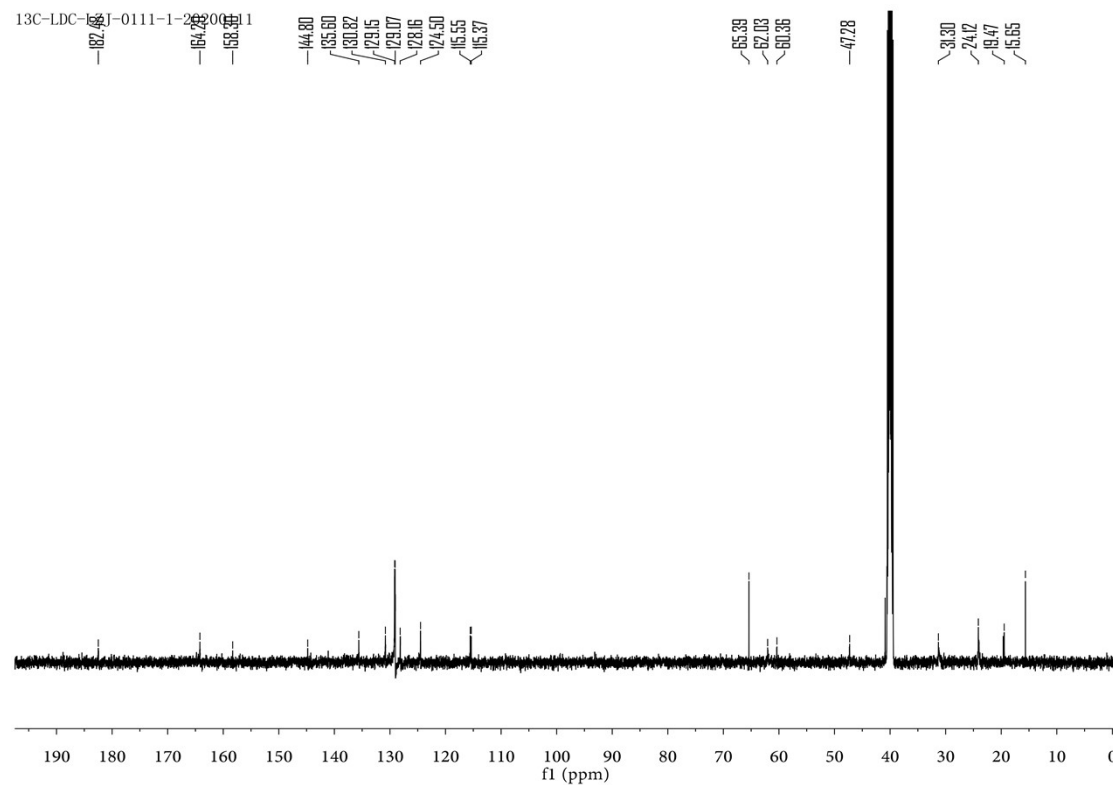
¹³C NMR spectrum for complex 2



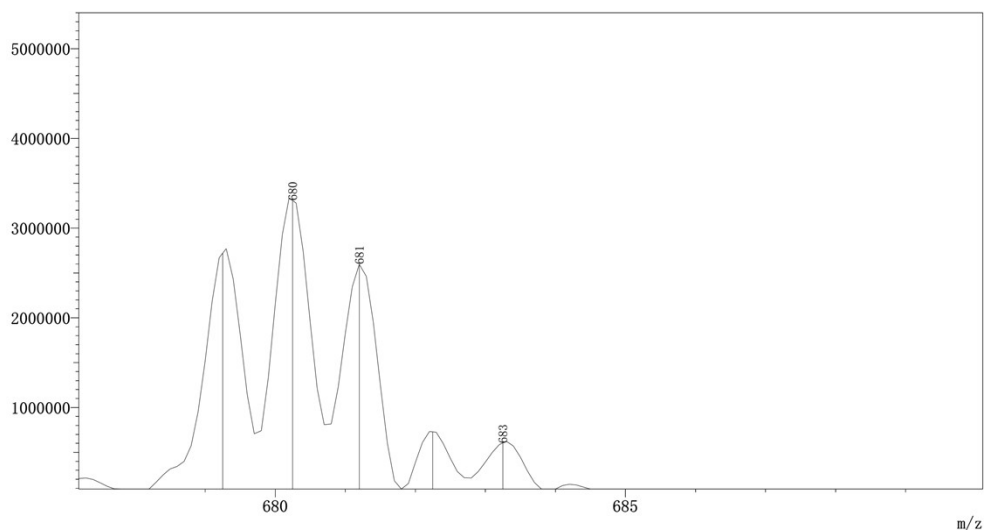
MS spectrum for complex 2



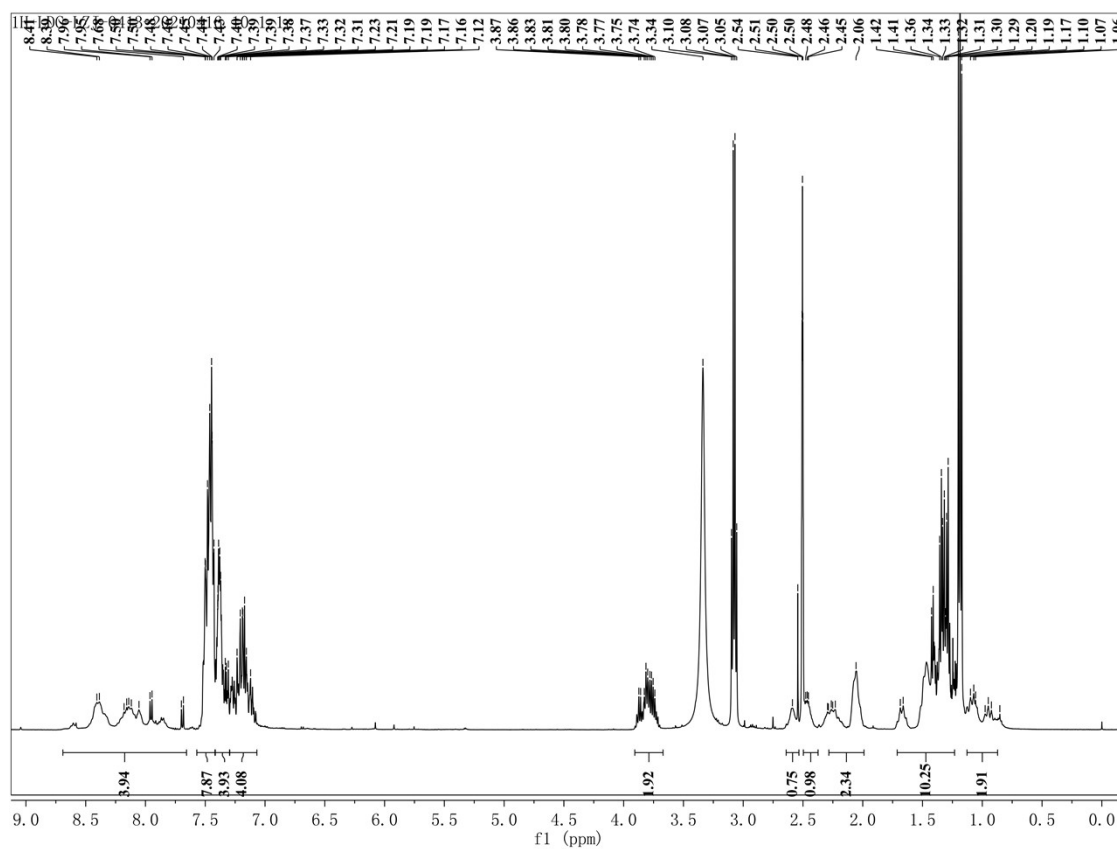
¹H NMR spectrum for complex **3**



