

## **An unnatural tripeptide structure containing intramolecular double**

### **H-bonds mimics turn hairpin conformation**

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## **Supporting Information**

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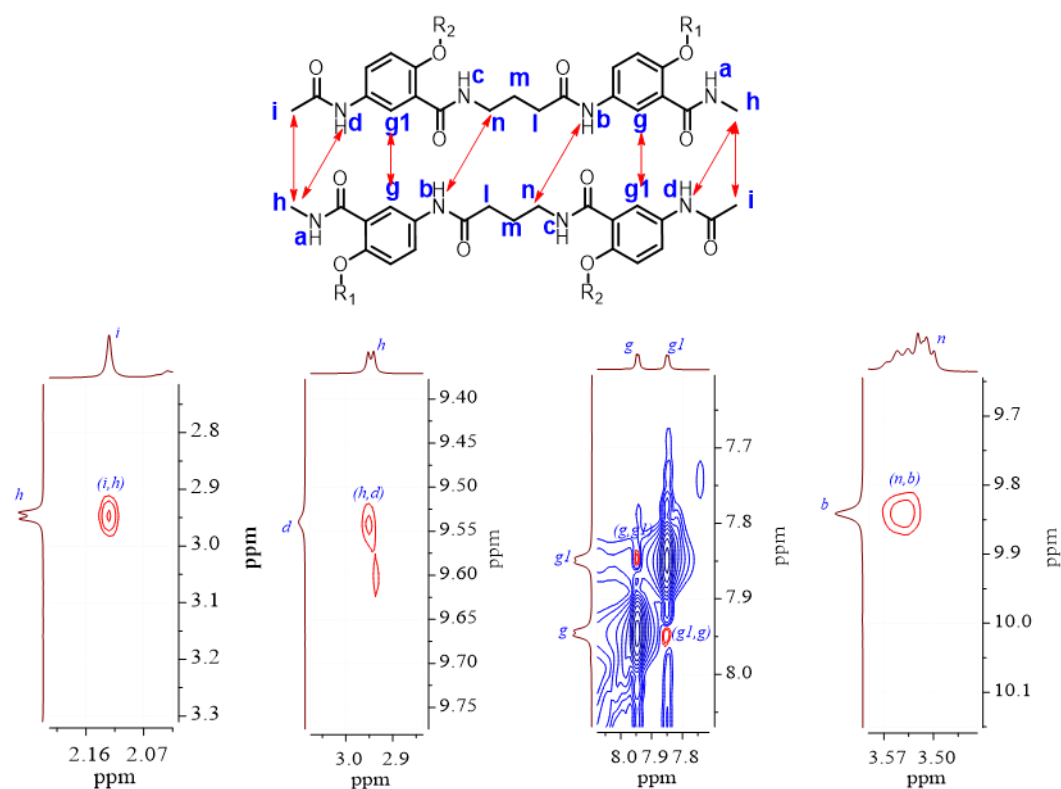
## I. Supplementary Tables and Figures

**Table S1.** Differences in the chemical shifts of the amide protons in CDCl<sub>3</sub> containing 0% and 50% DMSO-*d*<sub>6</sub><sup>a</sup>

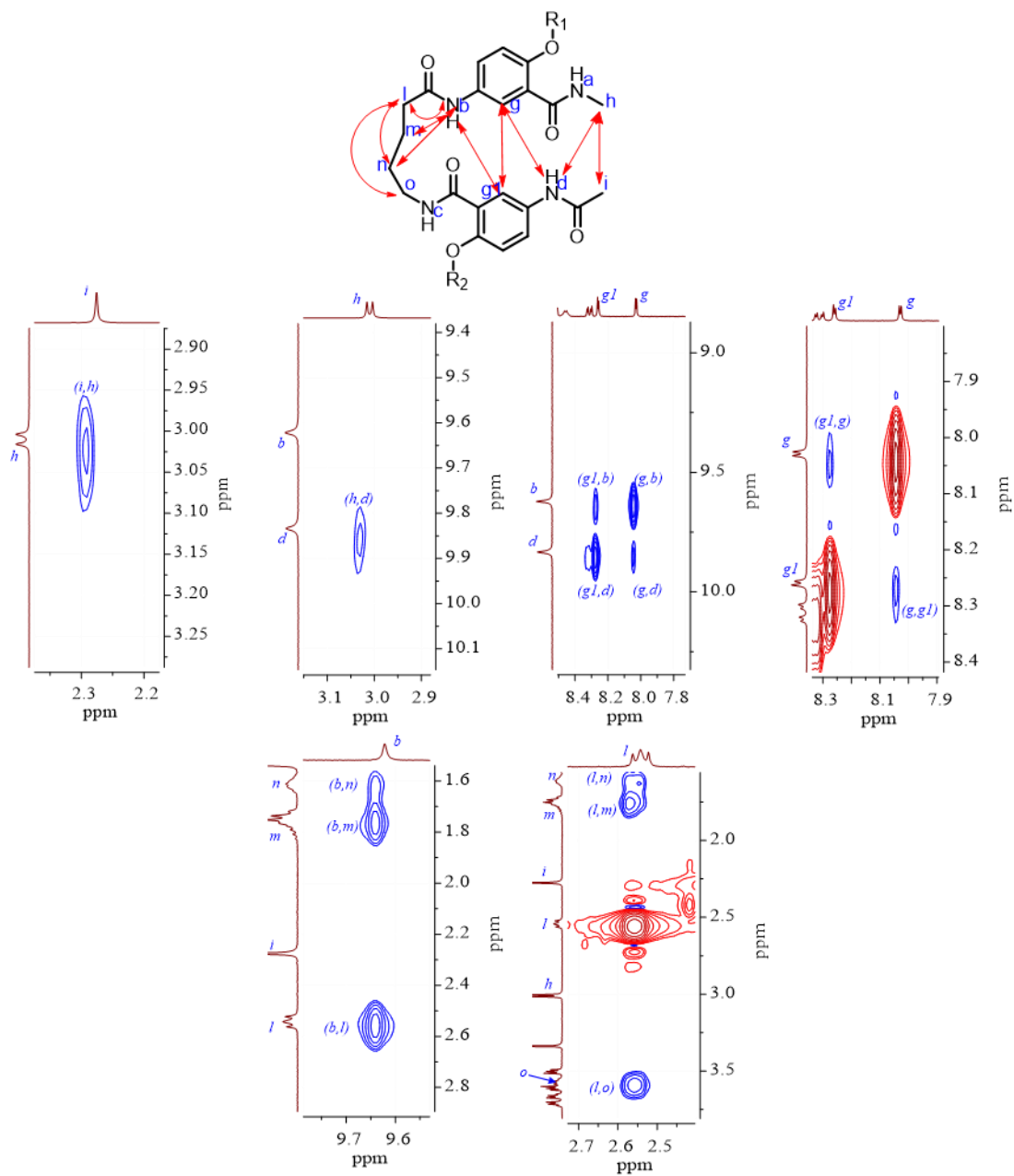
| Entry | $\Delta\delta_{\text{NH}}$ (ppm) <sup>b</sup> |         |         |         |
|-------|---|---------|---------|---------|
|       | A   | b       | c       | D       |
| 1a    | -0.1232                                       | 0.1549  | -0.1323 | 0.5538  |
| 1b    | -0.2040                                       | 0.1916  | -0.5477 | -0.0291 |
| 1c    | -0.1950                                       | 0.3570  | -0.3215 | 0.2378  |
| 2a    | -0.1845                                       | -0.1336 | -0.7653 | 0.0389  |
| 2b    | -0.3019                                       | -0.5583 | -0.2910 | 0.1557  |

<sup>a</sup><sup>1</sup>H-NMR spectra were recorded in CDCl<sub>3</sub> (600 MHz, 298 K).

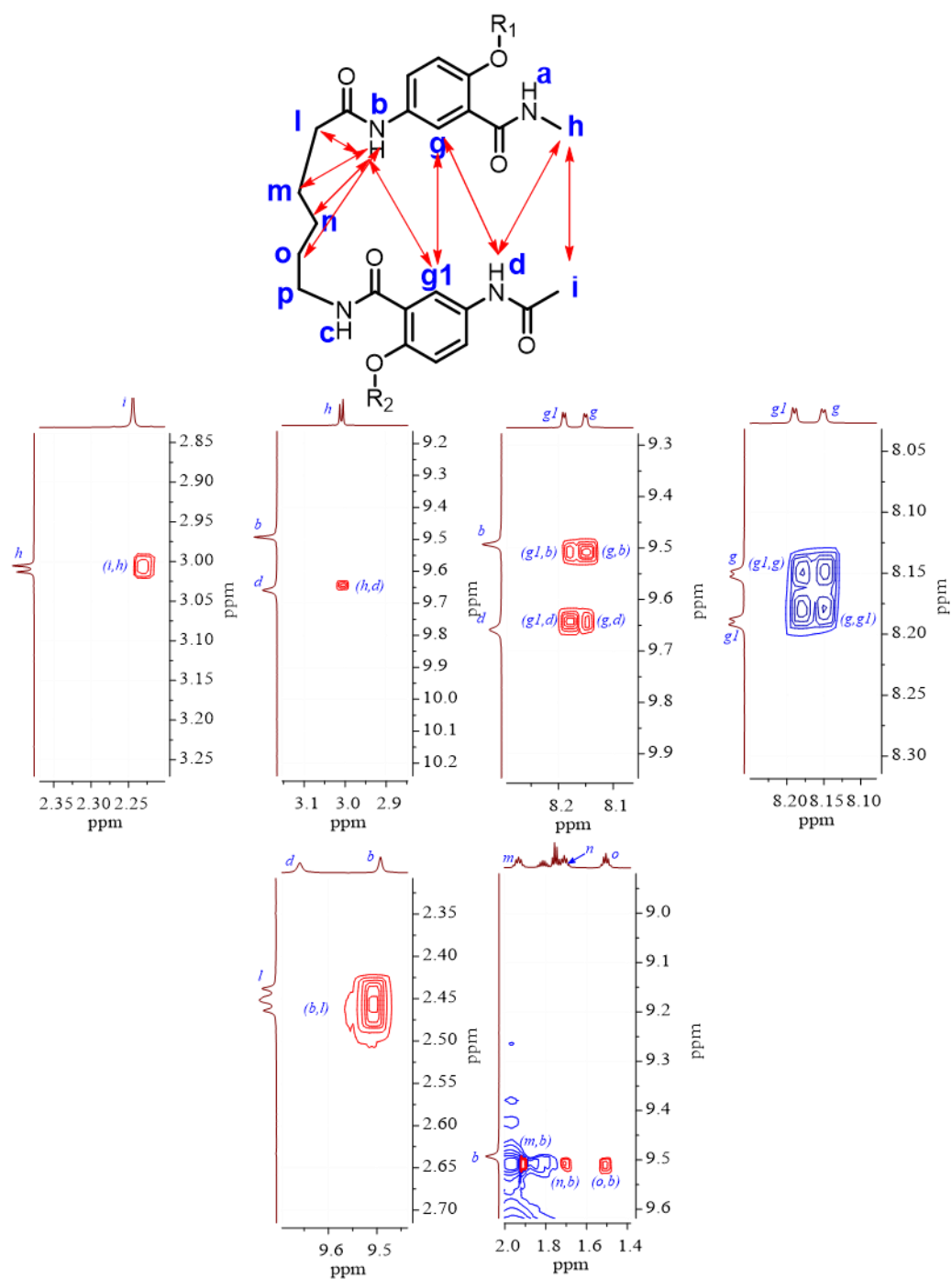
<sup>b</sup> $\Delta\delta_{\text{NH}} = \delta_{(50\% \text{ DMSO})} - \delta_{(0\% \text{ DMSO})}$ .



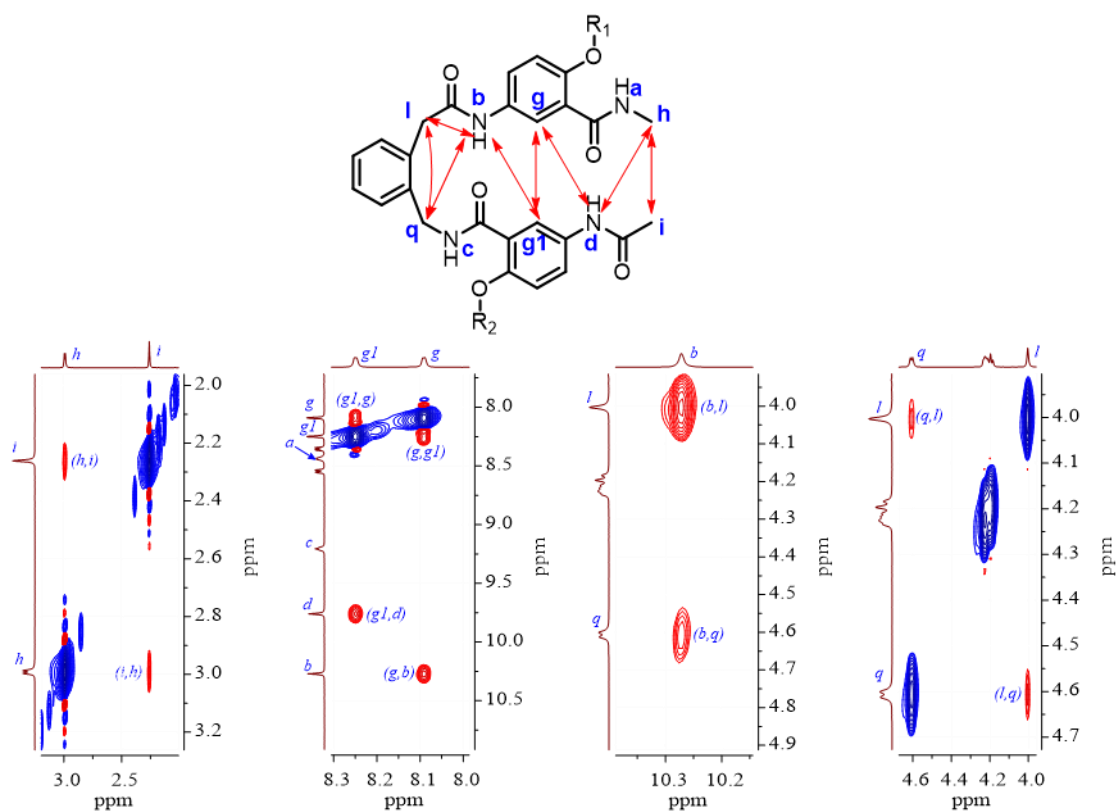
**Figure S1a.** Labeled structure including observed NOE and partial NOESY spectra of **1a** (3 mM, 298 K, 500 MHz, mixing time: 400 ms) in CDCl<sub>3</sub>.



