Supporting Information

A novel quinoline-based two-photon fluorescent probe for detecting Cd^{2+} *in vitro* and *in vivo*

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1. Cell cytotoxicity

2. Spectroscopic properties of APQ and APQ-Cd^{2+} complex

3. MALDI-TOF MS spectra of APQ、APQ-Cd^{2+} complex and APQ-Zn^{2+} complex

4. ^{1}H NMR spectra of APQ and APQ-Cd^{2+} complex

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1. Cell cytotoxicity

To ascertain the cytotoxic effect, the MTT (5-dimethylthiazol-2-yl-2, 5-diphenyltetrazolium bromide) assay was performed as previously reported. HeLa cells were passed and plated to ~ 70% confluence in 96-well plates 24h before treatment. Prior to APQ treatment, the DMEM was removed and replaced with fresh DMEM, and aliquots of APQ stock solutions (5 mM DMSO) were added to obtain final concentrations of 10, 30, and 50 μM. The treated cells were incubated for 24 h at 37 ^{o}C and under 5% CO_2. Subsequently, the cells were treated with 5 mg/mL MTT (40 μL /well) and incubated for an additional 4 h (37oC, 5% CO2). Then the cells were dissolved in DMSO (150 μL/well), and the absorbance at 570 nm was recorded. The cell viability (%) was calculated according to the following Equation: Cell viability%=OD570(sample)/OD570(control)×100, where OD570 (sample) represents the optical density of the wells treated with various concentration of 6-MPQ and OD570(control) represents that of the wells treated with DMEM+10% FCS. percent cell survival values are relative to untreated control cells.

Table S1. Cytotoxicity Data of APQ (HeLa, 24 h)

Probe concentration	0μM	10μM	30μM	50μM
Cell survival %	97.37±6.7%	93.25±6.9%	52.75±7.10%	43.68±7.0%

2. Spectroscopic Properties of APQ

Fluorescence quantum yields were determined using the quinine sulfate (in 0.1 N H_2SO_4, Φ =0.55) as the standard (excited by 320 nm).

$$\Phi_U = \frac{\Phi_S(F_U A_S)}{F_S A_U}$$

The quantum yields are calculated by the following equation:
Au and As are the UV absorption of analyte and the standard. Fu and Fs are integrated fluorescence emission of analyte and the standard.
The dissociation constant K_d is caculated by following equation:

$$\frac{F}{F_0} = 1 + \left(\frac{F_{max}}{2F_0} - \frac{1}{2}\right)\left\{1 + \frac{C_M}{C_L} + \frac{1}{K_S C_L} - \left[\left(1 + \frac{C_M}{C_L} + \frac{1}{K_S C_L}\right)^2 - 4\frac{C_M}{C_L}\right]^{\frac{1}{2}}\right\}$$

where F and F_0 are the fluorescence intensity of APQ in the presence and absence of Cd^{2+}, C_M and C_L are the concentrations of Cd^{2+} and APQ ; K_S is the stability constant. K_d is the reciprocal of K_s.

Table S2 Spectroscopic Properties of **APQ** and **APQ-Cd^{2+}**

absorption maxima λ (nm), ε×10^4 (M^{-1}cm^{-1})		emission maxima λ (nm)		fluorescence quantum yields *Φ*	
APQ	APQ+Cd^{2+}	APQ	APQ+Cd^{2+}	APQ	APQ+Cd^{2+}
27044	23980	480	520	0.057	0.2124

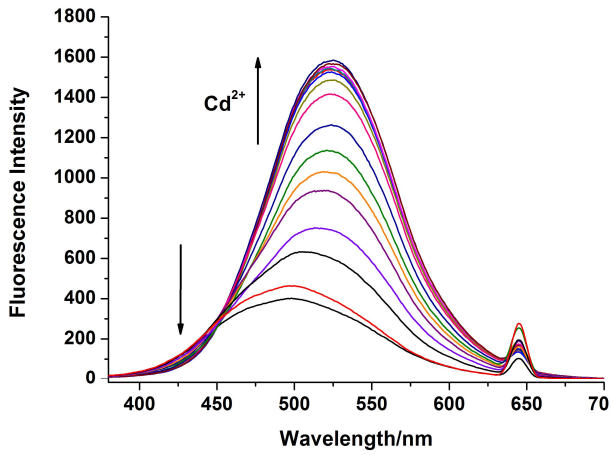


Fig S1. Fluorescence emission spectra of **APQ** (25 μM) with the excitation at 320 nm upon the titration of Cd^{2+} (0-1.5 equiv) in the methanol-water solutions (1:9, v/v, 50 mM HEPES buffer, pH = 7.4).

3. MALDI-TOF MS spectra of APQ、APQ-Cd^{2+} complex and APQ-Zn^{2+} complex

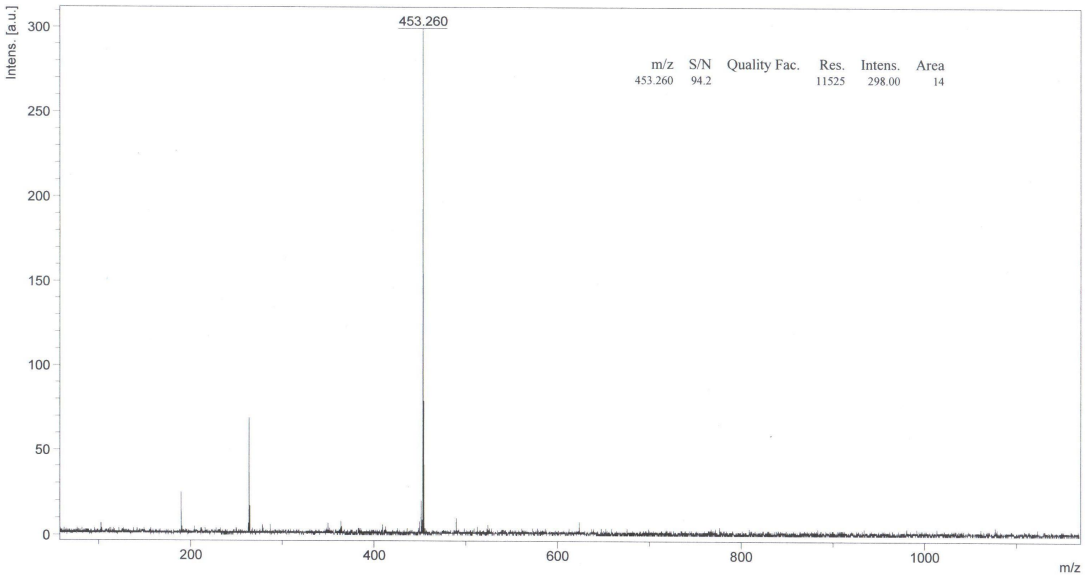


Fig S2. MALDI TOF MS of **APQ** (DCTB as matrix)