¹³C NMR spectrum for complex **3**

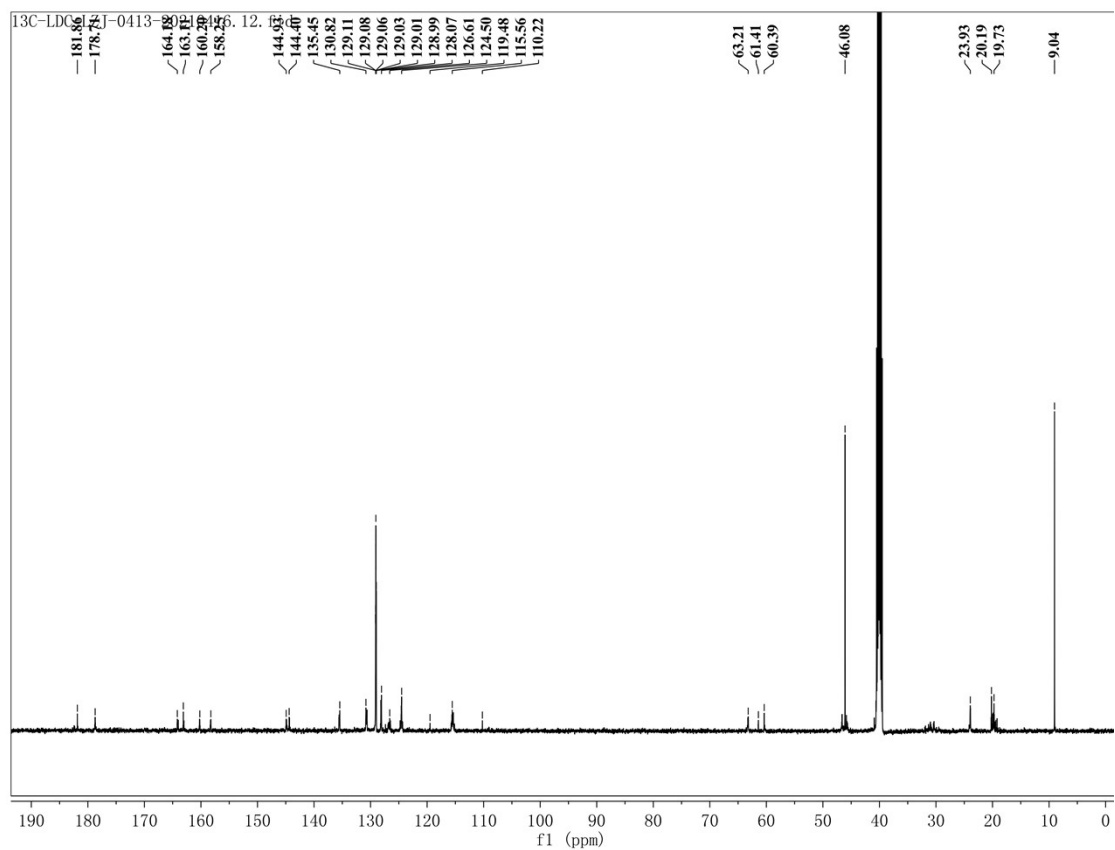


MS spectrum for complex 3



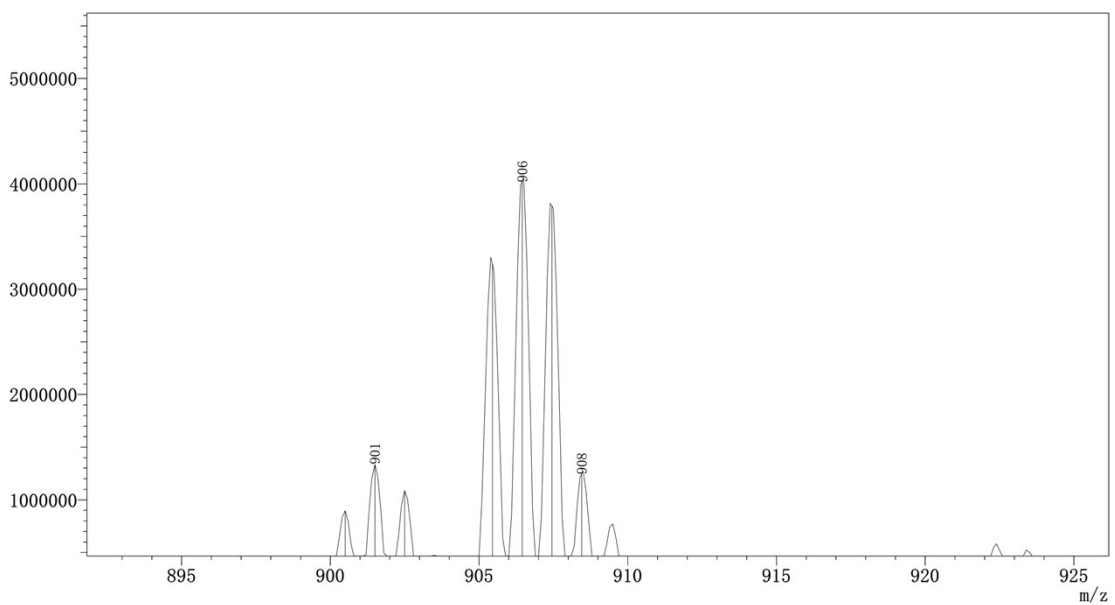
* Peaks 3.07 ppm (q) and 1.19 ppm (t) are ascribed to Et₂O.