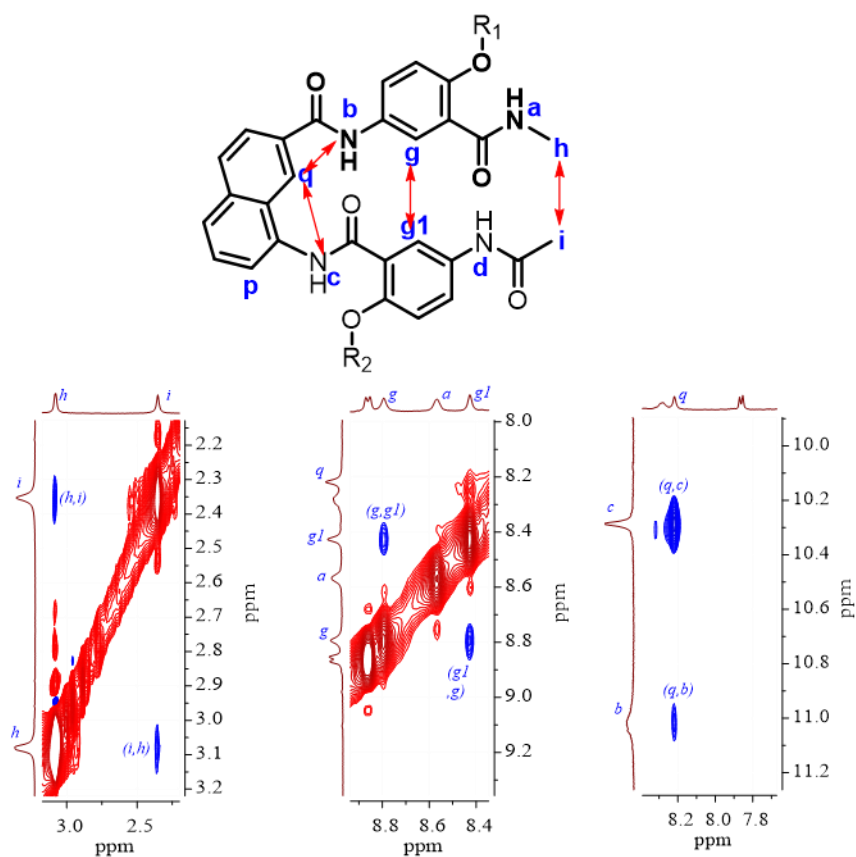
**Figure S1b.** Labeled structure including observed NOE and partial NOESY spectra of **1b** (3 mM, 298 K, 500 MHz, mixing time: 400 ms) in CDCl<sub>3</sub>.



**Figure S1c.** Labeled structure including observed NOE and partial NOESY spectra of **1c** (3 mM, 298 K, 500 MHz, mixing time: 400 ms) in CDCl<sub>3</sub>.



**Figure S1d.** Labeled structure including observed NOE and partial NOESY spectra of **1d** (3 mM, 298 K, 500 MHz, mixing time: 400 ms) in  $\text{CDCl}_3$ .



**Figure S1e.** Labeled structure including observed NOE and partial NOESY spectra of **1e** (3 mM, 298 K, 500 MHz, mixing time: 400 ms) in  $\text{CDCl}_3$ .

mM, 298 K, 500 MHz, mixing time: 400 ms) in CDCl<sub>3</sub>.

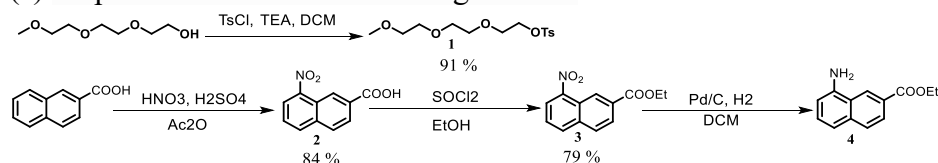
## II. Synthesis and Characterization

### General remarks

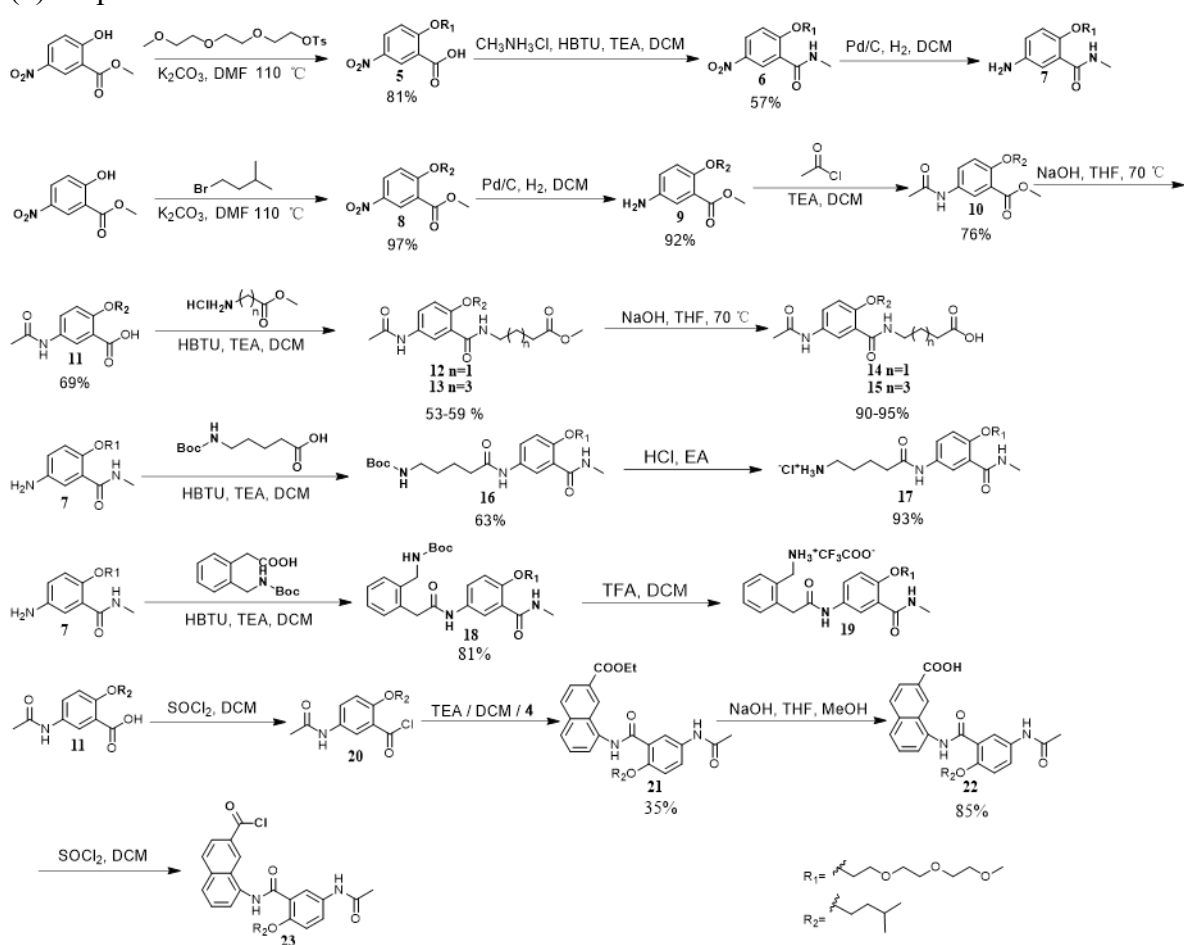
Reagents and solvents were purchased from commercial sources and used without further purification. Column chromatography was carried out on silica gel (300~400 mesh).  $^1\text{H}$  NMR spectra were recorded at 400 MHz and 600 MHz on a Bruker-400 spectrometer and JEOL-400 and 600 spectrometers at ambient temperature.  $^{13}\text{C}$  NMR spectra were measured at 100 MHz and 150 MHz on the same spectrometers. Chemical shifts are reported in parts per million downfield from TMS (tetramethylsilane). Coupling constant in  $^1\text{H}$  NMR are expressed in Hertz. Electrospray ionization high resolution mass spectra (ESI-HRMS) were recorded on a High resolution mass spectra using a waters LCT Premier XE spectrometer (Waters, Milford, MA, USA). The single crystal data were collected on XtaLAB Synergy R, DW system. Data reduction was performed using the CrysAlisPro (Rigaku OD) software packages.

### Scheme S1. Synthesis of 1a-1e

#### (a) Preparation of side chain and rigid linker



#### (b) Preparation of strands of 1a-1e







reaction process by TLC. After completion, washed with 5% citric acid aqueous solution, saturated NaHCO<sub>3</sub> aqueous solution and saturated NaCl aqueous solution, combined the organic layer, dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>, then the crude product was obtained by vacuum distillation, and the purified product as white solid (0.362 g, 54%) was obtained by silica gel column chromatography (PE:EA=1:1). <sup>1</sup>H NMR (600 MHz, Chloroform-*d*) δ 8.28 (s, 1H), 8.14 (dd, *J* = 8.9, 2.8 Hz, 2H), 7.90 (d, *J* = 2.8 Hz, 1H), 6.93 (d, *J* = 9.0 Hz, 1H), 4.12 (t, *J* = 6.5 Hz, 2H), 3.64 (s, 3H), 3.47 (q, *J* = 6.5 Hz, 2H), 2.39 (t, *J* = 7.4 Hz, 2H), 2.15 (s, 3H), 1.91 (p, *J* = 7.2 Hz, 2H), 1.79 (q, *J* = 6.5 Hz, 1H), 1.74 (q, *J* = 6.6 Hz, 2H), 0.98 (d, *J* = 6.3 Hz, 6H). <sup>13</sup>C NMR (101 MHz, Chloroform-*d*) δ 173.47, 169.01, 165.46, 153.56, 132.07, 128.93, 125.57, 123.41, 121.10, 113.03, 67.99, 51.75, 39.25, 37.99, 31.52, 25.39, 24.90, 24.29, 22.59, 0.07. **ESI-MS**: calcd for C<sub>19</sub>H<sub>28</sub>N<sub>2</sub>O<sub>5</sub> (M+H<sup>+</sup>) 365.2071 found 365.3317.

**Compound 13.** In a 100 mL flask, **11** (1.325 g, 5.0 mmol) and dichloromethane were added. Then added HBTU (2.843 g, 7.5 mmol), TEA (1.515 g, 6.0 mmol) and Methyl 6- aminocaproate hydrochloride (0.922 g, 6.0 mmol). Stirred evenly at room temperature, and monitored the reaction process by TLC. After completion, washed with 5% citric acid aqueous solution, saturated NaHCO<sub>3</sub> aqueous solution and saturated NaCl aqueous solution, combined the organic layer, dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>, then the crude product was obtained by vacuum distillation, and the purified product as white solid (1.633 g, 77%) was obtained by silica gel column chromatography (PE:EA=1:1). <sup>1</sup>H NMR (400 MHz, Chloroform-*d*) δ 8.37 (s, 1H), 8.18 (dd, *J* = 9.0, 2.8 Hz, 1H), 8.15 – 8.11 (m, 1H), 7.92 (d, *J* = 2.8 Hz, 1H), 6.95 (d, *J* = 9.0 Hz, 1H), 4.14 (t, *J* = 6.5 Hz, 2H), 3.66 (s, 3H), 3.50 – 3.38 (m, 2H), 2.31 (t, *J* = 7.5 Hz, 2H), 2.27 (s, 3H), 1.83 (q, *J* = 6.5 Hz, 1H), 1.76 (t, *J* = 6.5 Hz, 2H), 1.70 – 1.65 (m, 2H), 1.61 (q, *J* = 7.6 Hz, 2H), 1.47 – 1.38 (m, 2H), 1.00 (d, *J* = 6.4 Hz, 6H). <sup>13</sup>C NMR (151 MHz, Chloroform-*d*) δ 173.99, 168.87, 165.18, 153.44, 132.26, 130.99, 128.93, 125.34, 123.42, 121.43, 112.99, 67.89, 51.59, 39.80, 38.10, 33.98, 29.34, 26.74, 25.33, 24.72, 24.36, 22.58. **ESI-MS**: calcd for C<sub>21</sub>H<sub>32</sub>N<sub>2</sub>O<sub>5</sub> (M+H<sup>+</sup>) 393.2384 found 393.3739.

**Compound 14.** In a 100 mL flask, **12** (0.336 g, 1 mmol) was dissolved in tetrahydrofuran, to which 2 *N* NaOH (1.0 mL, 2.0 mmol) was added. The mixture was heated under reflux in oil bath, and monitored the reaction process by TLC. Upon completion, remove the tetrahydrofuran under vacuum, the aqueous layer was acidulated by addition of 2 *N* HCl to pH 4 extracted with EtOAc, dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>, concentrated to remove the solvent, and the crude product as yellow oil was obtained. This compound was used without further purification.

**Compound 15.** The compound **13** (1.503 g, 4.5 mmol) was dissolved in tetrahydrofuran, to which 2 *N* NaOH (5.6 mL, 9.2 mmol) was added. The mixture was heated under reflux in oil bath and monitored the reaction process by TLC. After removed the tetrahydrofuran under vacuum, the aqueous layer was acidulated by addition of 2 *N* HCl to pH 4 followed by filtration to obtain the crude product as white solid. This compound was used without further purification.

**Compound 16.** In a 100 mL flask, Boc-aminopentanoic (0.521 g, 2.4 mmol) and dichloromethane were added. Then added HBTU (1.137 g, 1.5 mmol), TEA (0.404 g, 2.0 mmol) and **7** (0.684 g, 2.0 mmol). Stirred evenly at room temperature, and monitored the reaction process by TLC. After completion, washed with 5% citric acid aqueous solution, saturated

NaHCO<sub>3</sub> aqueous solution and saturated NaCl aqueous solution, combined the organic layer, dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>, then the crude product was obtained by vacuum distillation, and the purified product as white solid (0.639 g, 63 %) was obtained by silica gel column chromatography (PE:Acetone=1:1). <sup>1</sup>H NMR (600 MHz, DMSO-*d*<sub>6</sub>) δ 9.87 (s, 1H), 8.22 (d, *J* = 4.7 Hz, 1H), 7.98 (d, *J* = 2.8 Hz, 1H), 7.78 (dd, *J* = 8.9, 2.8 Hz, 1H), 7.11 (d, *J* = 9.0 Hz, 1H), 6.79 (t, *J* = 5.6 Hz, 1H), 4.25 – 4.16 (m, 2H), 3.81 – 3.77 (m, 2H), 3.64 – 3.60 (m, 2H), 3.58 – 3.54 (m, 2H), 3.53 – 3.49 (m, 2H), 3.45 – 3.39 (m, 2H), 3.22 (s, 3H), 2.92 (q, *J* = 6.6 Hz, 2H), 2.82 (d, *J* = 4.7 Hz, 3H), 2.26 (t, *J* = 7.4 Hz, 2H), 1.55 (p, *J* = 7.5 Hz, 2H), 1.37 (s, 9H). <sup>13</sup>C NMR (151 MHz, DMSO-*d*<sub>6</sub>) δ 171.40, 165.30, 152.47, 133.62, 123.57, 123.19, 122.09, 114.96, 77.87, 71.79, 70.41, 70.37, 70.13, 69.15, 69.07, 58.57, 36.47, 29.68, 28.81, 26.70, 23.06. **ESI-MS**: calcd for C<sub>27</sub>H<sub>30</sub>N<sub>2</sub>O<sub>5</sub> (M+H<sup>+</sup>) 512.2967 found 512.4729.

**Compound 17.** The compound **16** (0.639 g, 0.5 mmol) was treated with hydrochloric solution of ethyl acetic (2 *N*, 20 mL), then the mixture was stirred at room temperature for 1 h. Removed the solvent to obtain compound **17** as a white solid. This compound was used without further purification.