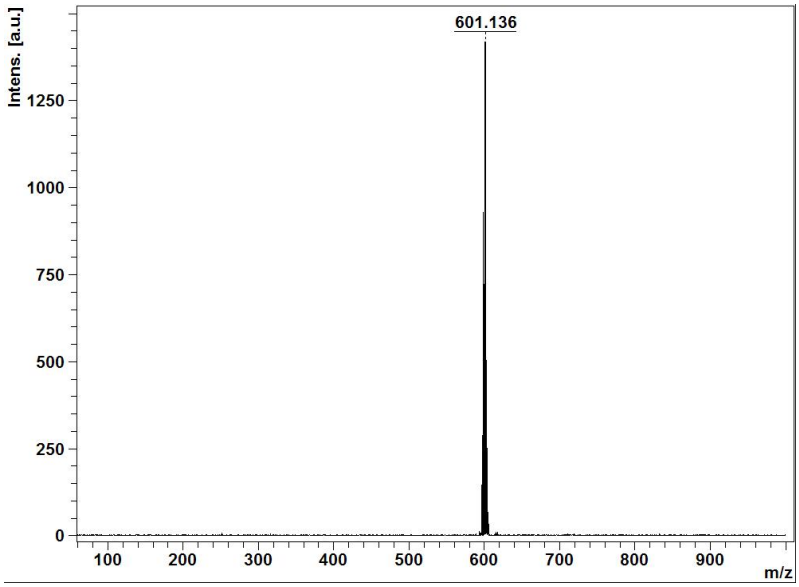


Fig S3. MALDI-TOF MS spectra of **APQ**-Cd^{2+} complex

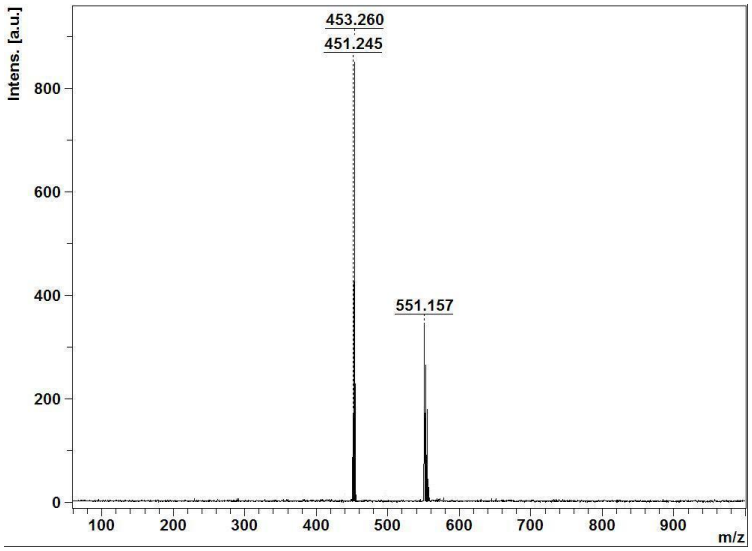