¹H NMR spectrum for complex 4

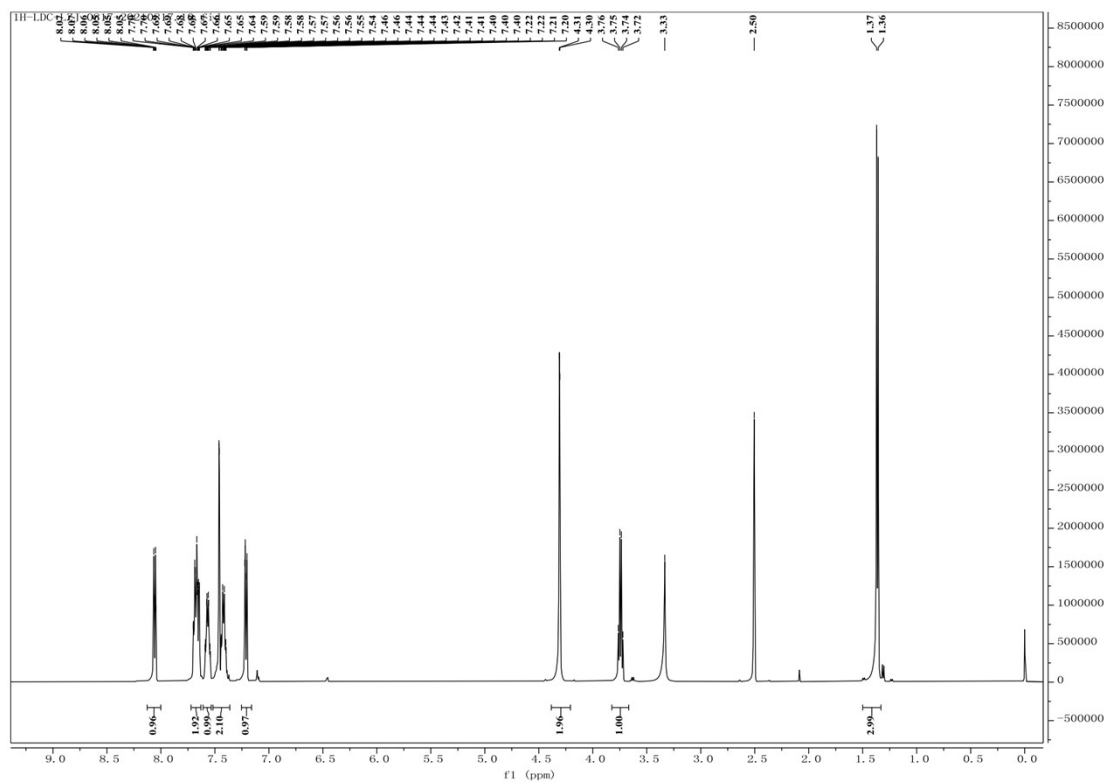


Peaks 19.73 ppm and 63.21 ppm are ascribed to Et₂O.