**Compound 18.** In a 100 mL flask, 2-(Boc-aminomethyl) phenylacetic acid (0.220 g, 0.83 mmol) and dichloromethane were added. Then added HATU (0.456 g, 1.2 mmol), TEA (0.101 g, 1.0 mmol) and **7** (0.285 g, 0.83 mmol). Stirred evenly at room temperature, and monitored the reaction process by TLC. After completion, washed with 5% citric acid aqueous solution, saturated NaHCO<sub>3</sub> aqueous solution and saturated NaCl aqueous solution, combined the organic layer, dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>, then the crude product was obtained by vacuum distillation, and the purified product as white flaky solid (0.409 g, 88 %) was obtained by silica gel column chromatography (PE:Acetone=1:1). <sup>1</sup>H NMR (600 MHz, DMSO-*d*<sub>6</sub>) δ 10.19 (s, 1H), 8.23 (d, *J* = 4.8 Hz, 1H), 7.99 (d, *J* = 2.8 Hz, 1H), 7.79 (dd, *J* = 8.9, 2.8 Hz, 1H), 7.28 – 7.25 (m, 2H), 7.23 (dd, *J* = 3.9, 2.3 Hz, 2H), 7.21 (d, *J* = 7.2 Hz, 1H), 7.13 (d, *J* = 9.0 Hz, 1H), 4.22 (t, *J* = 4.8 Hz, 4H), 3.81 – 3.77 (m, 2H), 3.71 (s, 2H), 3.64 – 3.59 (m, 2H), 3.57 – 3.53 (m, 2H), 3.52 – 3.49 (m, 2H), 3.44 – 3.37 (m, 2H), 3.21 (s, 3H), 2.82 (d, *J* = 4.7 Hz, 3H), 1.38 (s, 9H). <sup>13</sup>C NMR (151 MHz, DMSO-*d*<sub>6</sub>) δ 169.34, 165.24, 152.69, 134.26, 133.41, 130.67, 127.89, 127.25, 123.67, 123.24, 122.24, 115.02, 78.36, 71.79, 70.41, 70.37, 70.13, 69.15, 69.06, 58.57, 41.70, 28.78, 26.71. **ESI-MS**: calcd for C<sub>29</sub>H<sub>41</sub>N<sub>3</sub>O<sub>8</sub> (M+H<sup>+</sup>) 560.2867 found 560.4871.

**Compound 19.** In a 100 mL flask, **18** (0.548 g, 1.0 mmol) and dichloromethane were added. Then added trifluoroacetic acid dropwise. The reaction mixture was stirred at room temperature and detected by TLC. Upon completion, removed the solvent to afford **19** as yellow oil. This compound was used without further purification.

**Compound 20.** **11** (0.398 g, 1.5 mmol) and dichloromethane were added into the 50 mL flask, While stirring the resultant mixture at room temperature, oxalyl chloride and a drop of *N,N*-Dimethylformamide were added dropwise. the reaction process was monitored by TLC. After completion, removed the solvent to afford crude product. This compound was used without further purification.

**Compound 21.** In a 100 mL flask, **4** (0.408 g, 1.9 mmol) and dichloromethane were added. Then added triethylamine (0.247 g, 2.2 mmol) and **20** (0.601 g, 2.2 mmol) The mixture was stirred at room temperature and monitored the reaction process by TLC. After completion,

washed with 5% citric acid aqueous solution and saturated NaCl aqueous solution, combined the organic layer, dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>, then the crude product was obtained by vacuum distillation, and the purified product as white flaky solid (0.562 g, 66 %) was obtained by silica gel column chromatography (DCM:MeOH=150:1~60:1). <sup>1</sup>H NMR (600 MHz, Chloroform-*d*) δ 10.66 (s, 1H), 8.81 (d, *J* = 1.5 Hz, 1H), 8.26 (dd, *J* = 8.9, 2.9 Hz, 1H), 8.22 (d, *J* = 7.5 Hz, 1H), 8.17 (s, 1H), 8.13 (d, *J* = 2.9 Hz, 1H), 8.10 (dd, *J* = 8.6, 1.6 Hz, 1H), 7.93 (d, *J* = 8.5 Hz, 1H), 7.74 (d, *J* = 8.2 Hz, 1H), 7.62 (t, *J* = 7.8 Hz, 1H), 7.08 (d, *J* = 9.0 Hz, 1H), 4.42 (q, *J* = 7.2 Hz, 2H), 4.39 (t, *J* = 7.0 Hz, 2H), 1.85 (q, *J* = 7.0 Hz, 2H), 1.81 (s, 3H), 1.74-1.68 (m, 1H), 1.42 (t, *J* = 7.1 Hz, 3H), 0.82 (d, *J* = 6.6 Hz, 6H). <sup>13</sup>C NMR (151 MHz, Chloroform-*d*) δ 168.83, 166.56, 163.97, 153.47, 136.27, 134.49, 132.58, 129.07, 128.47, 128.12, 126.51, 125.99, 125.54, 125.21, 124.27, 123.71, 121.47, 113.37, 68.69, 61.32, 37.53, 25.25, 24.03, 22.43, 14.49. ESI-MS: calcd for C<sub>27</sub>H<sub>30</sub>N<sub>2</sub>O<sub>5</sub> (M+H<sup>+</sup>) 463.2228 found 463.3814.

**Compound 22.** The compound **21** (0.110 g, 0.25 mmol) was dissolved in tetrahydrofuran, to which 2 *N* NaOH (0.3 mL, 0.5 mmol) and a few methanol was added. The mixture was heated under reflux in oil bath and monitored the reaction process by TLC. After removed the tetrahydrofuran under vacuum, the aqueous layer was acidulated by addition of 2 *N* HCl to pH 3 followed by filtration to obtain the crude product as white solid. This compound was used without further purification.

**Compound 23.** **22** (0.102 g, 0.23 mmol) and dichloromethane were added into the 50 mL round bottom flask. While the resultant mixture was stirred in an ice bath, the dichloromethane solution of sulfoxide chloride was dripped slowly from the constant pressure funnel. Then removed the ice bath, stirred the reaction solution in oil bath for refluxing and monitored the reaction process by TLC. After completion, removed the solvent to afford crude product as yellow oil. This compound was used without further purification.

**1a.** In a 100 mL flask, **14** (0.869 g, 2.7 mmol) and dichloromethane were added. Then added HBTU (1.737 g, 3.6 mmol), TEA (0.606 g, 6.0 mmol) and **7** (0.989 g, 3.0 mmol). Stirred evenly at room temperature, and monitored the reaction process by TLC. After completion, washed with 5% citric acid aqueous solution, saturated NaHCO<sub>3</sub> aqueous solution and saturated NaCl aqueous solution, combined the organic layer, dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>, then the crude product was obtained by vacuum distillation, and the purified product as white solid (0.237 g, 21 %) was obtained by silica gel column chromatography (PE:Acetone=1:2). <sup>1</sup>H NMR (400 MHz, Chloroform-*d*) δ 9.87 (s, 1H), 9.62 (s, 1H), 8.35 (d, *J* = 4.7 Hz, 1H), 8.30 – 8.17 (m, 3H), 7.94 (d, *J* = 2.7 Hz, 1H), 7.84 (d, *J* = 2.8 Hz, 1H), 6.79 (d, *J* = 9.1 Hz, 1H), 6.74 (d, *J* = 8.9 Hz, 1H), 4.20 (t, *J* = 4.6 Hz, 2H), 4.07 (t, *J* = 6.3 Hz, 2H), 3.88 (t, *J* = 5.7 Hz, 2H), 3.72 (t, *J* = 6.1 Hz, 2H), 3.67 (t, *J* = 6.0 Hz, 2H), 3.62 (t, *J* = 6.0 Hz, 2H), 3.58 – 3.47 (m, 4H), 3.34 (s, 3H), 2.94 (d, *J* = 4.7 Hz, 3H), 2.48 (t, *J* = 6.6 Hz, 2H), 2.12 (s, 3H), 2.03-2.00 (m, 2H), 1.78 (t, *J* = 6.0 Hz, 3H), 0.98 (d, *J* = 6.0 Hz, 6H). <sup>13</sup>C NMR (151 MHz, Chloroform-*d*) δ 171.65, 169.13, 166.03, 165.88, 153.05, 152.50, 133.74, 133.14, 124.53, 124.00, 122.90, 122.69, 121.67, 120.90, 113.61, 112.53, 72.00, 70.77, 70.70, 69.38, 68.56, 67.92, 59.09, 39.96, 37.89, 35.41, 26.76, 25.65, 25.43, 24.26, 22.66. HRMS (ESI), calcd C<sub>33</sub>H<sub>48</sub>N<sub>4</sub>O<sub>9</sub> [M+H]<sup>+</sup> 645.3500 found 645.3498.

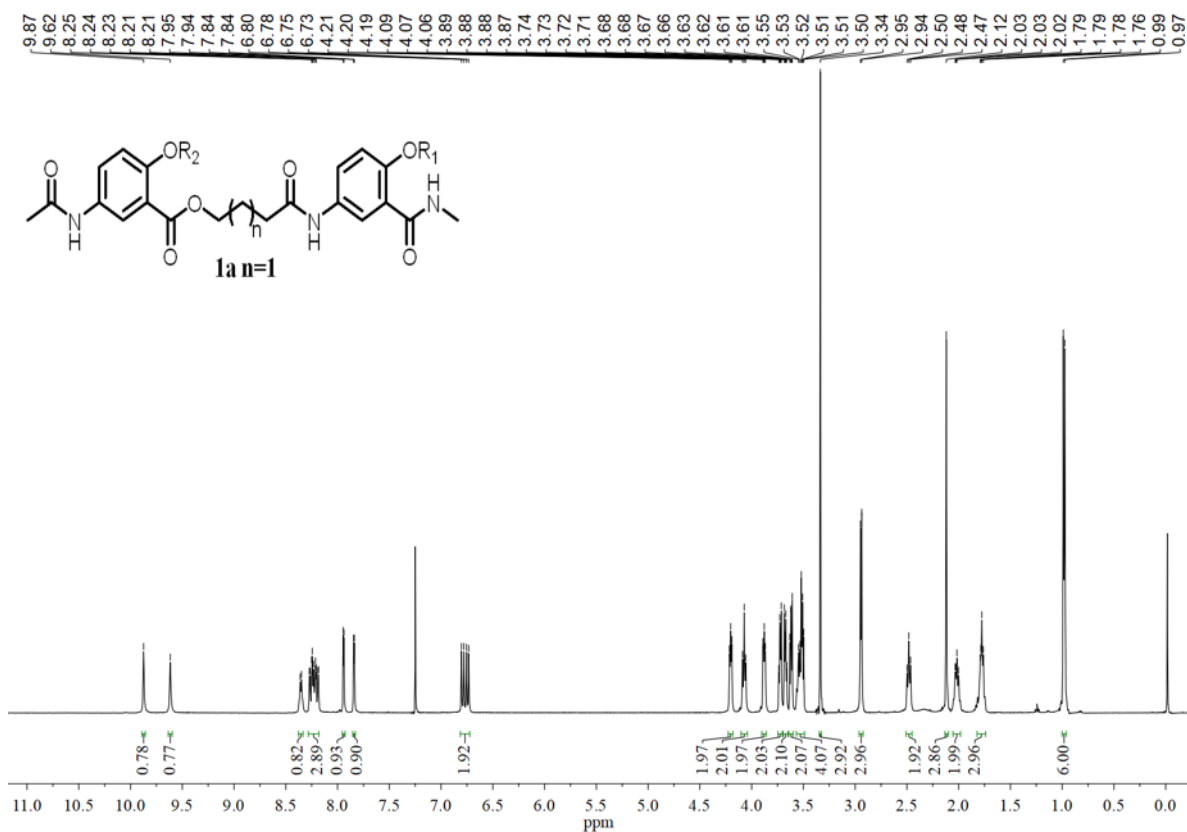
**1b.** In a 100 mL flask, **11** (0.582 g, 1.3 mmol) and dichloromethane were added. Then added HBTU (0.758 g, 2.0 mmol), TEA (0.405 g, 4.0 mmol) and **17** (0.561 g, 1.6 mmol). Stirred evenly at room temperature, and monitored the reaction process by TLC. After completion, washed with 5% citric acid aqueous solution, saturated NaHCO<sub>3</sub> aqueous solution and saturated NaCl aqueous solution, combined the organic layer, dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>, then the crude product was obtained by vacuum distillation, and the purified product as white solid (0.155 g, 26 %) was obtained by silica gel column chromatography (PE:Acetone=1:2). <sup>1</sup>H NMR (600 MHz, Chloroform-*d*) δ 9.88 (s, 1H), 9.65 (s, 1H), 8.67 (t, *J* = 6.6 Hz, 1H), 8.53 (dd, *J* = 9.0, 2.8 Hz, 1H), 8.51 – 8.46 (m, 1H), 8.32 (dd, *J* = 8.9, 2.8 Hz, 1H), 8.28 (d, *J* = 2.8 Hz, 1H), 8.04 (d, *J* = 2.8 Hz, 1H), 6.96 (d, *J* = 9.0 Hz, 1H), 6.92 (d, *J* = 9.0 Hz, 1H), 4.24 (t, *J* = 4.5 Hz, 2H), 4.17 (t, *J* = 6.4 Hz, 2H), 3.88 (t, *J* = 4.5 Hz, 2H), 3.73 (t, *J* = 5.8 Hz, 2H), 3.68 (t, *J* = 5.8 Hz, 2H), 3.63 (t, *J* = 5.8 Hz, 2H), 3.59 (d, *J* = 5.8 Hz, 2H), 3.52 (t, *J* = 4.8 Hz, 2H), 3.35 (s, 3H), 3.03 (d, *J* = 4.7 Hz, 3H), 2.56 (t, *J* = 8.1 Hz, 2H), 2.30 (s, 3H), 1.82-1.74 (m, 5H), 1.63 (p, *J* = 6.0 Hz, 2H), 1.00 (d, *J* = 6.4 Hz, 6H). <sup>13</sup>C NMR (151 MHz, Chloroform-*d*) δ 172.88, 169.43, 166.77, 166.34, 153.48, 152.18, 134.44, 133.24, 126.27, 123.93, 123.24, 122.23, 121.73, 120.44, 114.24, 113.04, 72.01, 70.77, 70.71, 69.30, 68.79, 68.10, 59.09, 38.07, 36.50, 36.39, 29.60, 27.11, 25.52, 24.41, 23.15, 22.64. HRMS (ESI), calcd C<sub>34</sub>H<sub>50</sub>N<sub>4</sub>O<sub>9</sub> [M+H]<sup>+</sup> 659.3656 found 659.3659.