Fig S4. MALDI-TOF MS spectra of **APQ**-Zn^{2+} complex

4. ^{1}H NMR spectra of APQ and APQ-Cd^{2+} complex

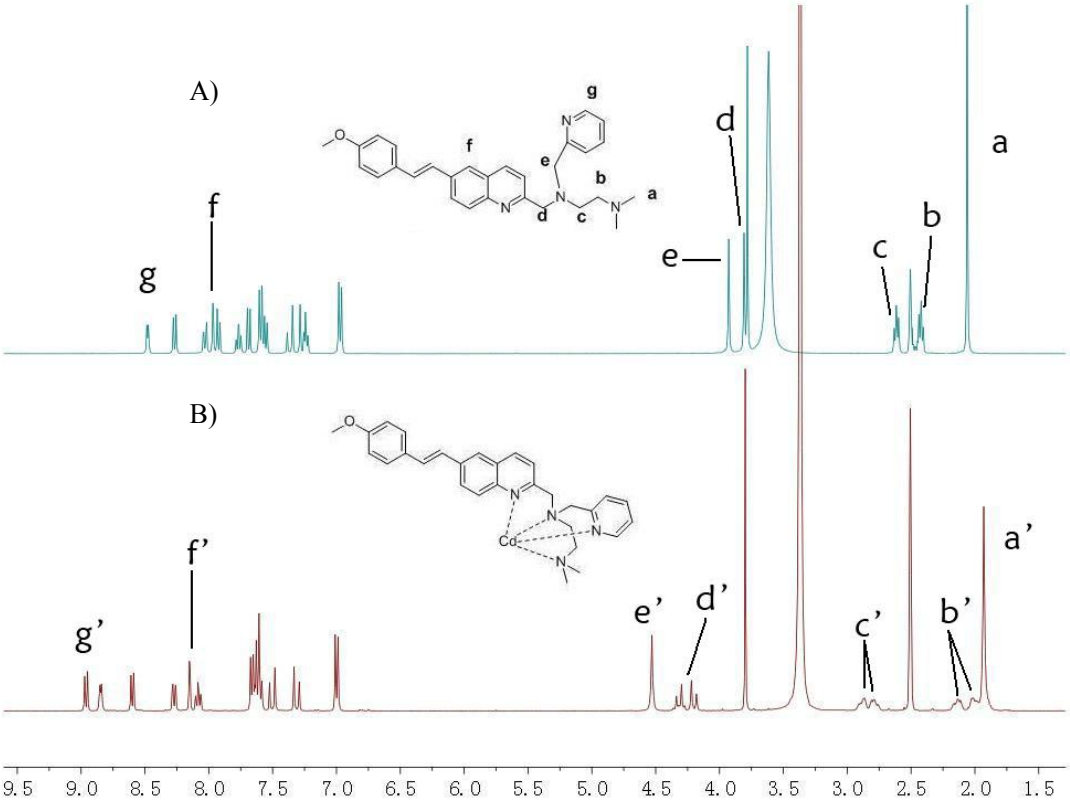