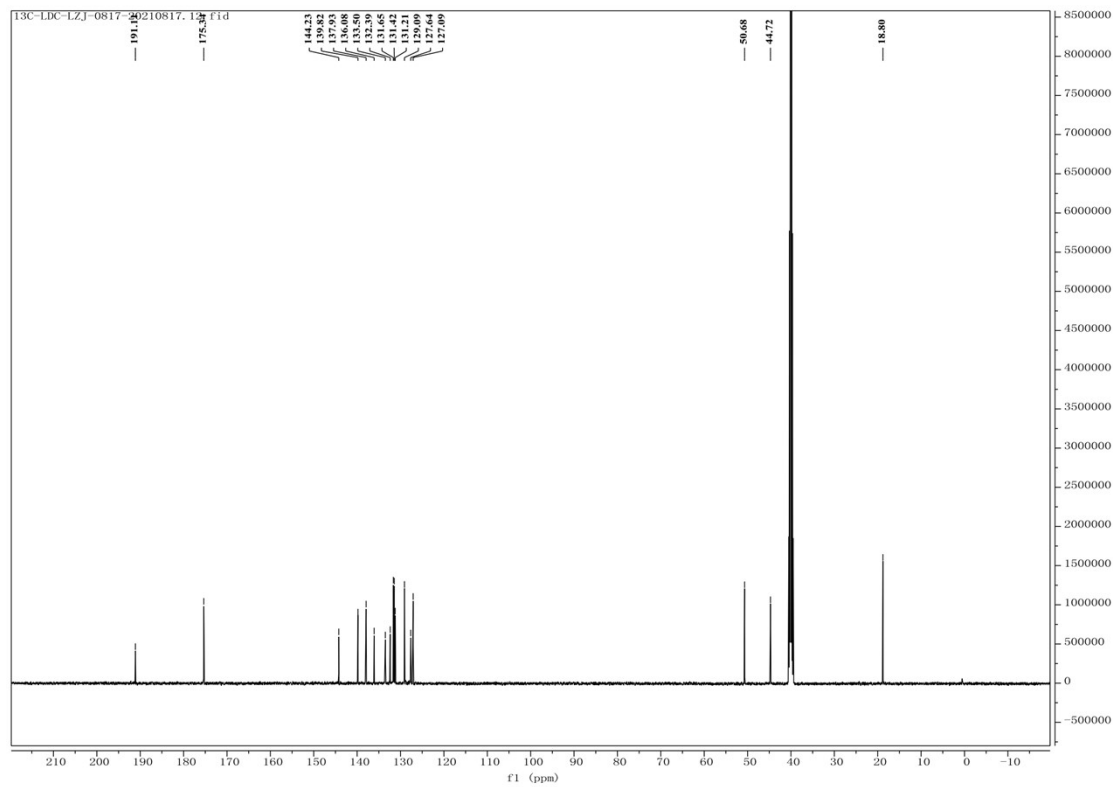
¹³C NMR spectrum for complex **4**



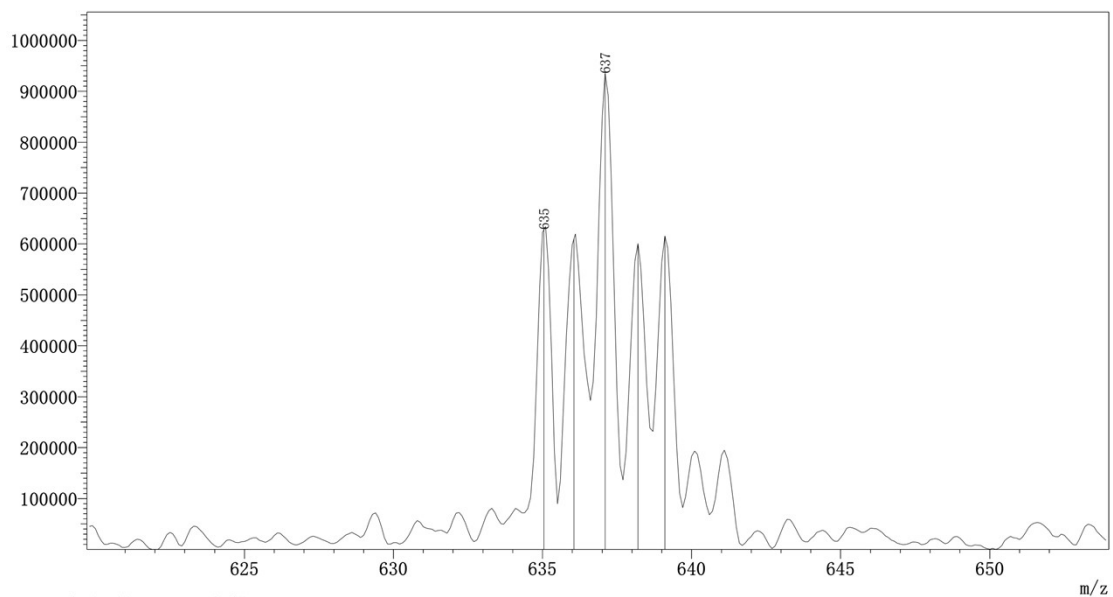
MS spectrum for complex **4**



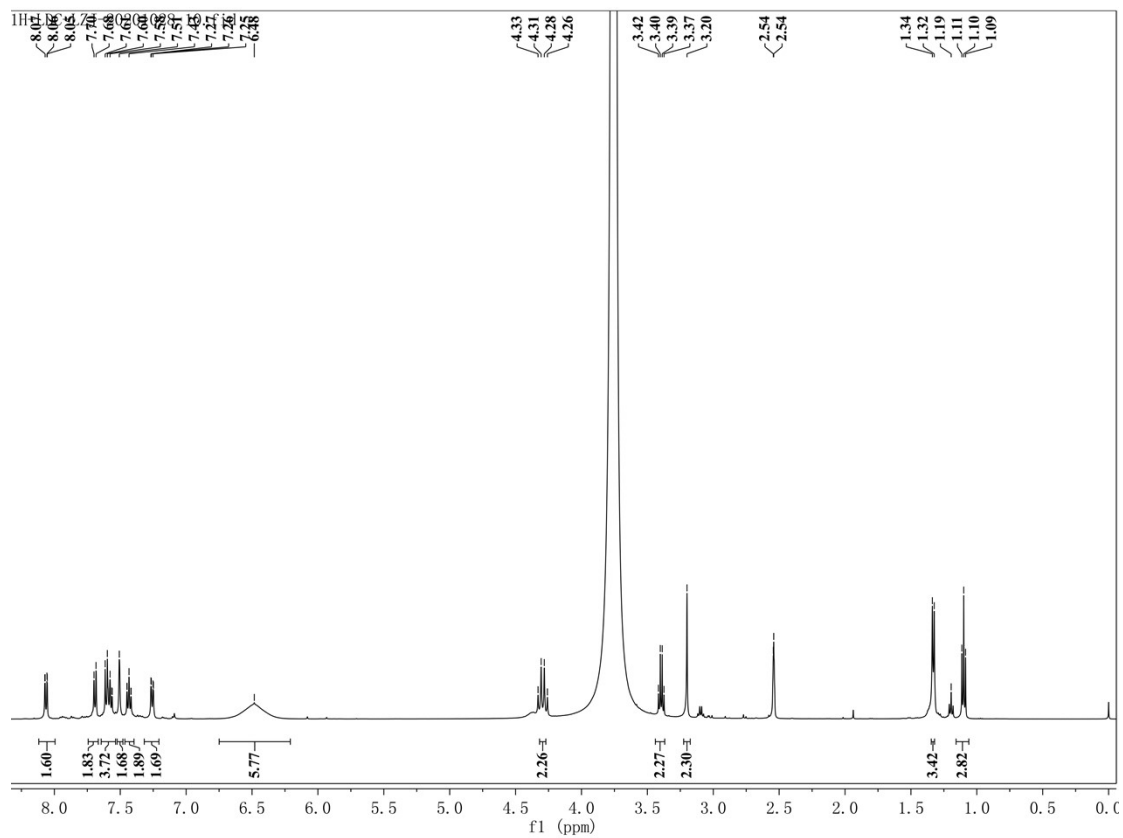
¹H NMR spectrum for complex 5



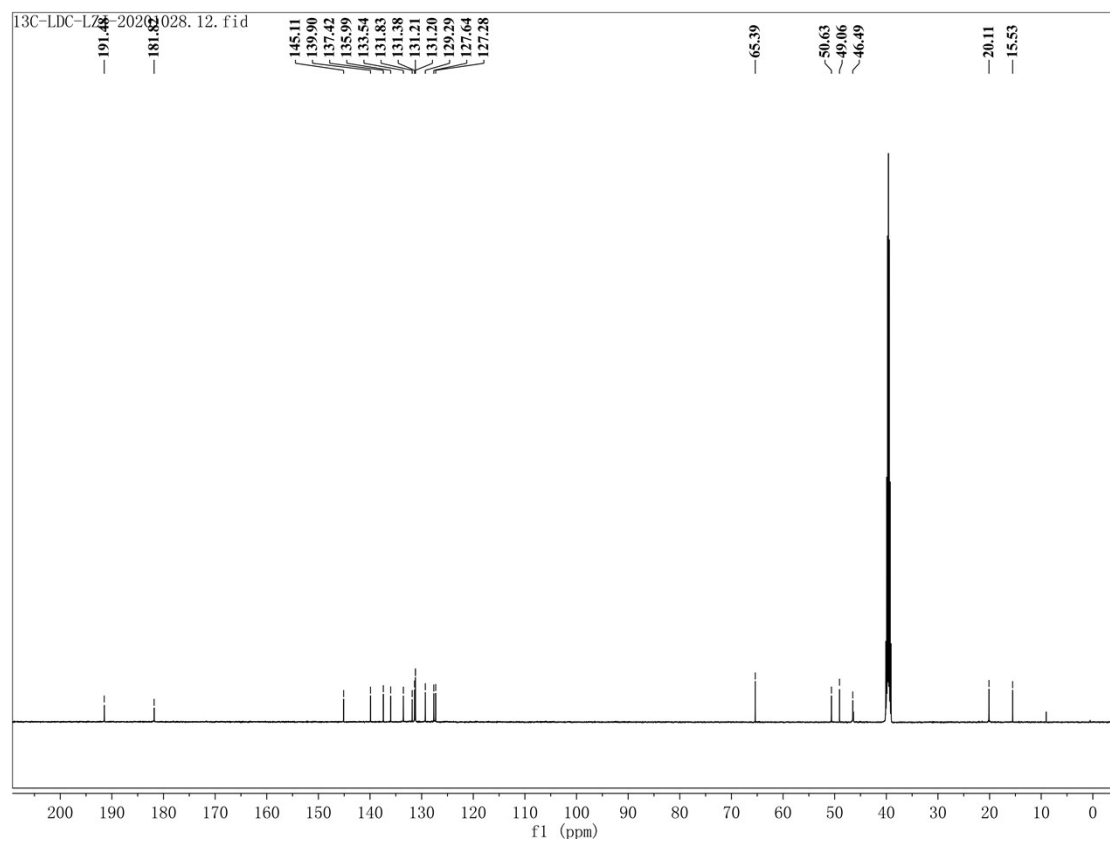
¹³C NMR spectrum for complex 5