**1c.** In a 100 mL flask, **15** (0.312 g, 0.8 mmol) and dichloromethane were added. Then added HBTU (0.455 g, 1.2 mmol), TEA (0.242 g, 2.4 mmol) and **7** (0.342 g, 1.0 mmol). Stirred evenly at room temperature, and monitored the reaction process by TLC. After completion, washed with 5% citric acid aqueous solution, saturated NaHCO<sub>3</sub> aqueous solution and saturated NaCl aqueous solution, combined the organic layer, dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>, then the crude product was obtained by vacuum distillation, and the purified product as white solid (0.215 g, 35 %) was obtained by silica gel column chromatography (PE:Acetone=2:1~1:3). <sup>1</sup>H NMR (600 MHz, Chloroform-*d*) δ 9.66 (s, 1H), 9.49 (s, 1H), 8.45 (d, *J* = 4.9 Hz, 1H), 8.40-8.40 (m, 1H), 8.19 (d, *J* = 2.8 Hz, 1H), 8.15 (d, *J* = 2.8 Hz, 1H), 6.94 (d, *J* = 1.4 Hz, 1H), 6.93 (d, *J* = 1.4 Hz, 1H), 4.30 – 4.23 (m, 2H), 4.14 (t, *J* = 6.5 Hz, 2H), 3.90 – 3.87 (m, 2H), 3.75 – 3.72 (m, 2H), 3.71 – 3.66 (m, 2H), 3.64 – 3.61 (m, 2H), 3.54 – 3.51 (m, 2H), 3.46 (q, *J* = 6.9 Hz, 2H), 3.35 (s, 3H), 3.01 (d, *J* = 4.7 Hz, 3H), 2.51 – 2.39 (m, 2H), 2.24 (s, 3H), 1.93 (t, *J* = 7.7 Hz, 2H), 1.82 (p, *J* = 6.6 Hz, 1H), 1.75 (q, *J* = 6.7 Hz, 2H), 1.72 – 1.69 (m, 2H), 1.51 (t, *J* = 6.9 Hz, 2H), 1.00 (d, *J* = 6.6 Hz, 6H). <sup>13</sup>C NMR (151 MHz, Chloroform-*d*) δ 172.55, 169.32, 166.27, 165.84, 153.14, 152.59, 133.90, 133.23, 125.10, 124.48, 123.31, 122.92, 122.15, 121.04, 114.17, 112.93, 72.00, 70.76, 70.71, 69.30, 68.77, 67.95, 59.10, 38.52, 38.09, 36.90, 28.23, 26.98, 25.43, 25.40, 24.40, 24.22, 22.63. HRMS (ESI), calcd C<sub>35</sub>H<sub>52</sub>N<sub>4</sub>O<sub>9</sub> [M+H]<sup>+</sup> 673.3813 found 673.3811.

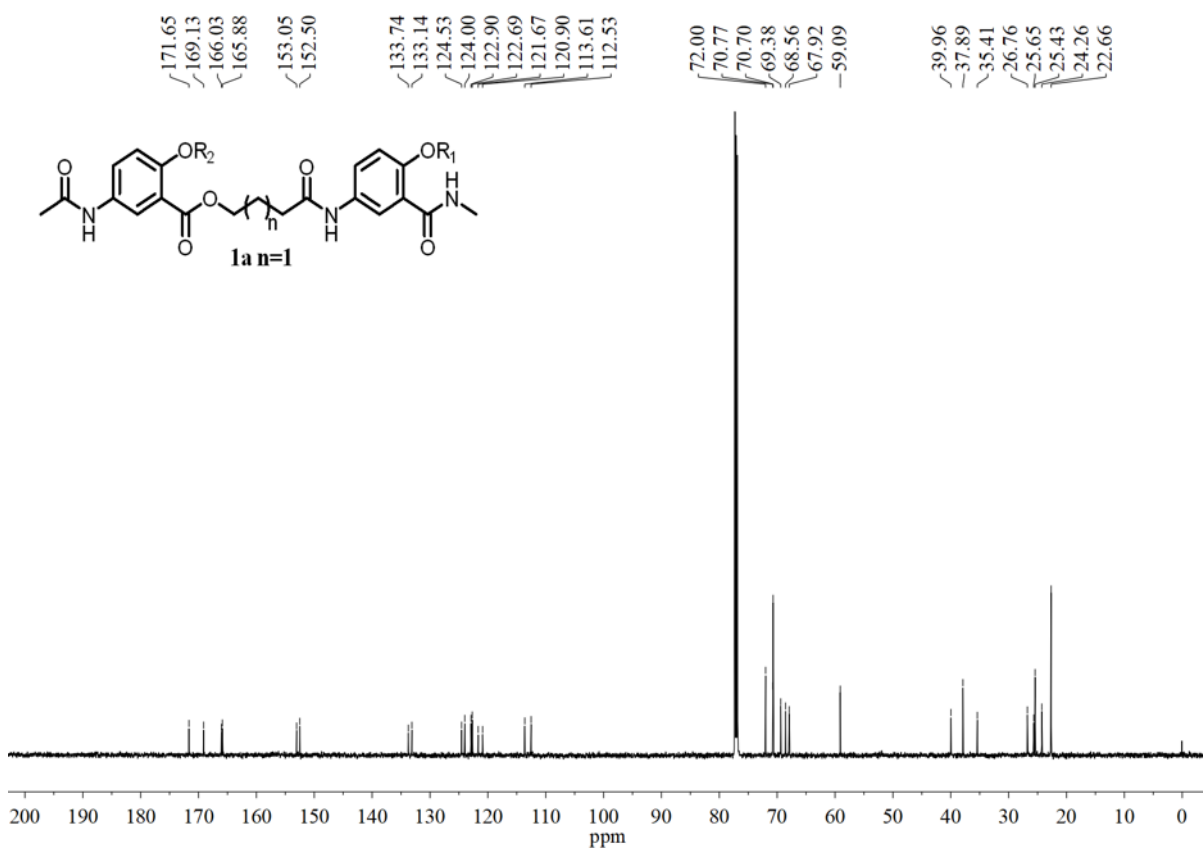
**1d.** In a 100 mL flask, **19** (0.573 g, 1.0 mmol) and dichloromethane were added. Then added triethylamine (0.505 g, 5.0 mmol) and **20** (0.398 g, 1.5 mmol). The mixture was stirred at room temperature and monitored the reaction process by TLC. After completion, washed with 5% citric acid aqueous solution and saturated NaCl aqueous solution, combined the organic layer, dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>, then the crude product was obtained by vacuum distillation. The crude product was further purified by silica gel column chromatography (1<sup>st</sup>: DCM:MeOH=60:1~40:1; 2<sup>nd</sup>: DCM:MeOH=45:1~30:1) and ether recrystallization to afford purified product as white flaky solid (0.040 g, 7 %). <sup>1</sup>H NMR (600 MHz, Chloroform-*d*) δ

10.28 (s, 1H), 9.82 (s, 1H), 9.17 (s, 1H), 8.52 (dd,  $J=9.0, 2.7$  Hz, 1H), 8.44 (d,  $J=5.1$  Hz, 1H), 8.35 (dd,  $J=9.1, 2.8$  Hz, 1H), 8.23 (d,  $J=2.7$  Hz, 1H), 8.08 (d,  $J=2.7$  Hz, 1H), 7.68 – 7.51 (m, 1H), 7.25 (d,  $J=3.7$  Hz, 1H), 7.23 – 7.21 (m, 2H), 6.93 (d,  $J=9.0$  Hz, 1H), 6.88 (d,  $J=9.0$  Hz, 1H), 4.61 (d,  $J=6.0$  Hz, 2H), 4.22 (t,  $J=4.5$  Hz, 2H), 4.19 (t,  $J=6.6$  Hz, 2H), 4.00 (s, 2H), 3.89 – 3.84 (m, 2H), 3.72 (t,  $J=5.7$  Hz, 2H), 3.69 – 3.64 (m, 2H), 3.61 (t,  $J=4.8$  Hz, 2H), 3.55 – 3.47 (m, 2H), 3.34 (d,  $J=1.2$  Hz, 3H), 2.99 (d,  $J=4.7$  Hz, 3H), 2.26 (s, 3H), 1.89 (p,  $J=6.6$  Hz, 1H), 1.82 (q,  $J=6.7$  Hz, 2H), 1.05 (d,  $J=6.5$  Hz, 6H).  $^{13}\text{C}$  NMR (151 MHz, Chloroform-*d*)  $\delta$  169.99, 169.27, 166.36, 165.67, 153.46, 152.22, 136.21, 134.50, 134.05, 133.33, 129.74, 129.64, 127.93, 127.41, 125.82, 123.75, 123.39, 122.11, 120.02, 114.03, 112.82, 71.99, 70.76, 70.70, 69.30, 68.71, 67.99, 59.10, 40.82, 40.57, 38.17, 27.14, 25.51, 24.52, 22.70. HRMS (ESI), calcd  $\text{C}_{38}\text{H}_{50}\text{N}_4\text{O}_9$   $[\text{M}+\text{H}]^+$  707.3656 found 707.3647.

**1e.** In a 100 mL flask, **7** (0.220 g, 0.66 mmol) and dichloromethane were added. Then added triethylamine (0.098 g, 0.9 mmol) and **23** (0.298 g, 0.66 mmol). The mixture was stirred at room temperature and monitored the reaction process by TLC. After completion, washed with 5% citric acid aqueous solution and saturated NaCl aqueous solution, combined the organic layer, dried over anhydrous  $\text{Na}_2\text{SO}_4$ , then the crude product was obtained by vacuum distillation. The crude product was further purified by silica gel column chromatography (DCM:MeOH=70:1~40:1) and ether recrystallization to afford purified product as white flaky solid (0.024 g, 5 %).  $^1\text{H}$  NMR (600 MHz, Chloroform-*d*)  $\delta$  11.06 (s, 1H), 10.28 (s, 1H), 9.79 (s, 1H), 8.85 (d,  $J=8.9$  Hz, 1H), 8.81 (s, 1H), 8.56 (s, 1H), 8.42 (s, 1H), 8.28 (d,  $J=8.8$  Hz, 1H), 8.21 (s, 1H), 7.85 (d,  $J=8.5$  Hz, 1H), 7.62 (d,  $J=7.4$  Hz, 1H), 7.49 (d,  $J=8.6$  Hz, 1H), 7.10 (d,  $J=8.8$  Hz, 2H), 6.98 (t,  $J=7.8$  Hz, 1H), 6.68 (s, 1H), 4.35 (t,  $J=4.4$  Hz, 2H), 4.17 (s, 2H), 3.94 (t,  $J=4.5$  Hz, 2H), 3.77 (t,  $J=4.1$  Hz, 2H), 3.73 – 3.69 (m, 2H), 3.65 (t,  $J=4.6$  Hz, 2H), 3.53 (t,  $J=4.6$  Hz, 2H), 3.36 (s, 3H), 3.07 (d,  $J=4.5$  Hz, 3H), 2.33 (s, 3H), 1.75 (d,  $J=7.2$  Hz, 2H), 1.44 – 1.35 (m, 1H), 0.52 (d,  $J=6.7$  Hz, 6H).  $^{13}\text{C}$  NMR (151 MHz, Chloroform-*d*)  $\delta$  168.67, 166.49, 166.33, 164.69, 152.94, 134.53, 134.43, 133.06, 132.29, 128.46, 126.68, 126.22, 125.28, 125.18, 124.95, 123.94, 123.09, 122.46, 121.19, 120.68, 119.48, 114.29, 112.15, 72.03, 70.81, 70.76, 69.36, 68.85, 68.19, 59.13, 37.60, 27.18, 25.15, 24.73, 22.03. HRMS (ESI), calcd  $\text{C}_{40}\text{H}_{48}\text{N}_4\text{O}_9$   $[\text{M}+\text{H}]^+$  729.3500 found 729.3502.