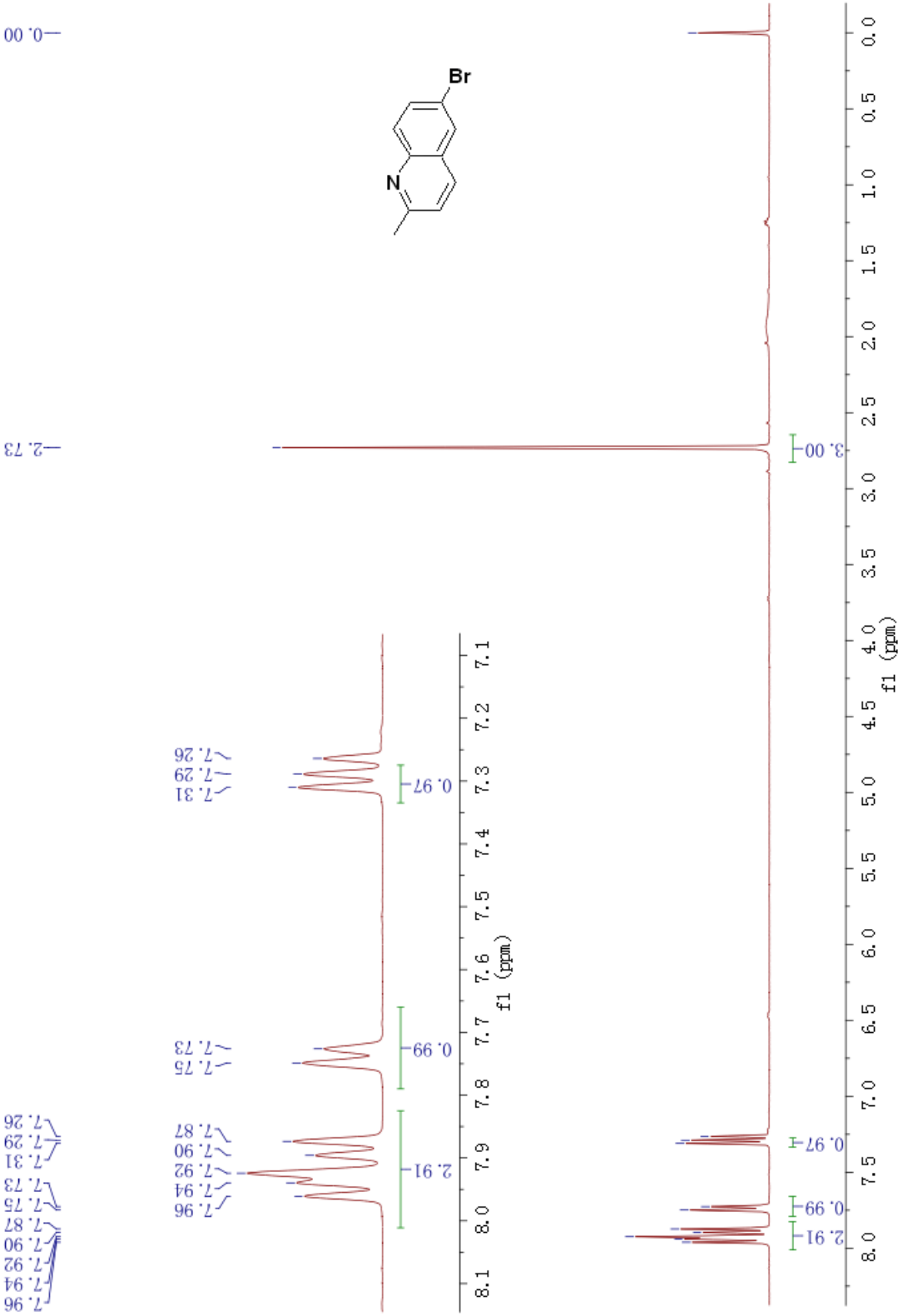
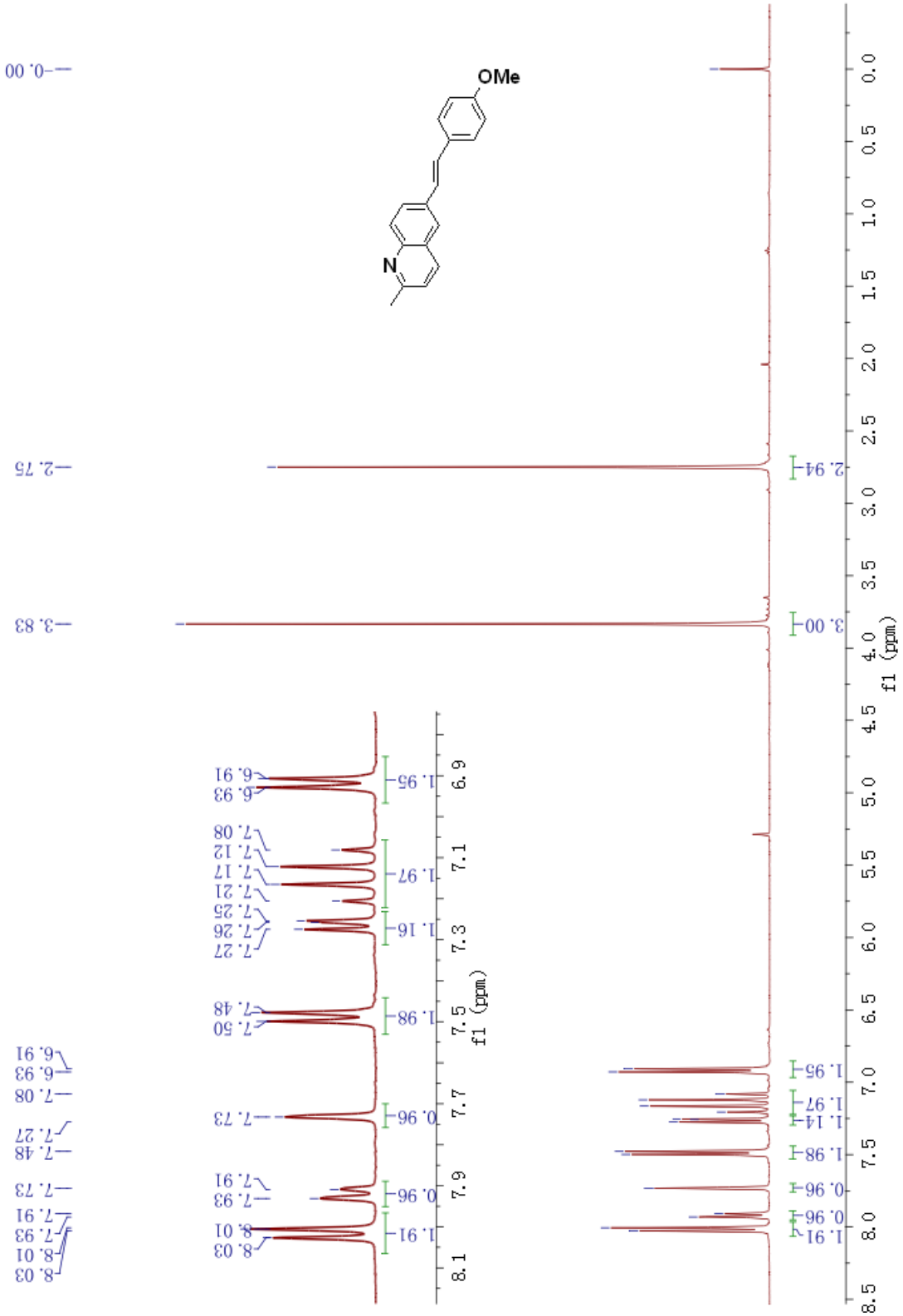
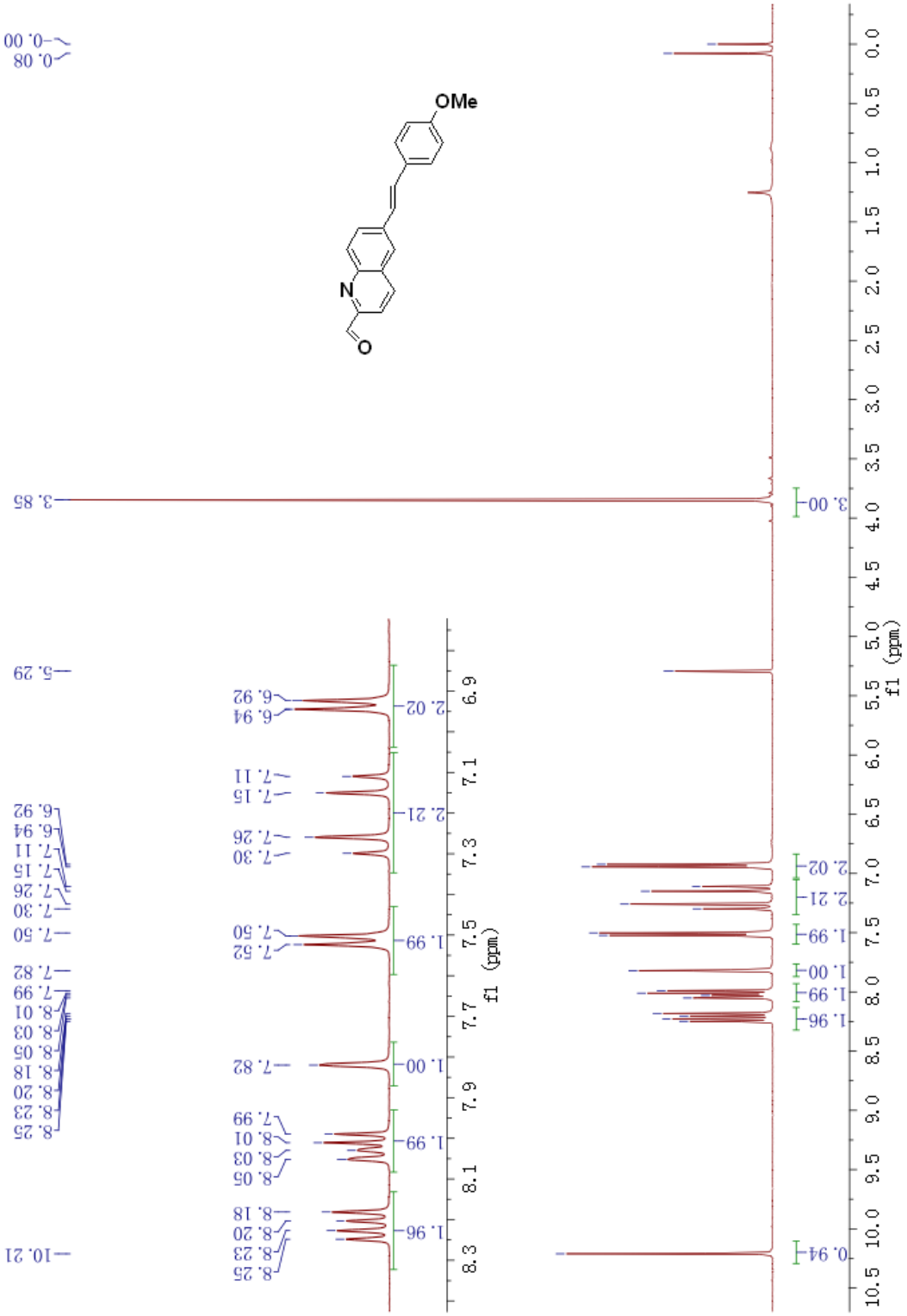