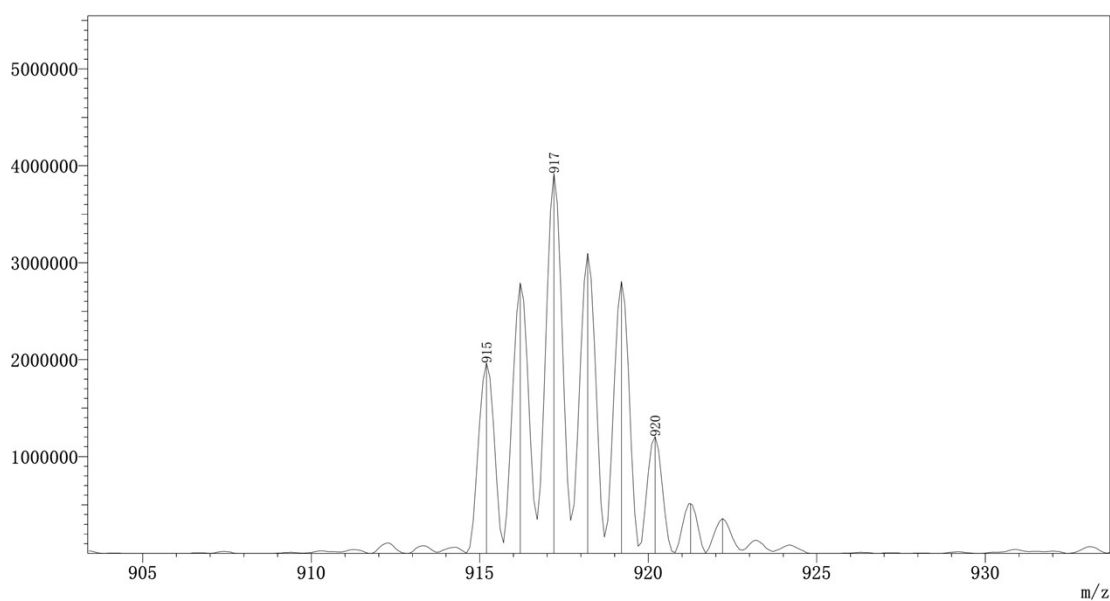
MS spectrum for complex 5



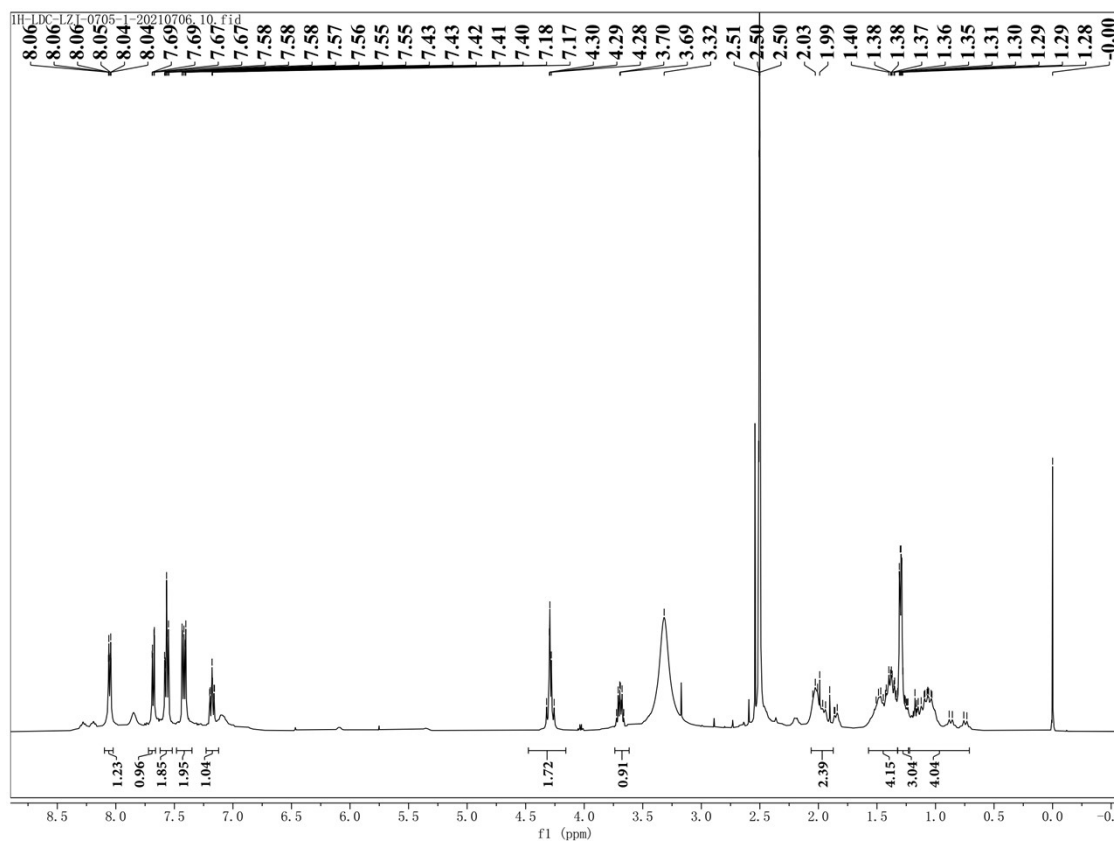
¹H NMR spectrum for complex 6



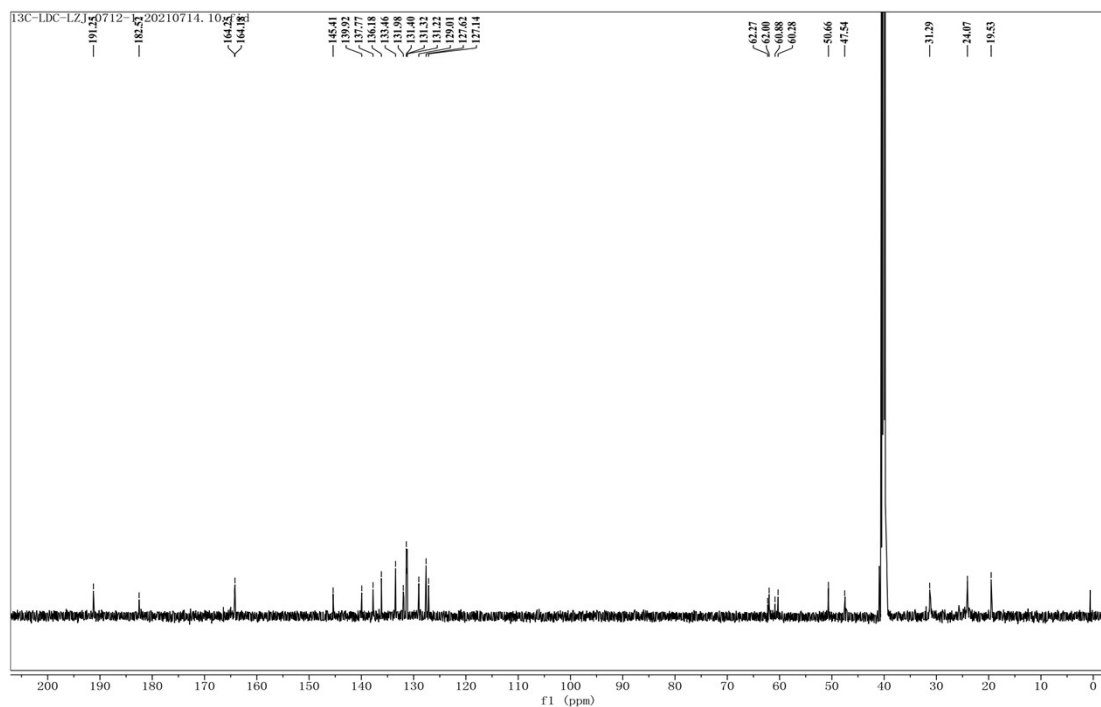
¹³C NMR spectrum for complex **6**



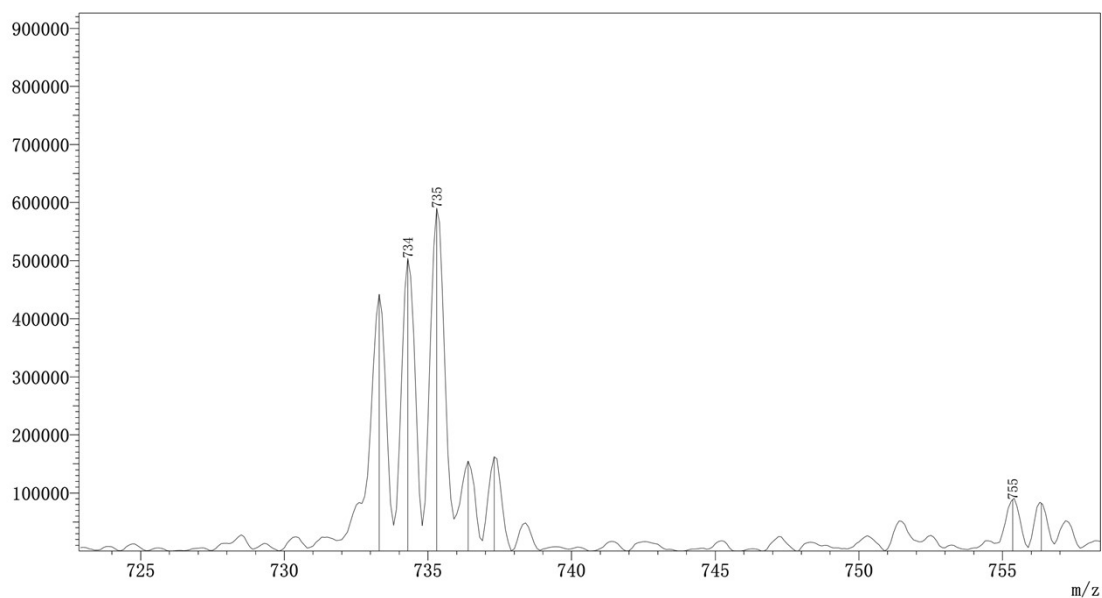
MS spectrum for complex **6**