**Figure S2.**  $^1\text{H NMR}$  spectrum of compound **1a** ( $\text{CDCl}_3$ , 298 K, 400 MHz)



**Figure S3.**  $^{13}\text{C NMR}$  spectrum of compound **1a** ( $\text{CDCl}_3$ , 298 K, 151 MHz)

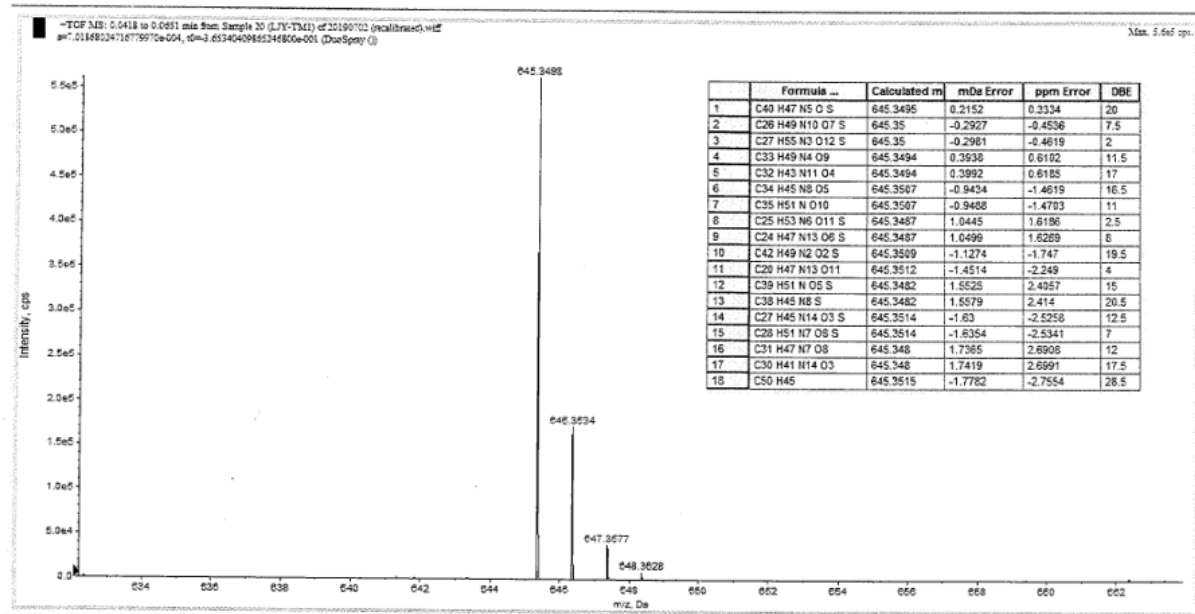


Figure S4. MS spectrum of compound **1a**

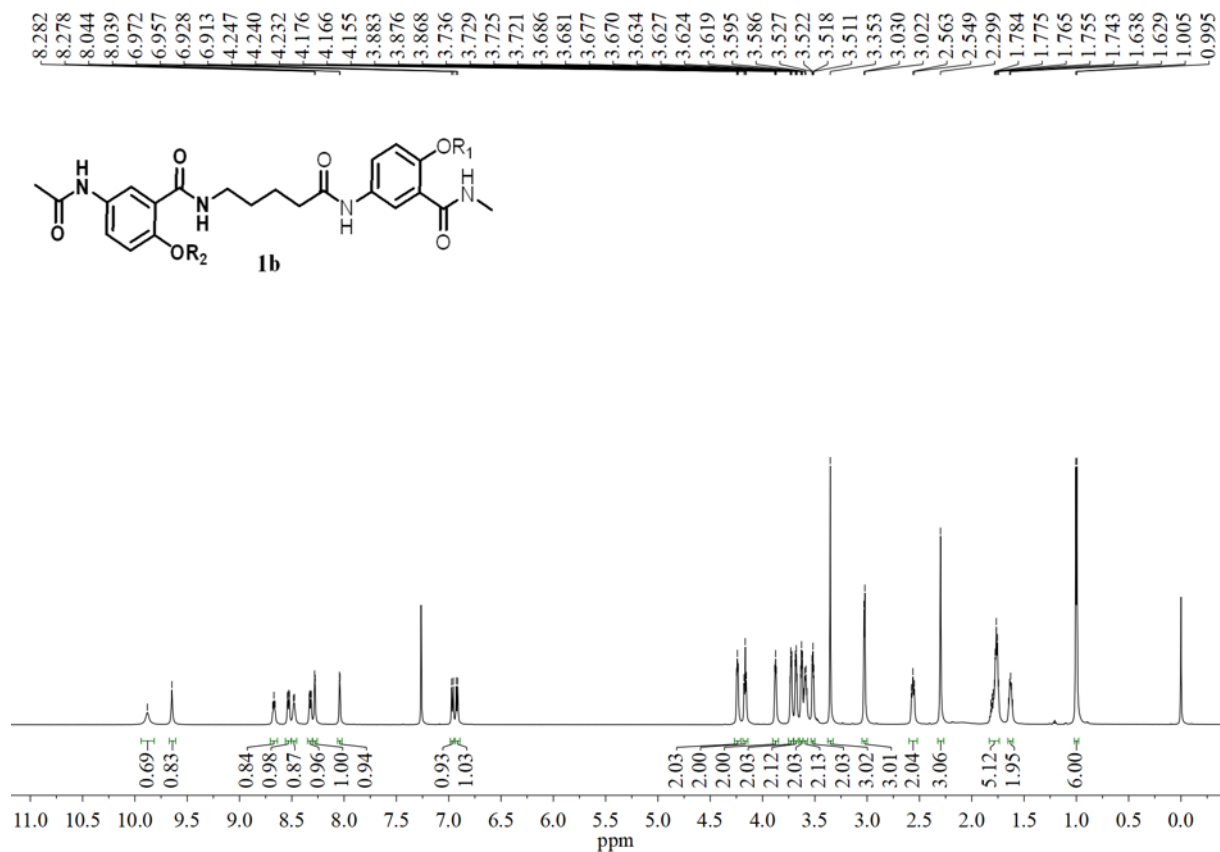
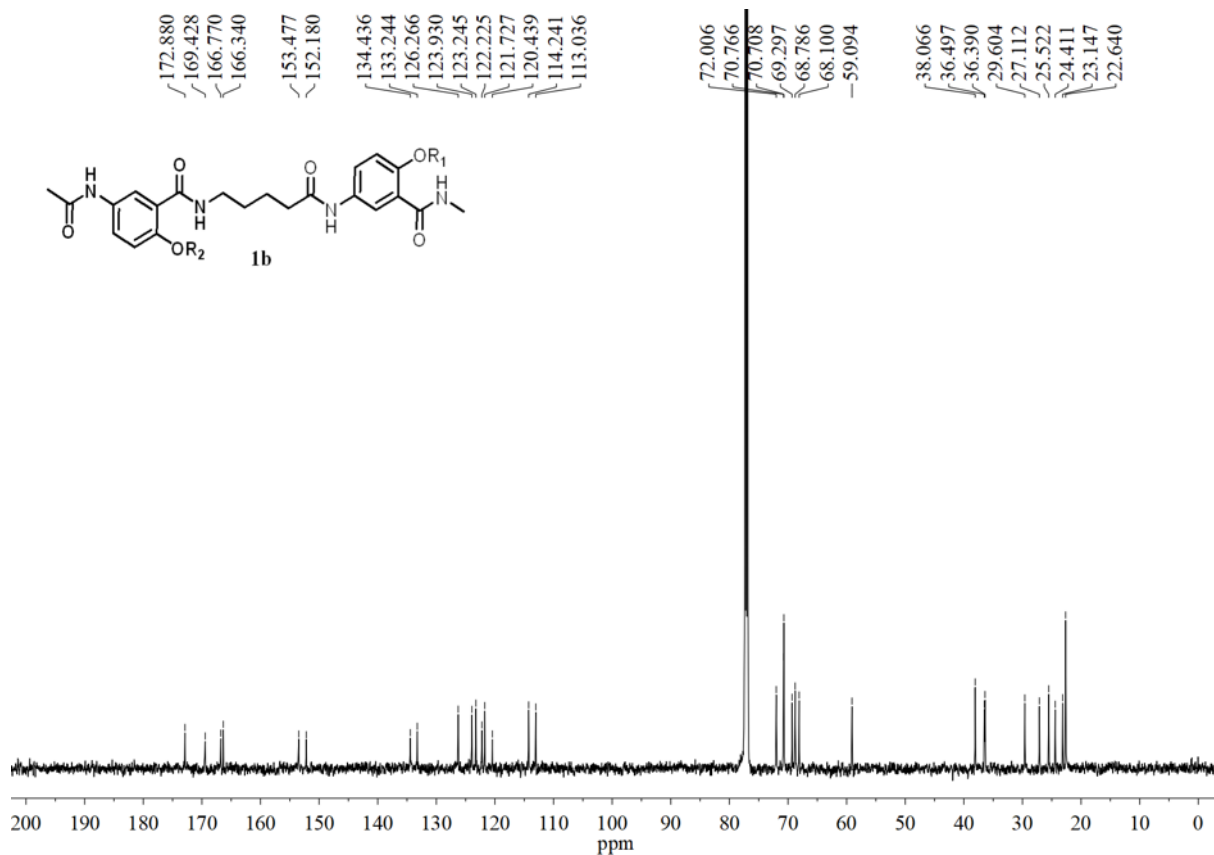
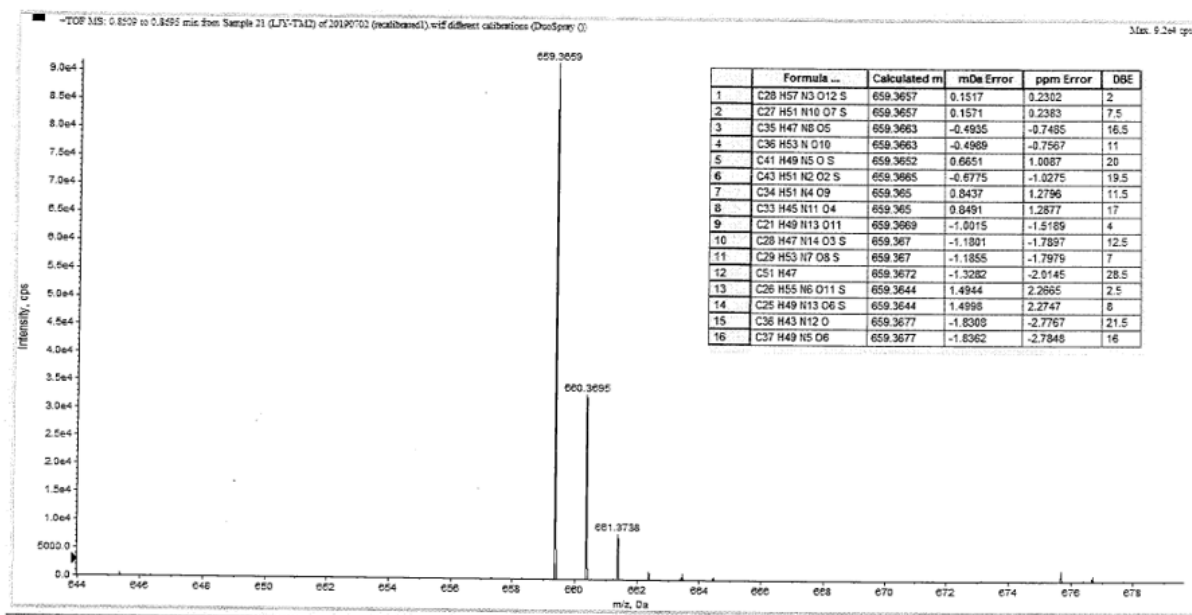


Figure S5. <sup>1</sup>H NMR spectrum of compound **1b** (CDCl<sub>3</sub>, 298 K, 600 MHz)