Fig S5. The ^{1}H-NMR spectra of the **APQ** (A) and **APQ**+1.2eq Cd^{2+} (B) in DMSO-d6.

5. NMR spectra



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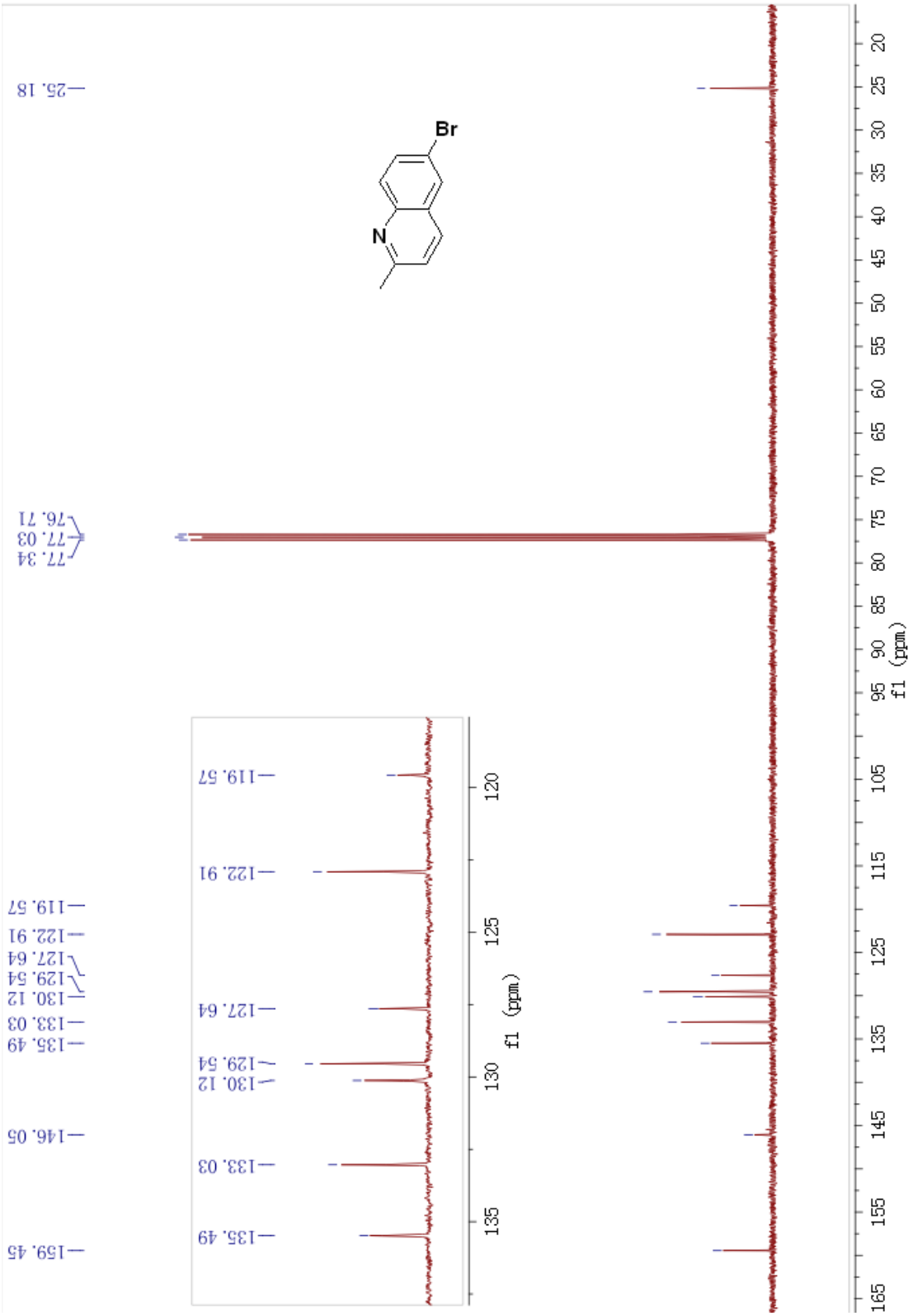
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