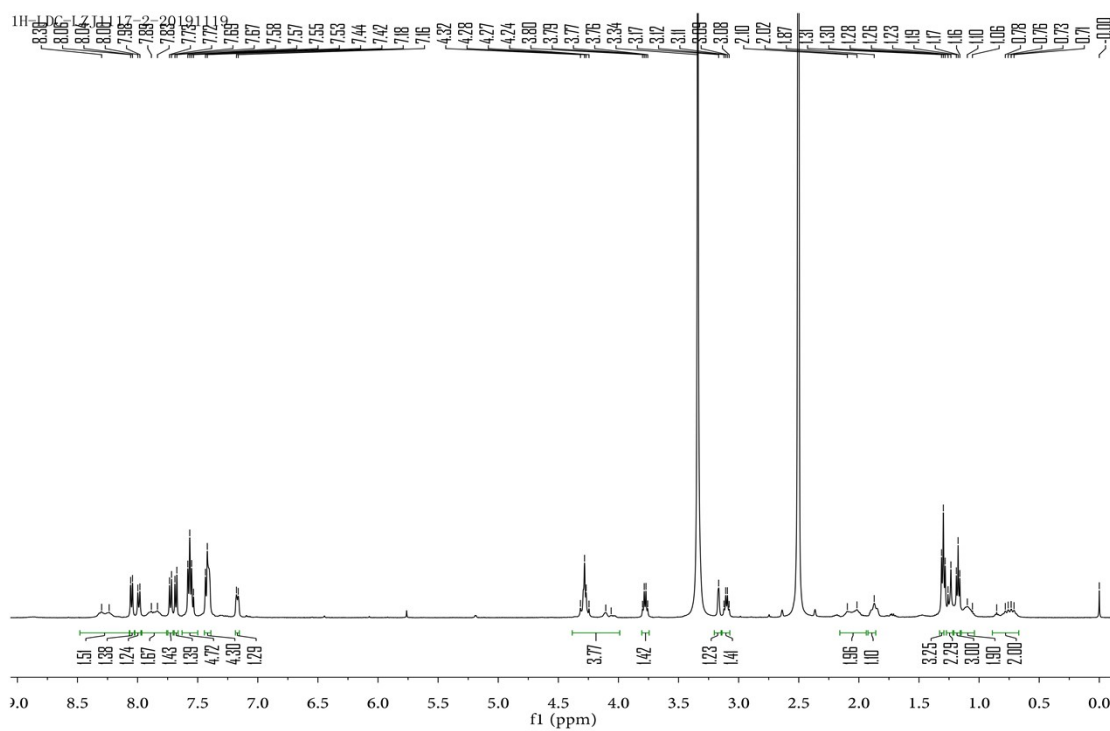
¹H NMR spectrum for complex 7



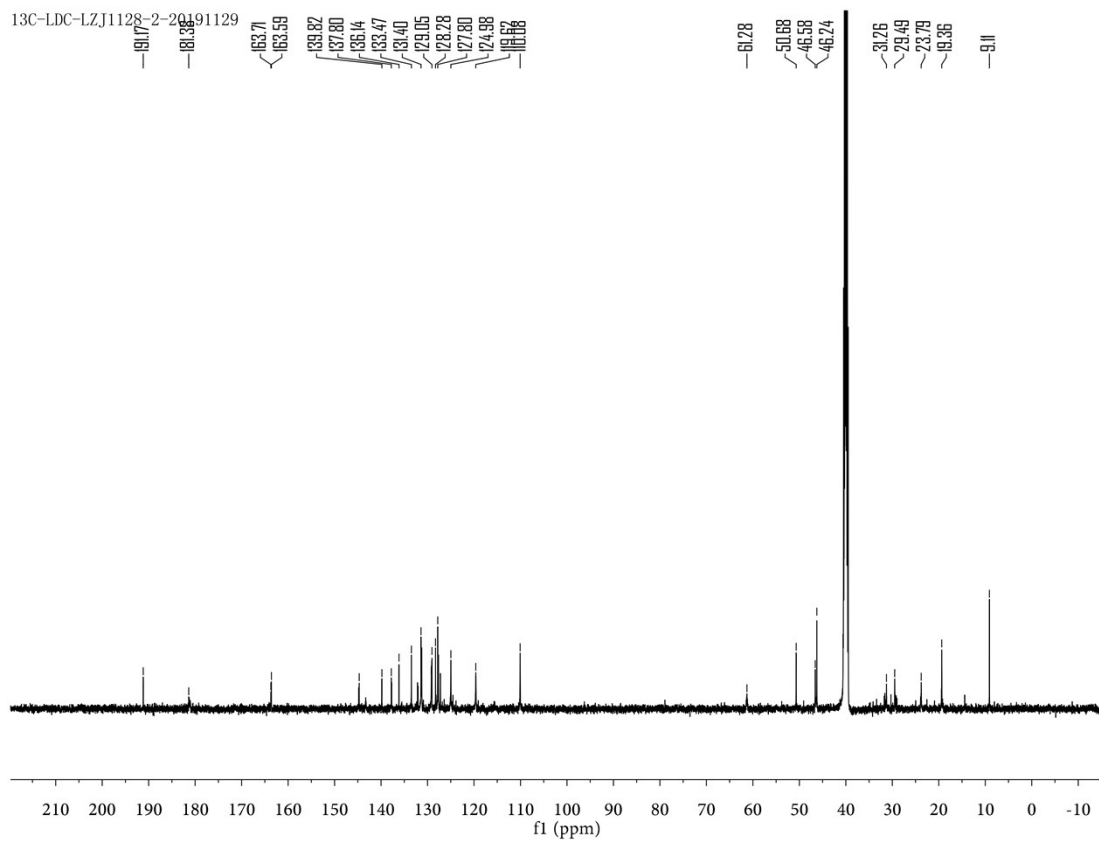
¹³C NMR spectrum for complex 7



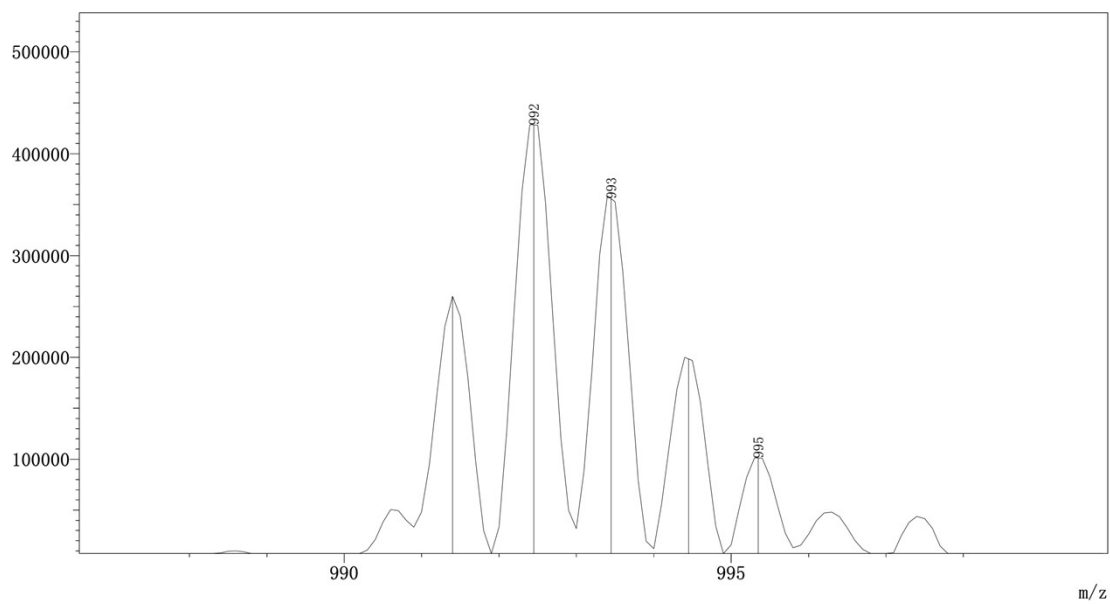
MS spectrum for complex 7



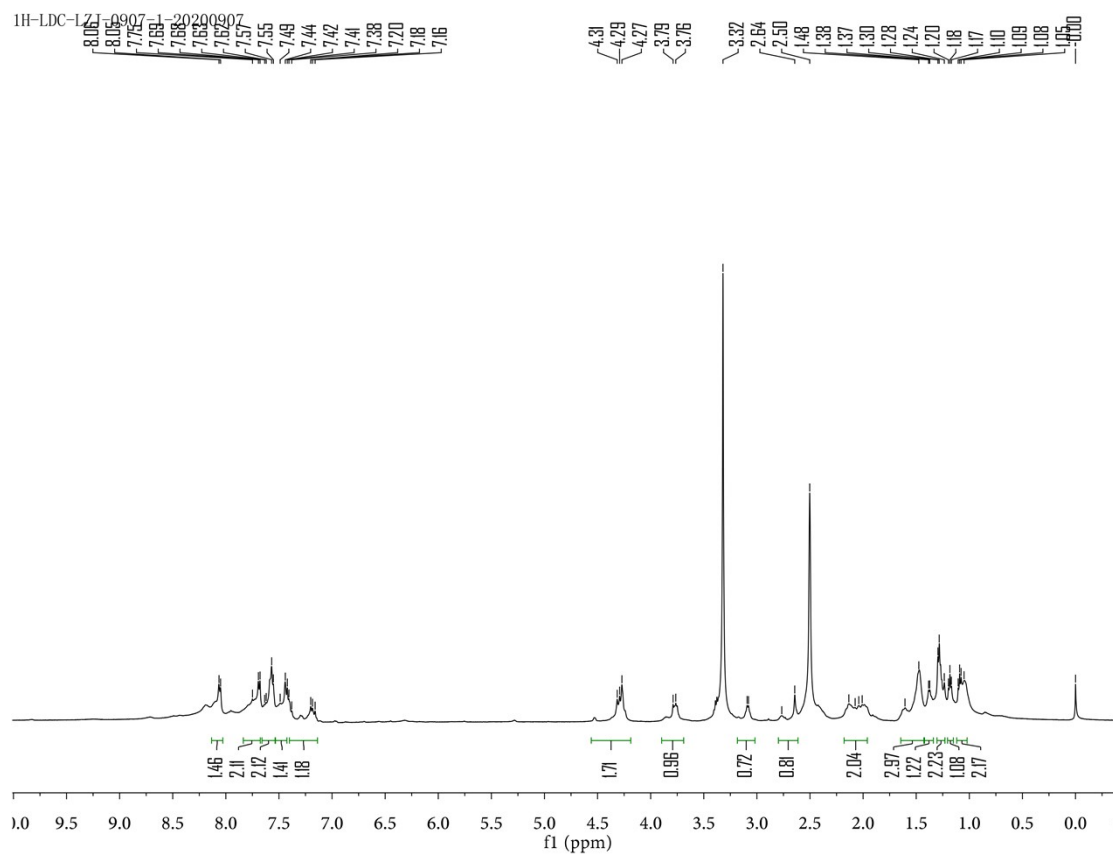
¹H NMR spectrum for complex 8



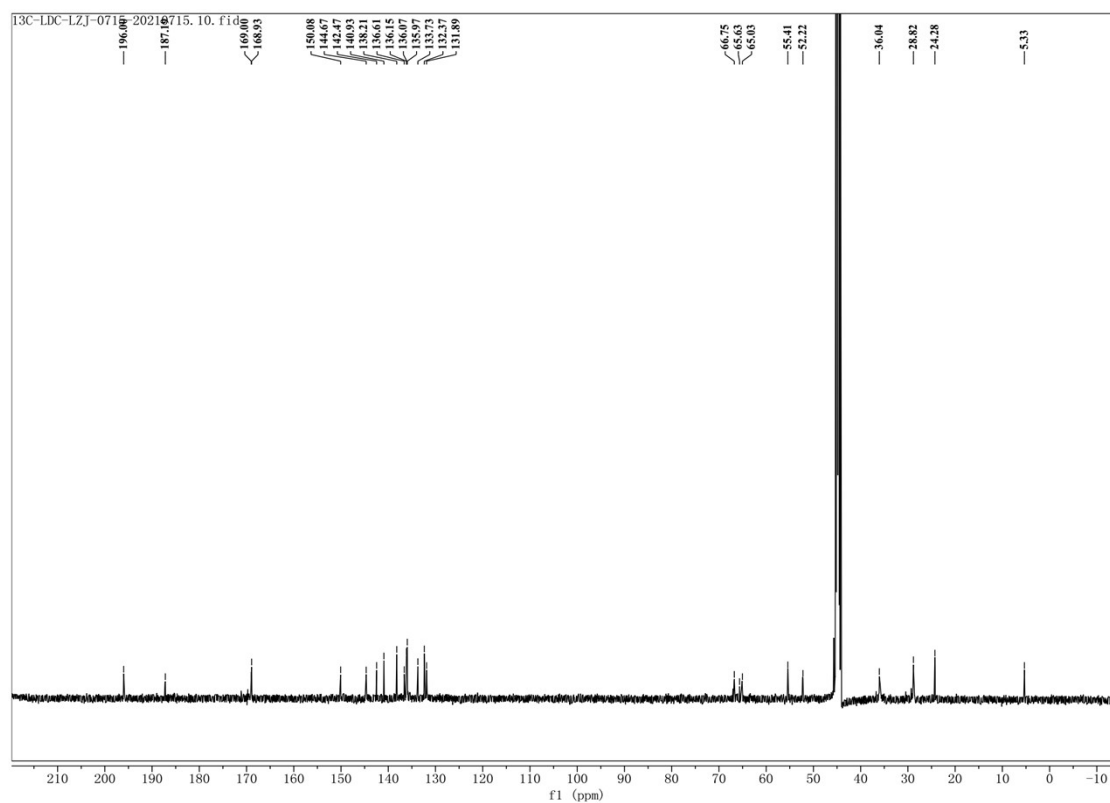