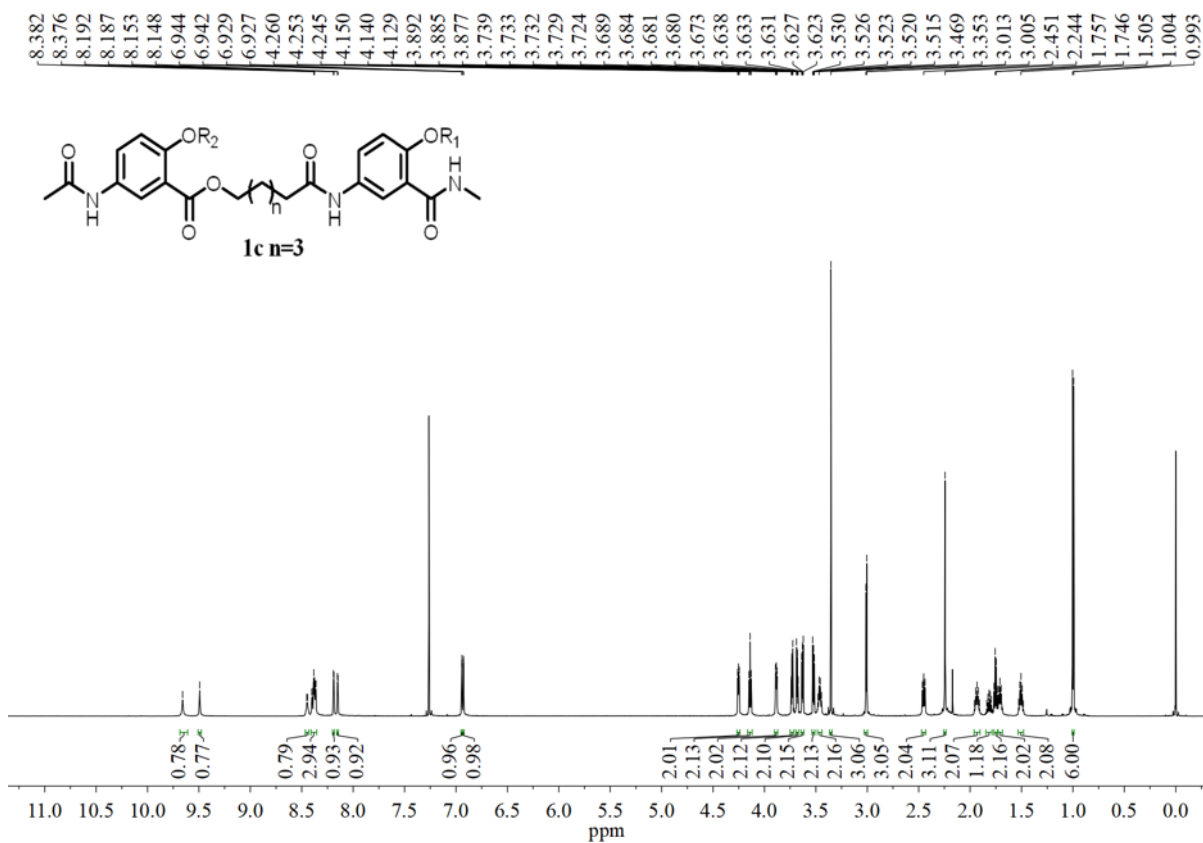


**Figure S6.** <sup>13</sup>C NMR spectrum of compound **1b** (CDCl<sub>3</sub>, 298 K, 151 MHz)

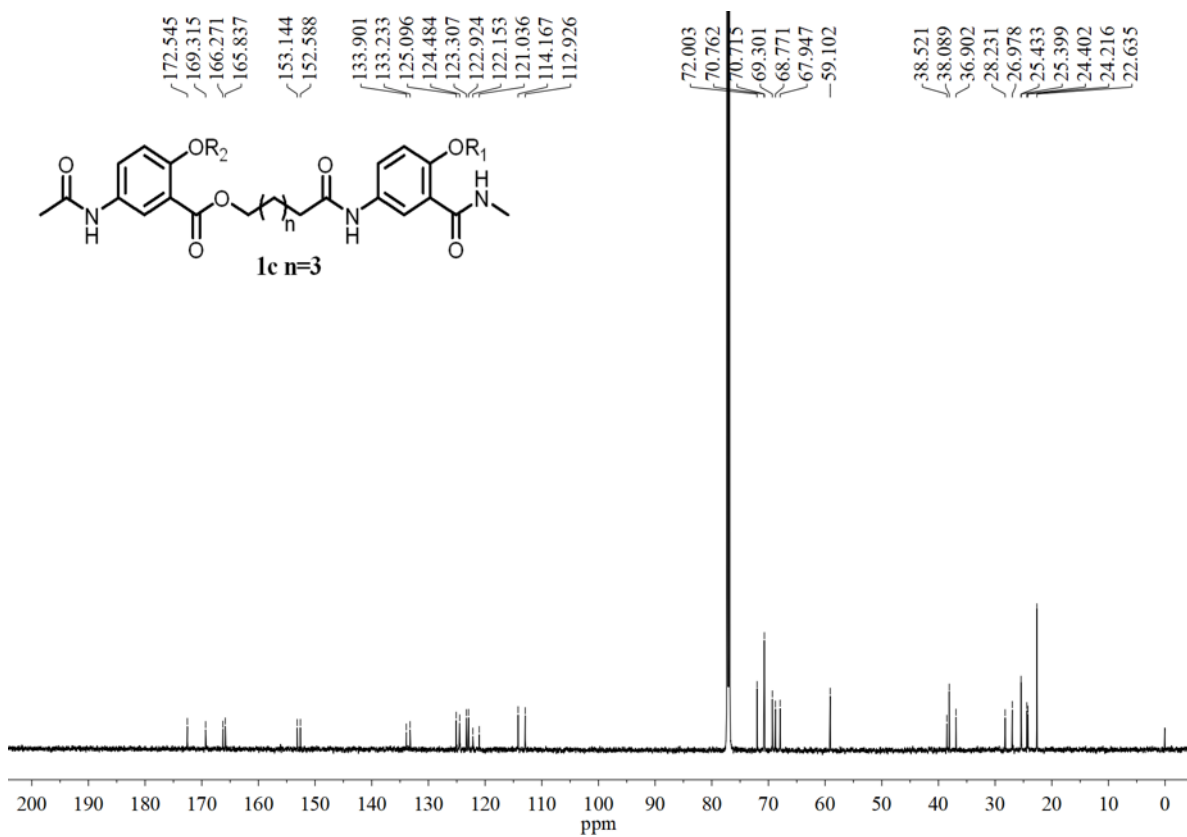


**Figure S7.** MS spectrum of compound **1b**



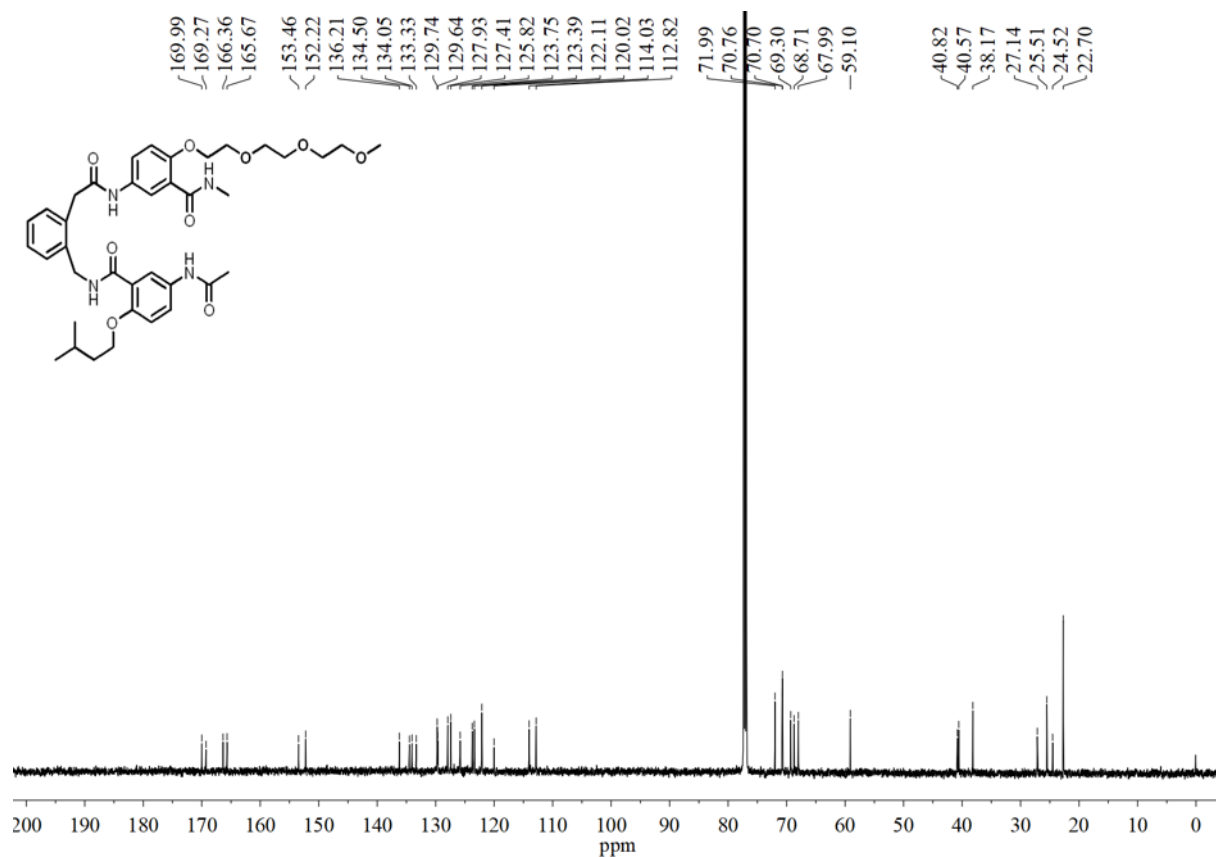


**Figure S8.**  $^1\text{H NMR}$  spectrum of compound **1c** ( $\text{CDCl}_3$ , 298 K, 600 MHz)

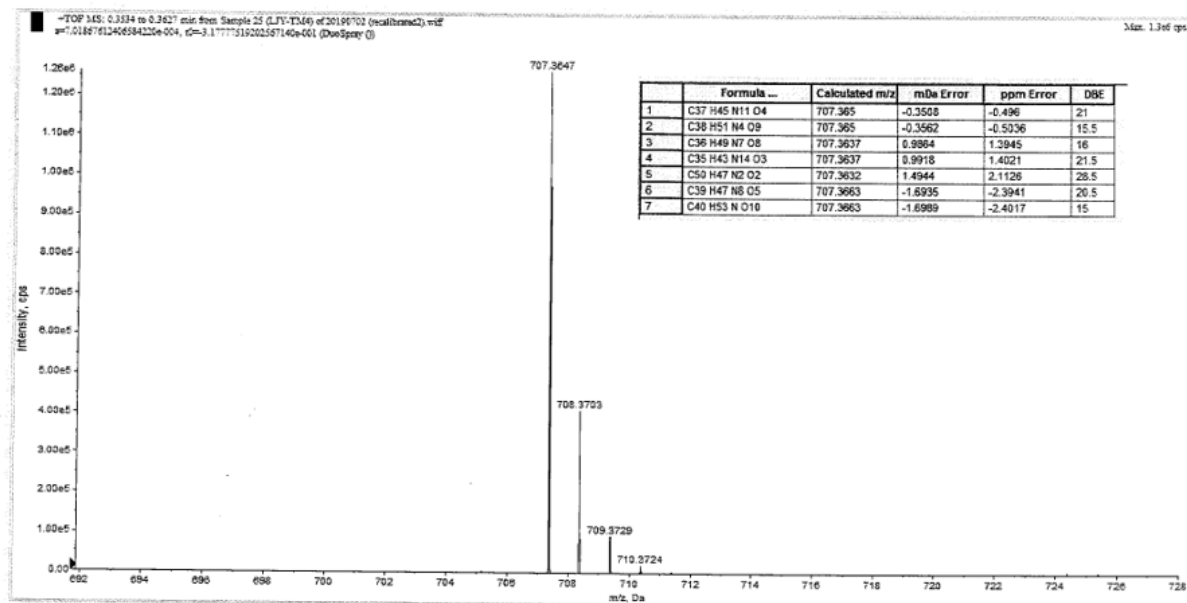


**Figure S9.**  $^{13}\text{C NMR}$  spectrum of compound **1c** ( $\text{CDCl}_3$ , 298 K, 151 MHz)

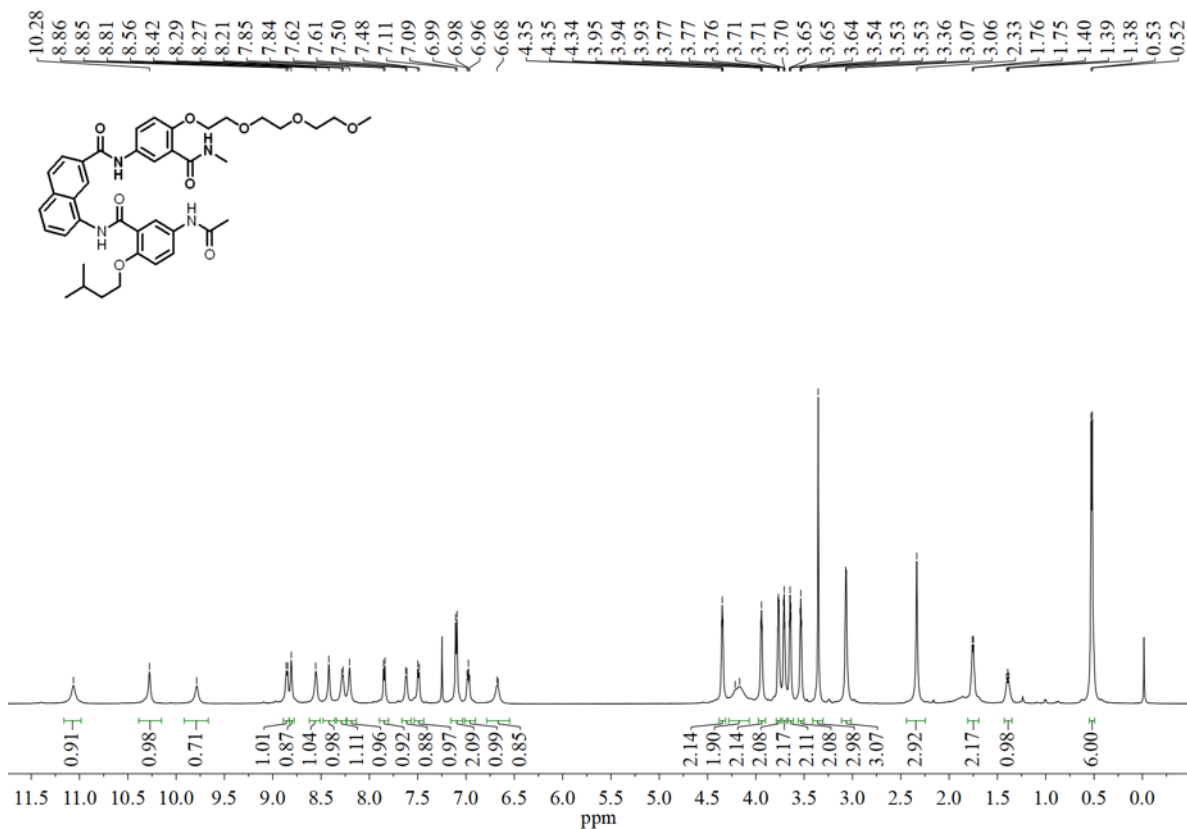




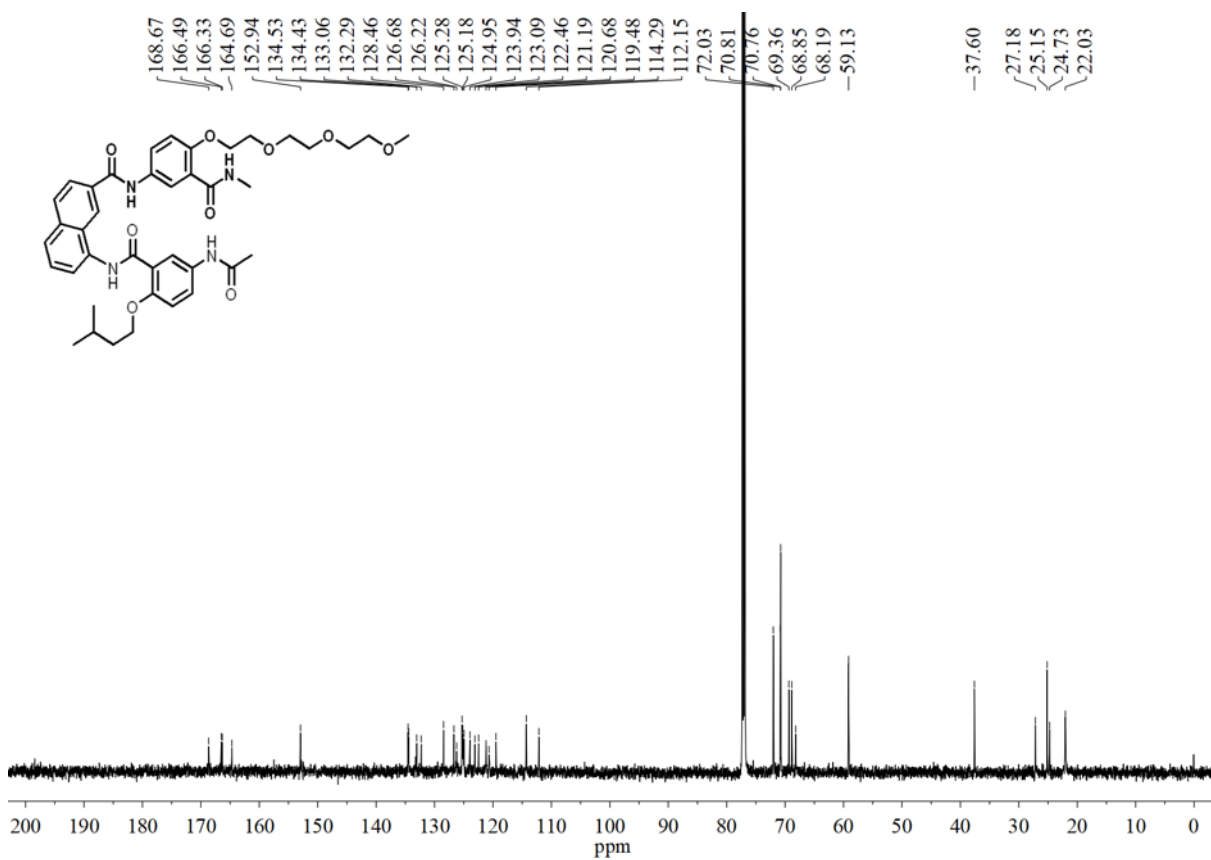
**Figure S12.** <sup>13</sup>C NMR spectrum of compound **1d** (CDCl<sub>3</sub>, 298 K, 151 MHz)