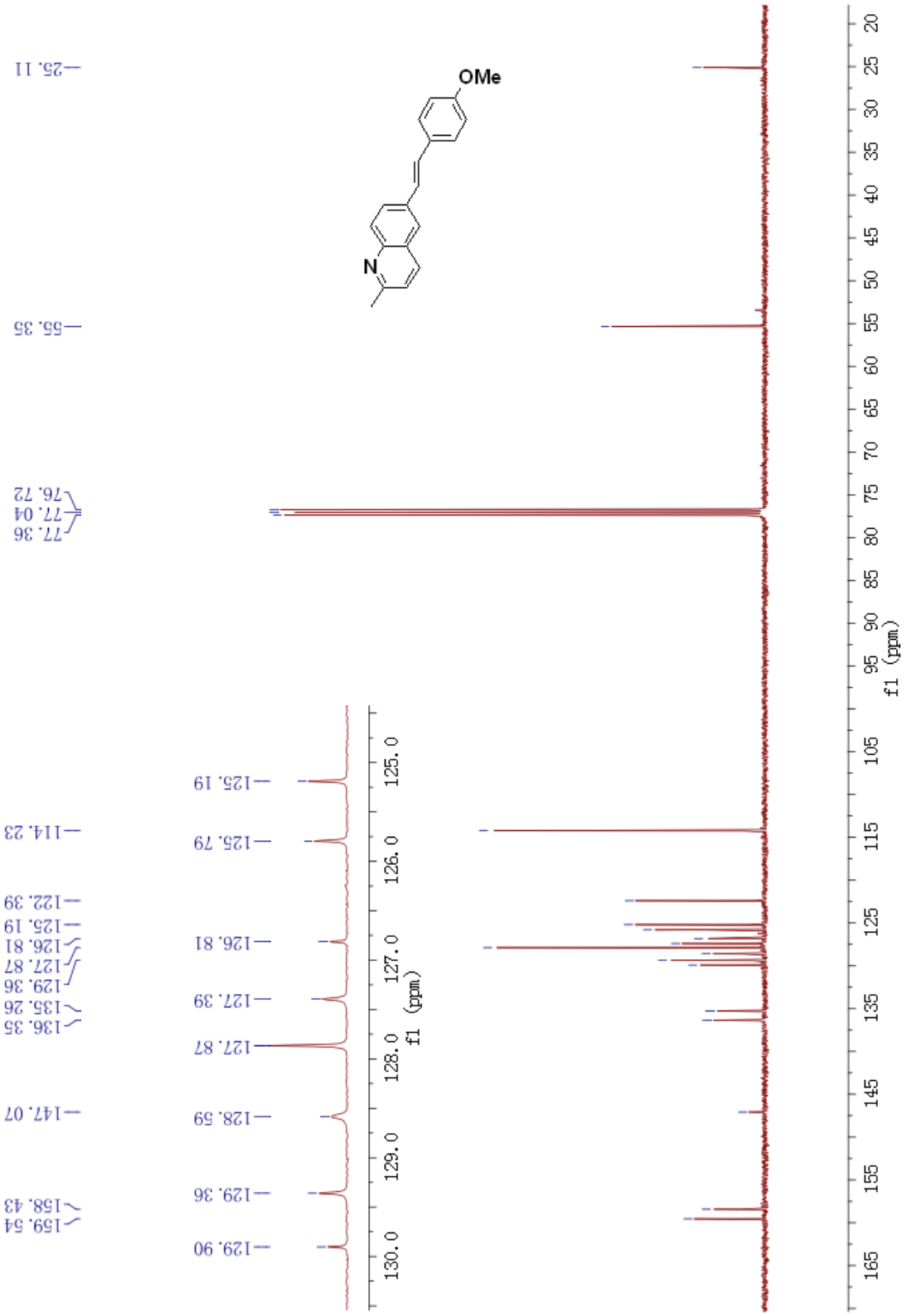
f1 (ppm)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f1 (ppm)
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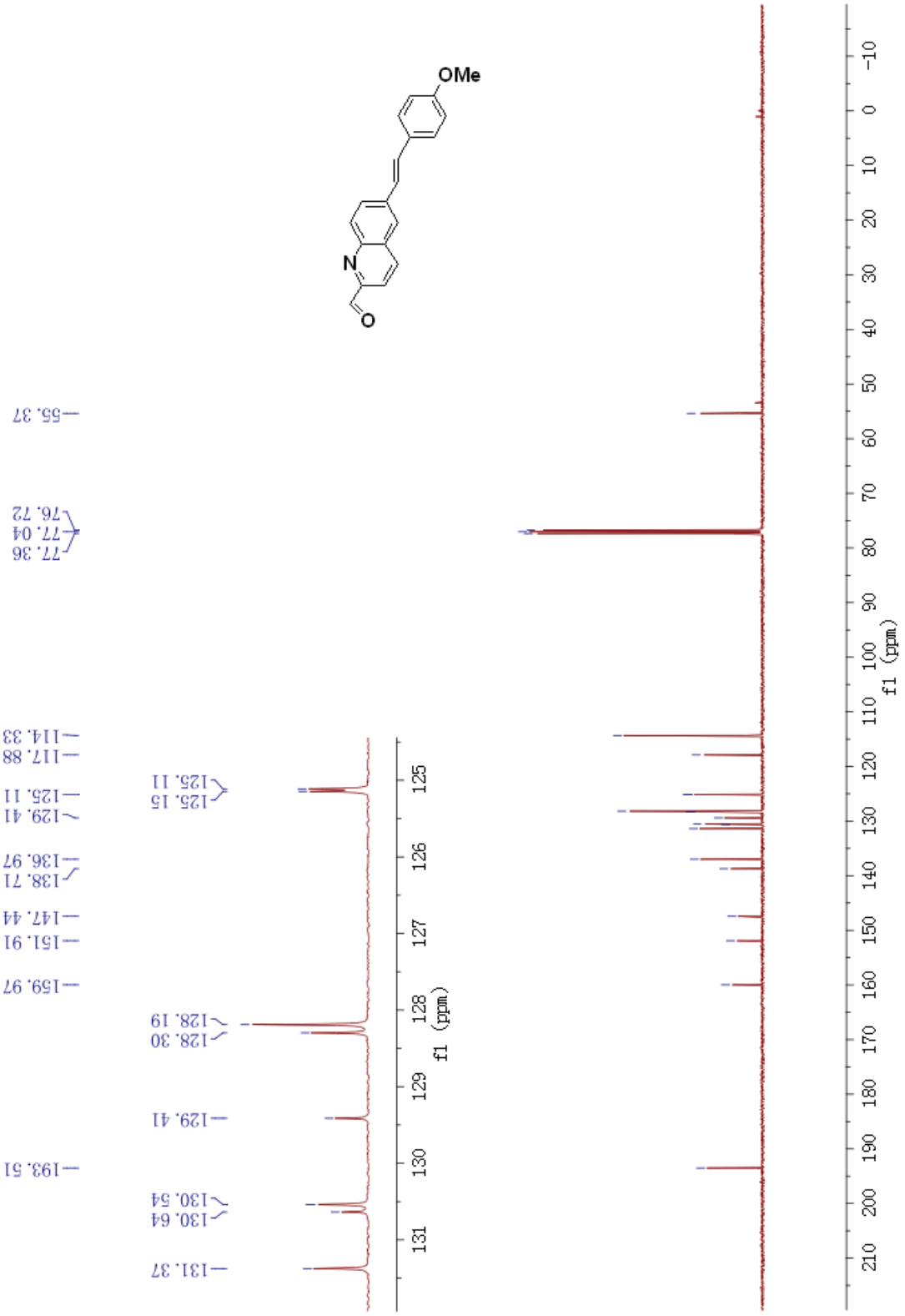