¹³C NMR spectrum for complex **8**



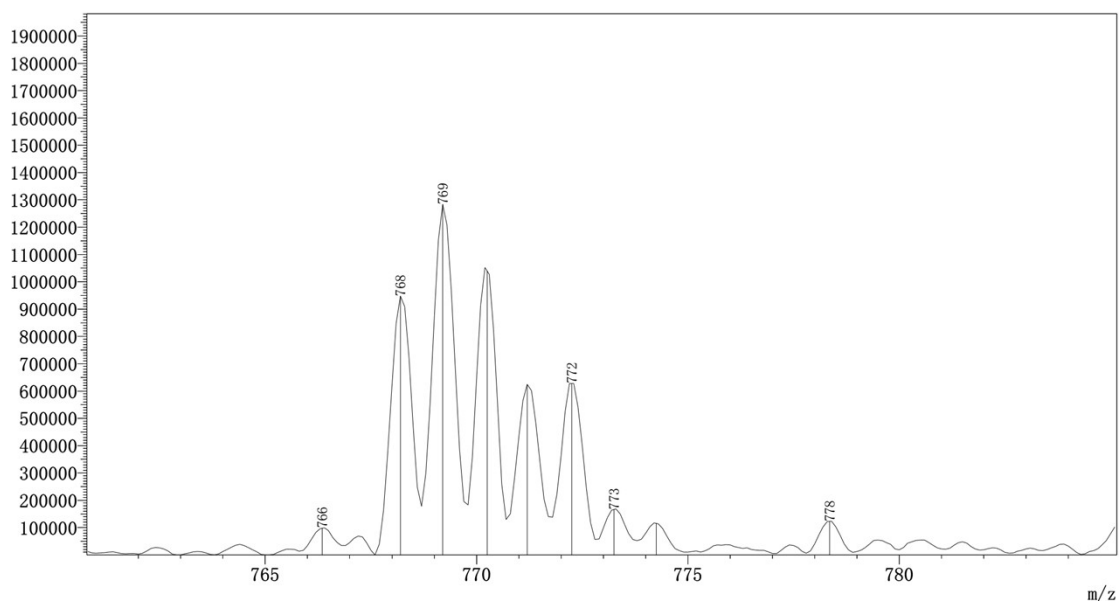
MS spectrum for complex **8**



¹H NMR spectrum for complex **9**



¹³C NMR spectrum for complex **9**



MS spectrum for complex **9**

Reference

- [S1] Z. Li, Q. Wang, L. Li, Y. Chen, J. Cui, M. Liu, N. Zhang, Z. Liu, J. Han and Z. Wang, *J. Med. Chem.*, 2021, 64, 17920–17935.
- [S2] Q. Wang, Z. Huang, J. Ma, X. Lu, L. Zhang, X. Wang, P.G. Wang, Design, synthesis and biological evaluation of a series of new glycosylated platinum(IV) complexes as antitumor agents. *Dalton Trans.* 45 (2016) 10366–10374.