**Figure S13.** MS spectrum of compound **1d**



**Figure S14.** <sup>1</sup>H NMR spectrum of compound 1e (CDCl<sub>3</sub>, 298 K, 600 MHz)



**Figure S15.** <sup>13</sup>C NMR spectrum of compound 1e (CDCl<sub>3</sub>, 298 K, 151 MHz)

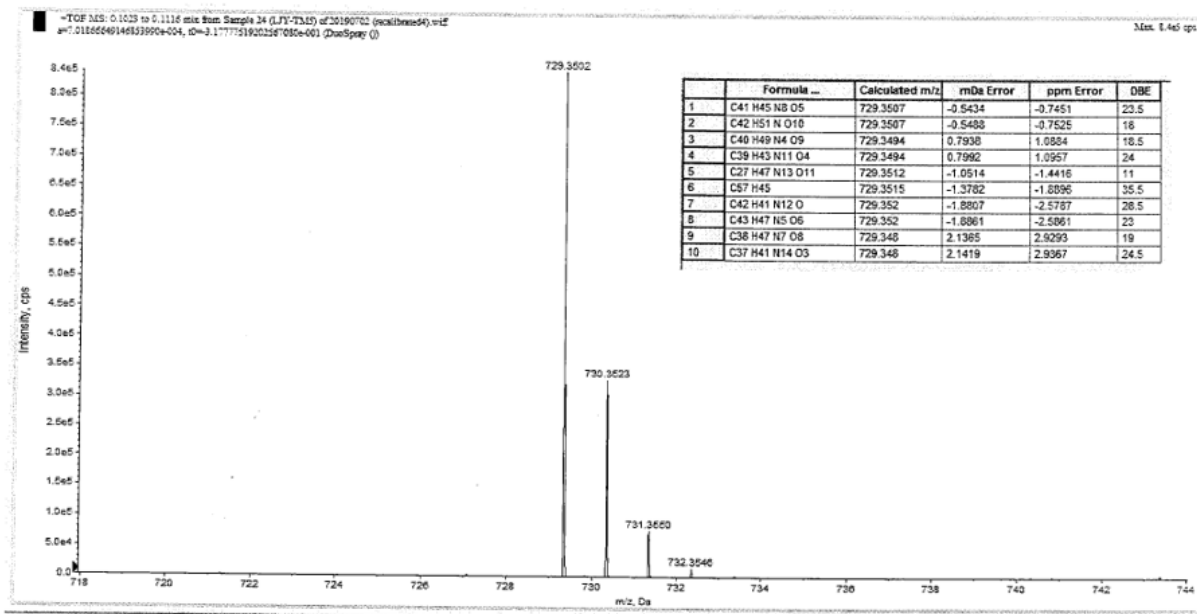


Figure S16. MS spectrum of compound 1e

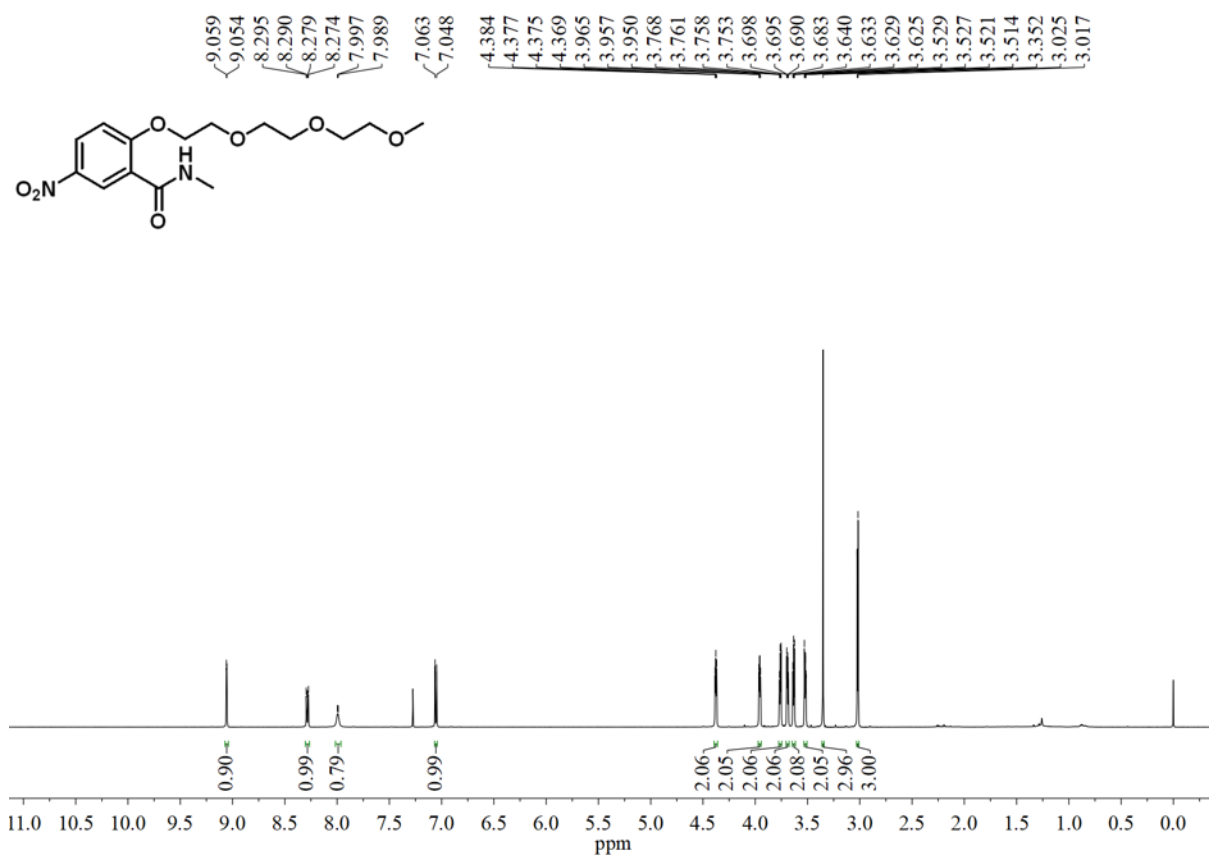
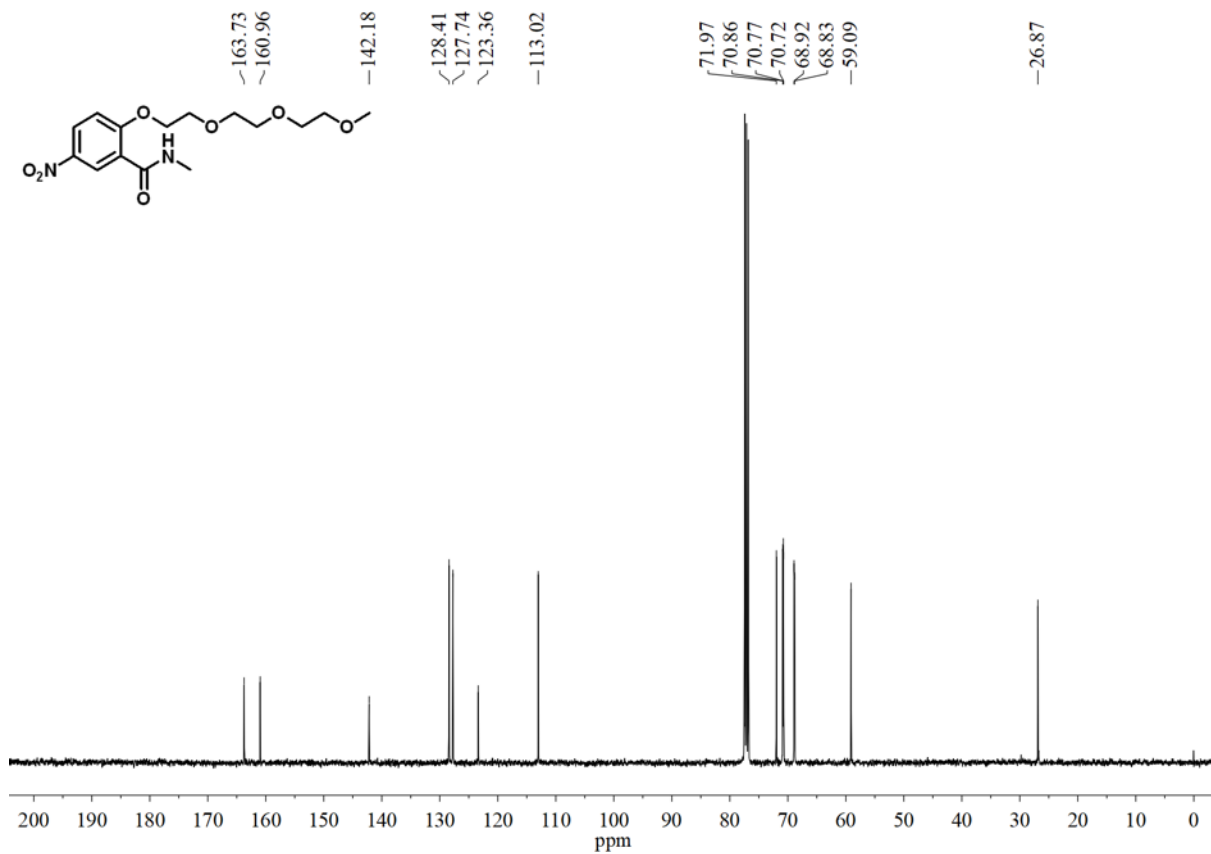
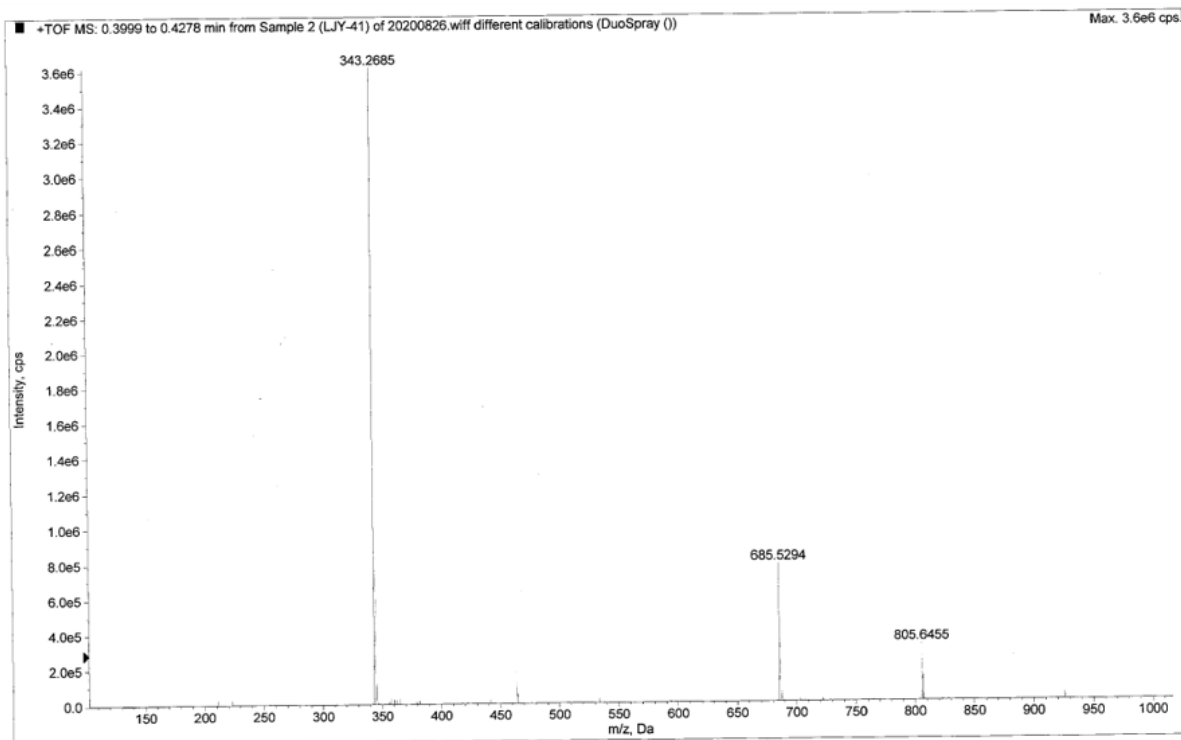


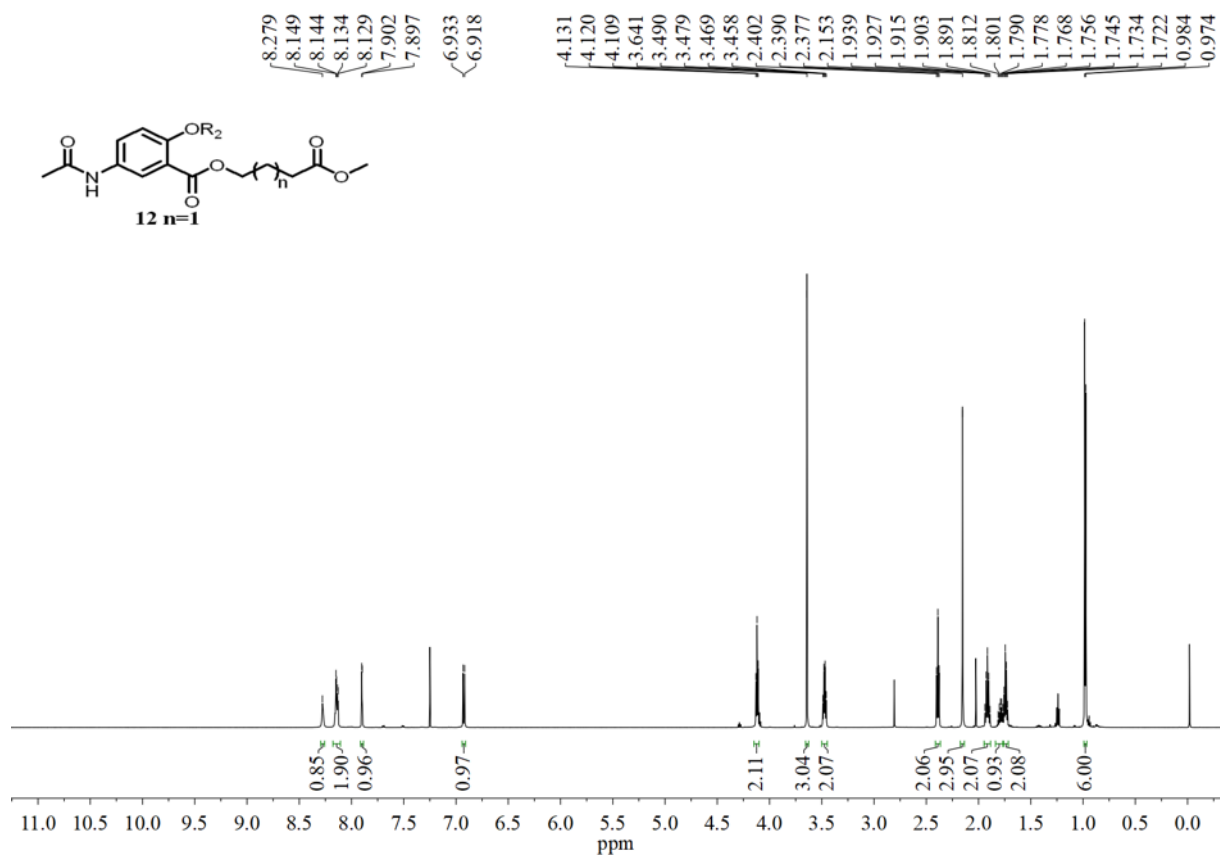
Figure S17. <sup>1</sup>H NMR spectrum of compound 6 (CDCl<sub>3</sub>, 298 K, 600 MHz)



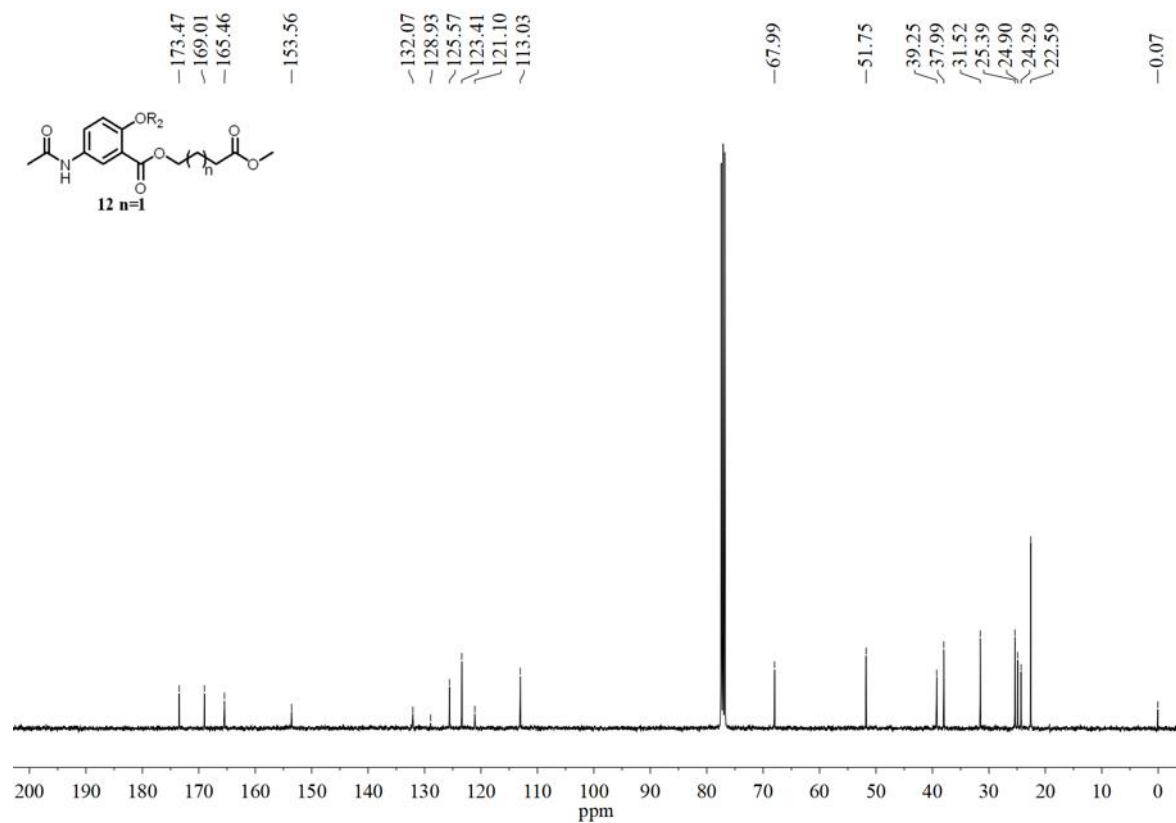
**Figure S18.**  $^{13}\text{C}$  NMR spectrum of compound **6** ( $\text{CDCl}_3$ , 298 K, 101 MHz)