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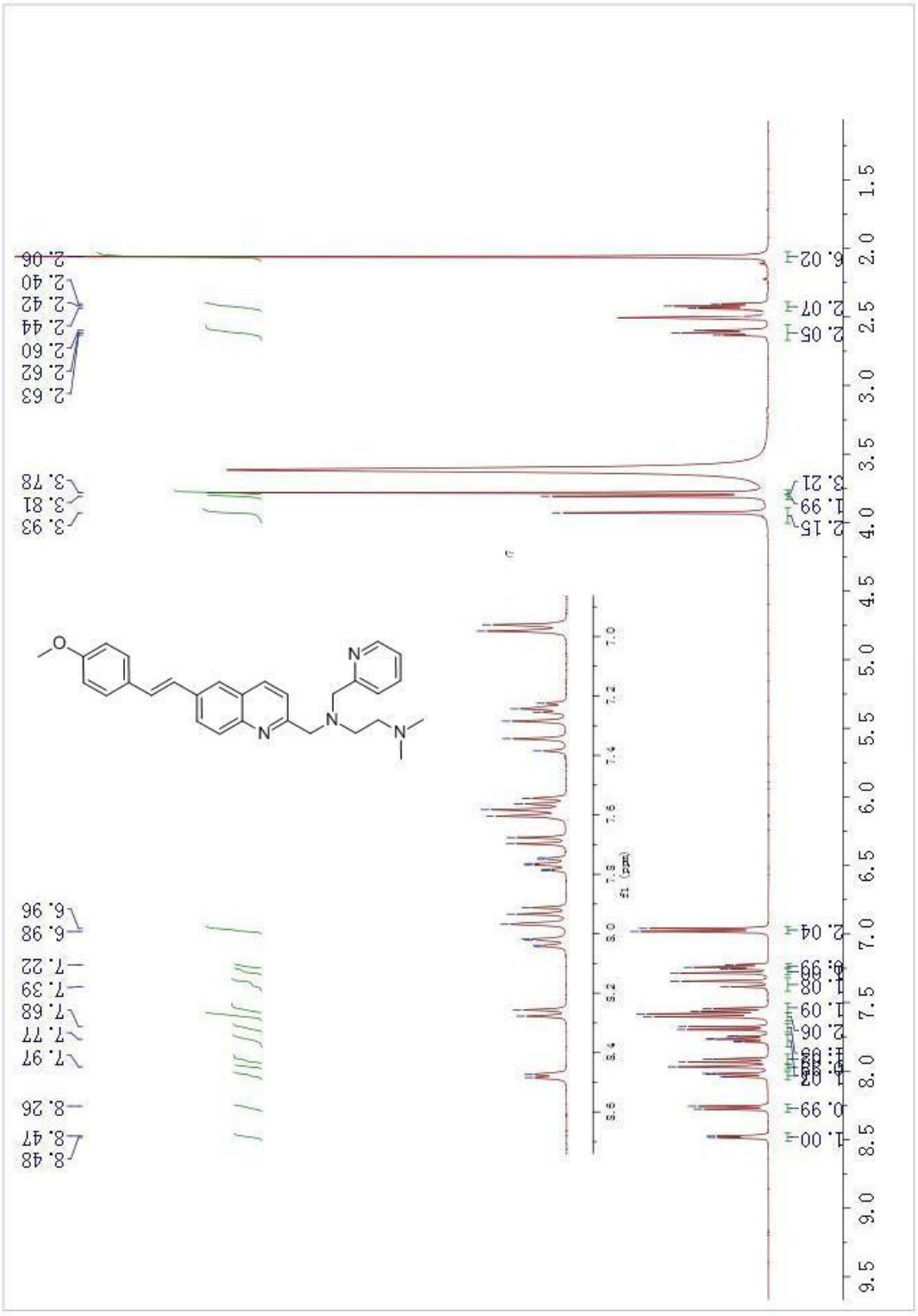
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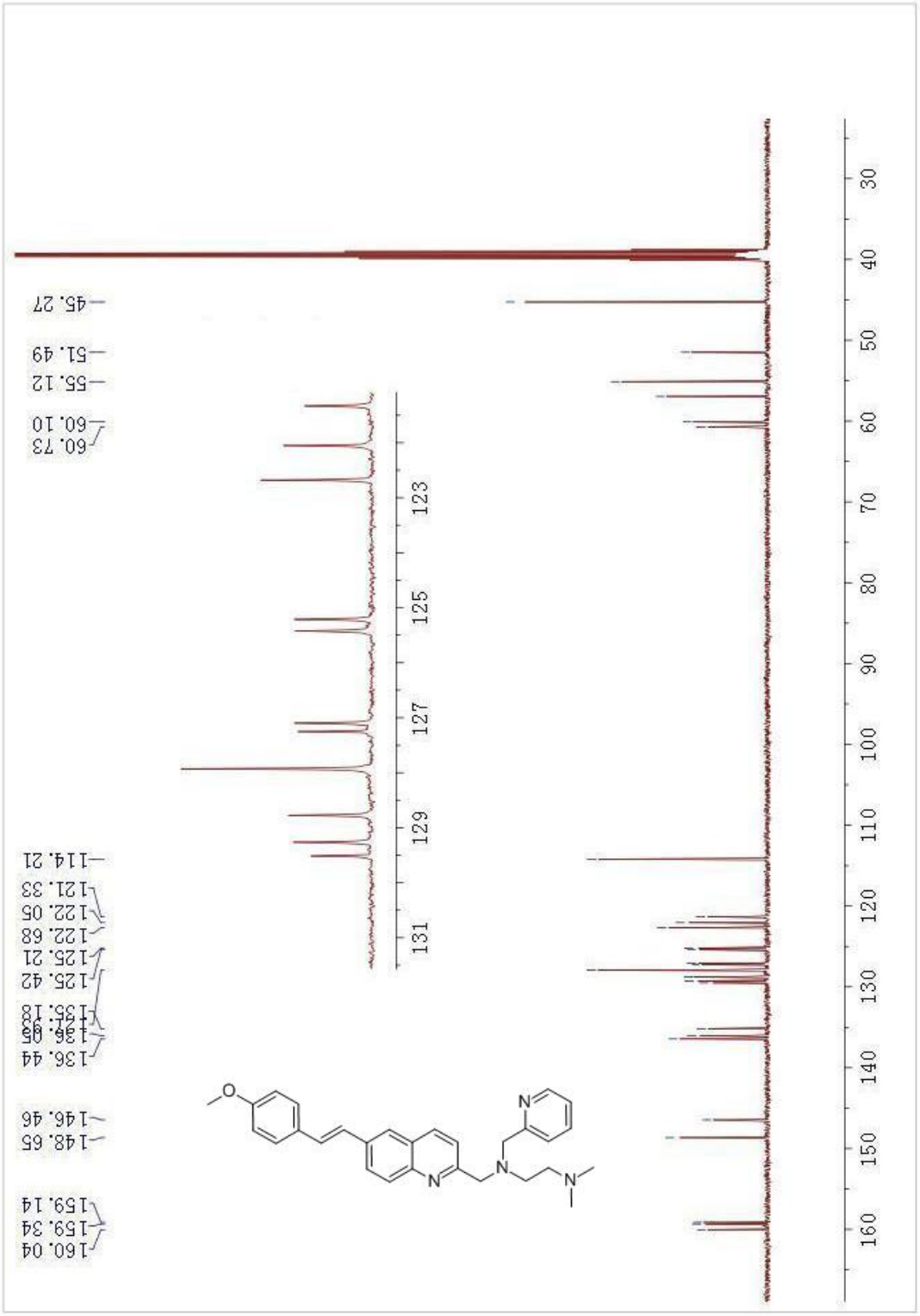
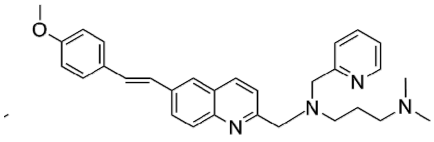
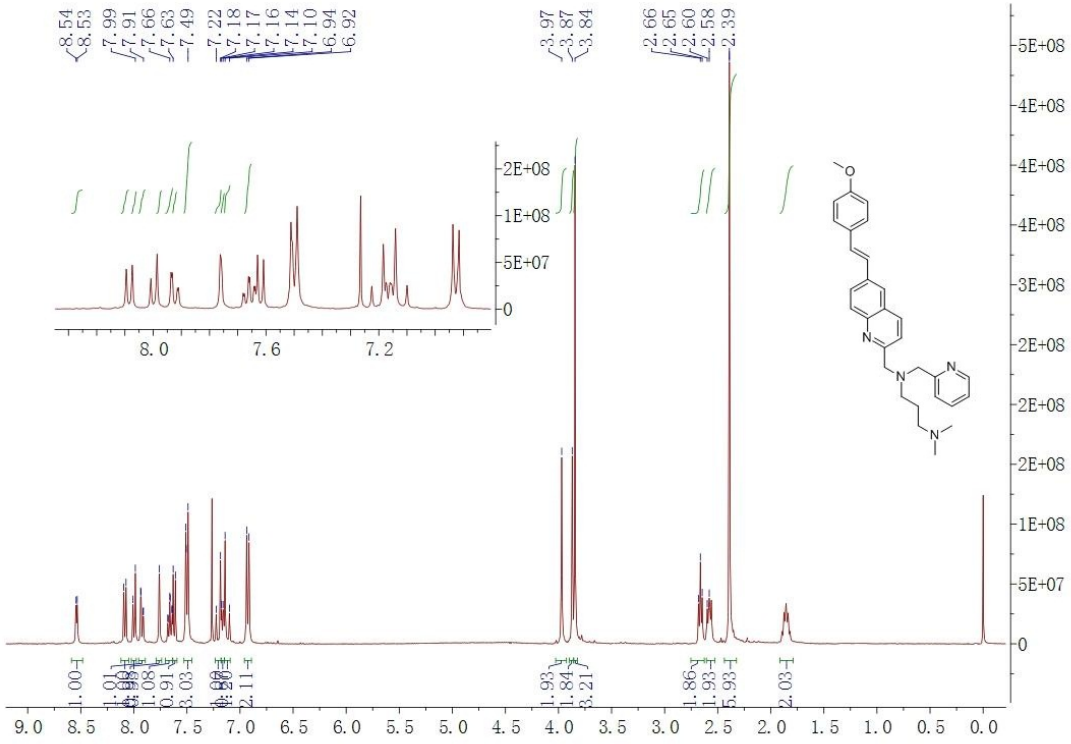




APPQ

^{1}HNMR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1.93
5.93
2.03
9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0
8.0
7.6
7.2
5E+08
4E+08
3E+08
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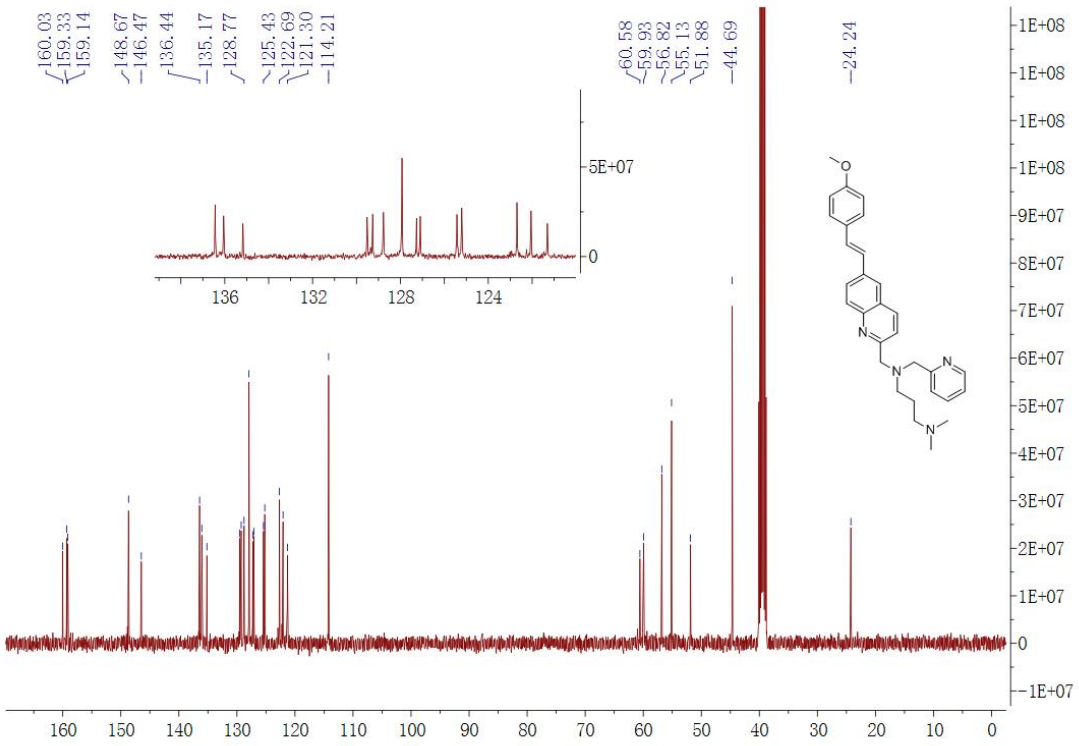
5E+07
0

^{13}CNMR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114.21
60.58
59.93
56.82
55.13
51.88
44.69
24.24
5E+07
0
136
132
128
124
1E+08
1E+08
1E+08
1E+08
9E+07
8E+07
7E+07
6E+07
5E+07
4E+07
3E+07
2E+07
1E+07
0
-1E+07
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0