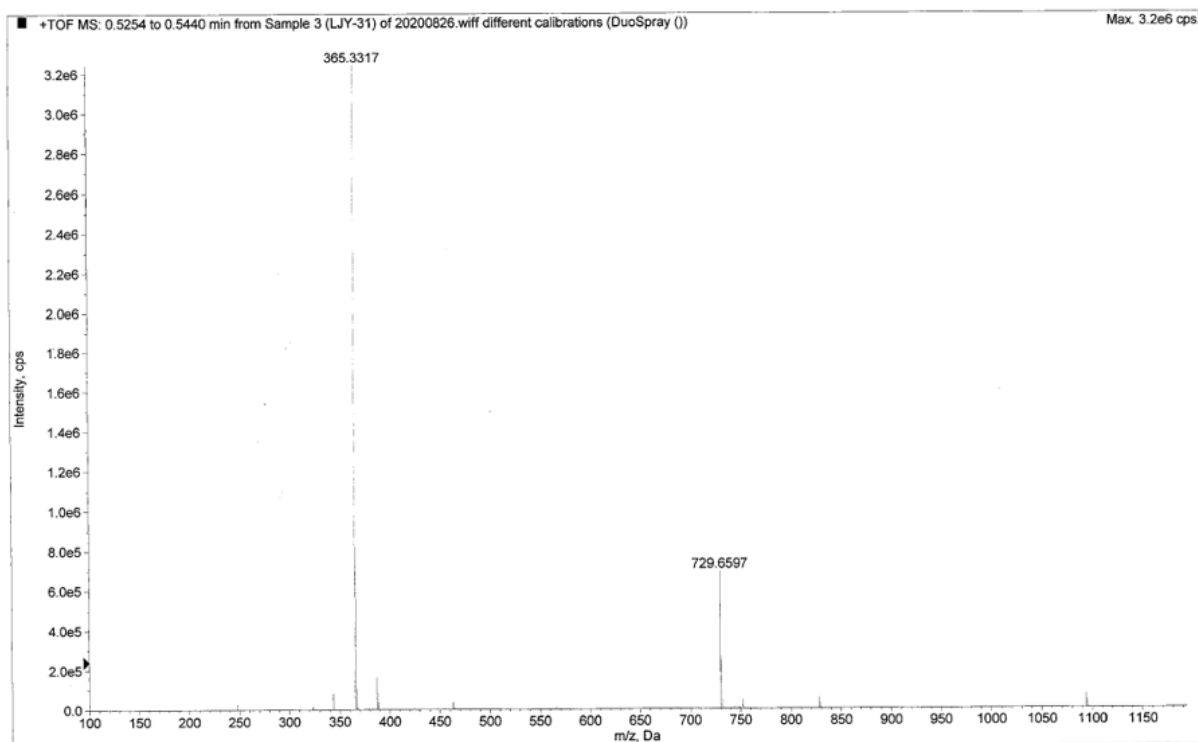
**Figure S19.** MS spectrum of compound **6**



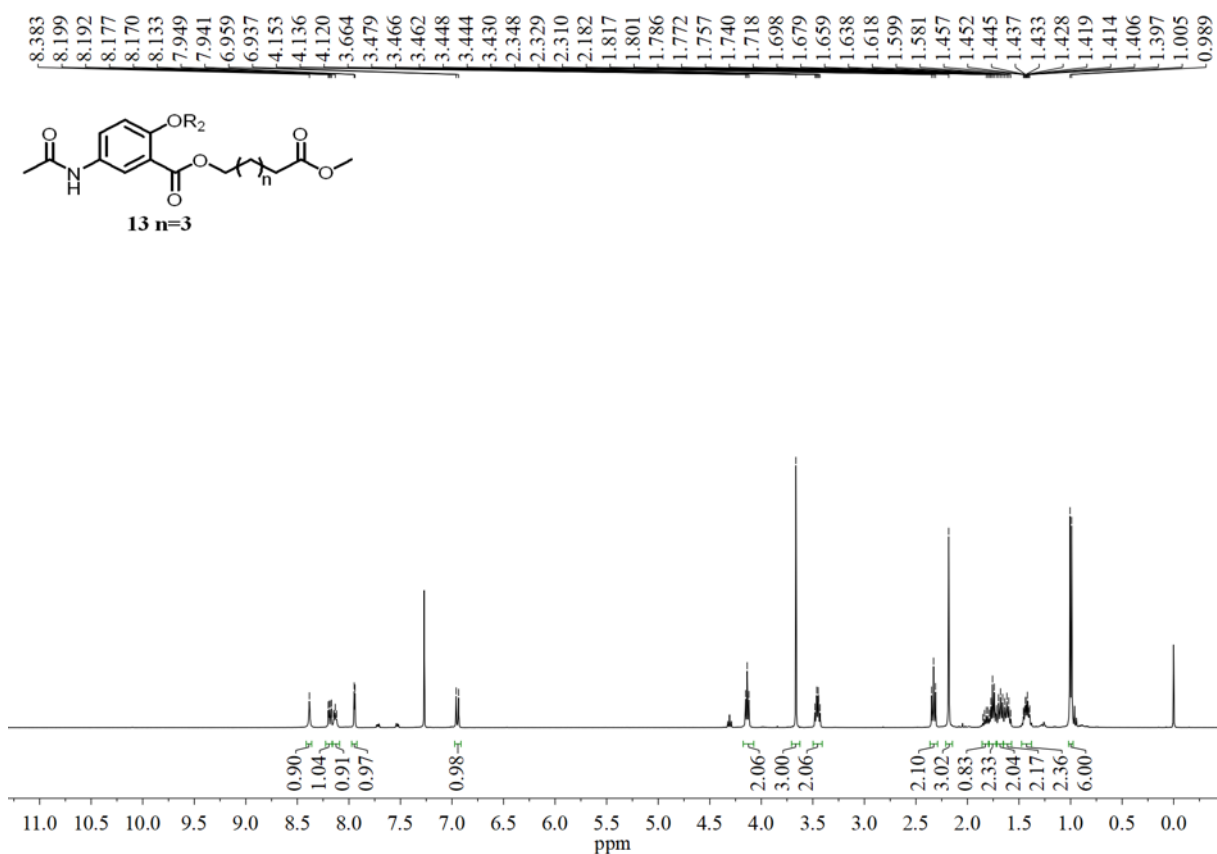
**Figure S20.**  $^1\text{H NMR}$  spectrum of compound **12** ( $\text{CDCl}_3$ , 298 K, 600 MHz)



**Figure S21.**  $^{13}\text{C NMR}$  spectrum of compound **12** ( $\text{CDCl}_3$ , 298 K, 101 MHz)

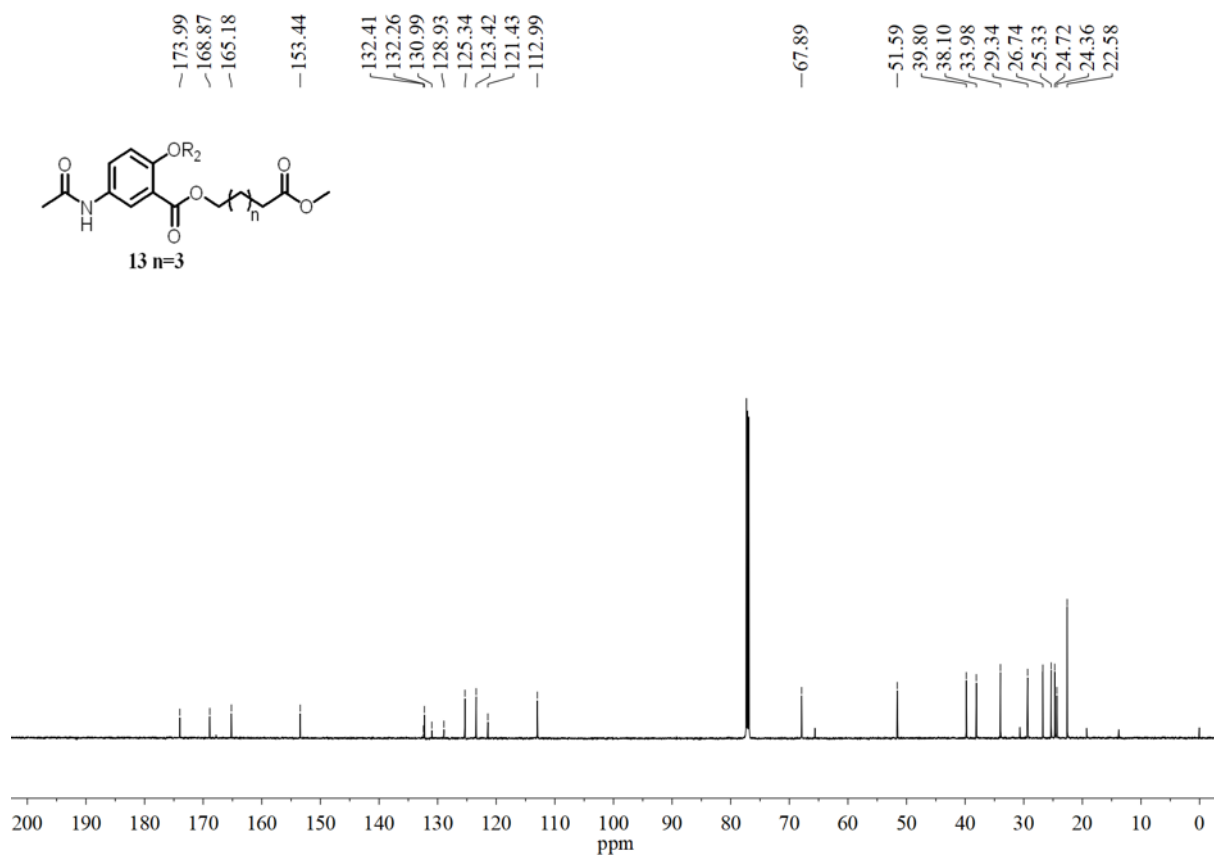


**Figure S22.** MS spectrum of compound **12**

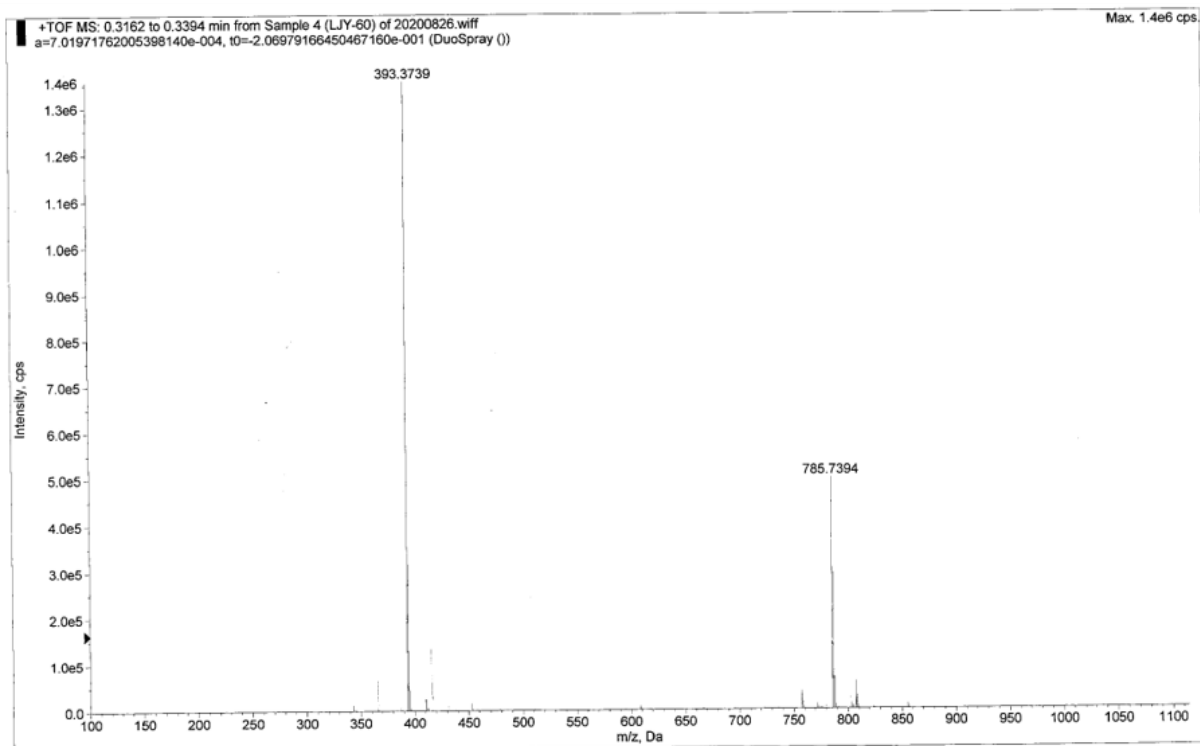


**Figure S23.**  $^1\text{H}$  NMR spectrum of compound **13** ( $\text{CDCl}_3$ , 298 K, 400 MHz)





**Figure S24.**  $^{13}\text{C}$  NMR spectrum of compound **13** (CDCl<sub>3</sub>, 298 K, 151 MHz)



**Figure S25.** MS spectrum of compound **13**

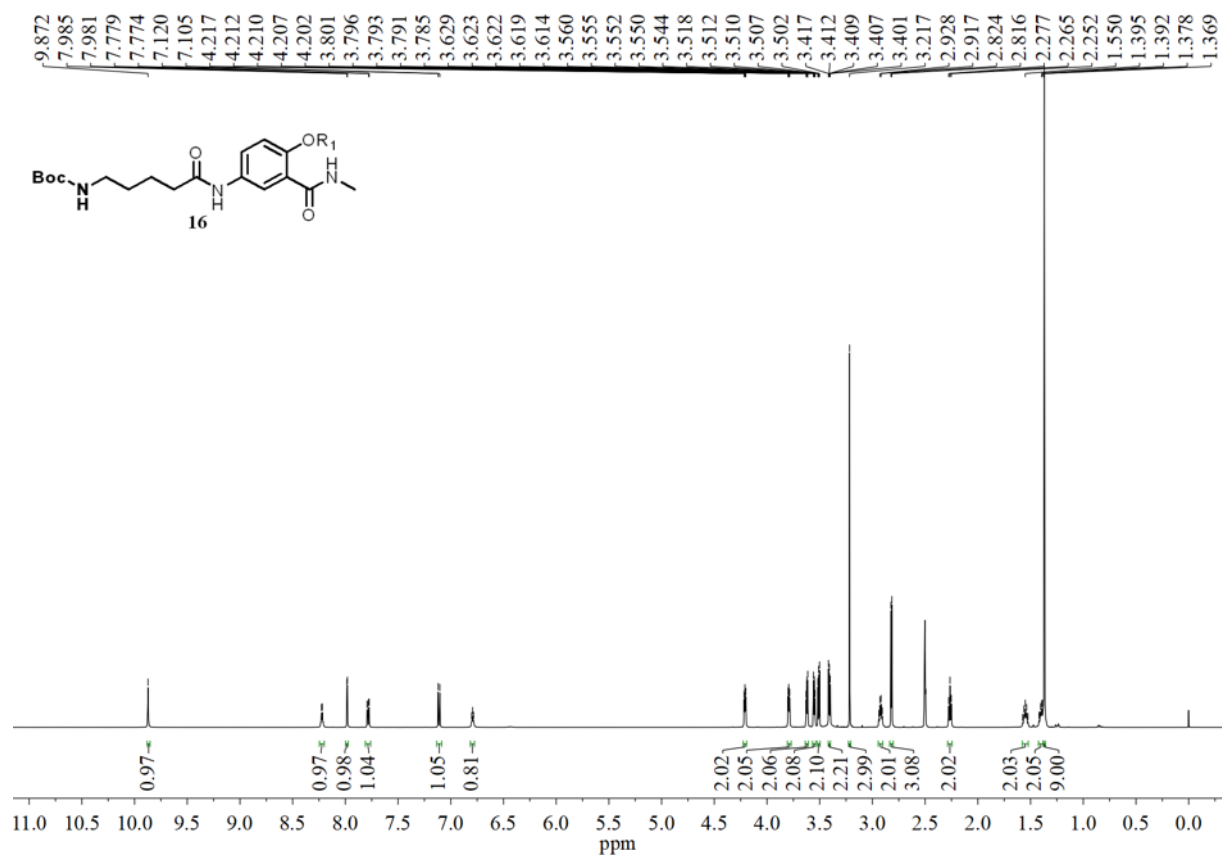


Figure S26.  $^1\text{H}$  NMR spectrum of compound 16 (DMSO, 298 K, 600 MHz)

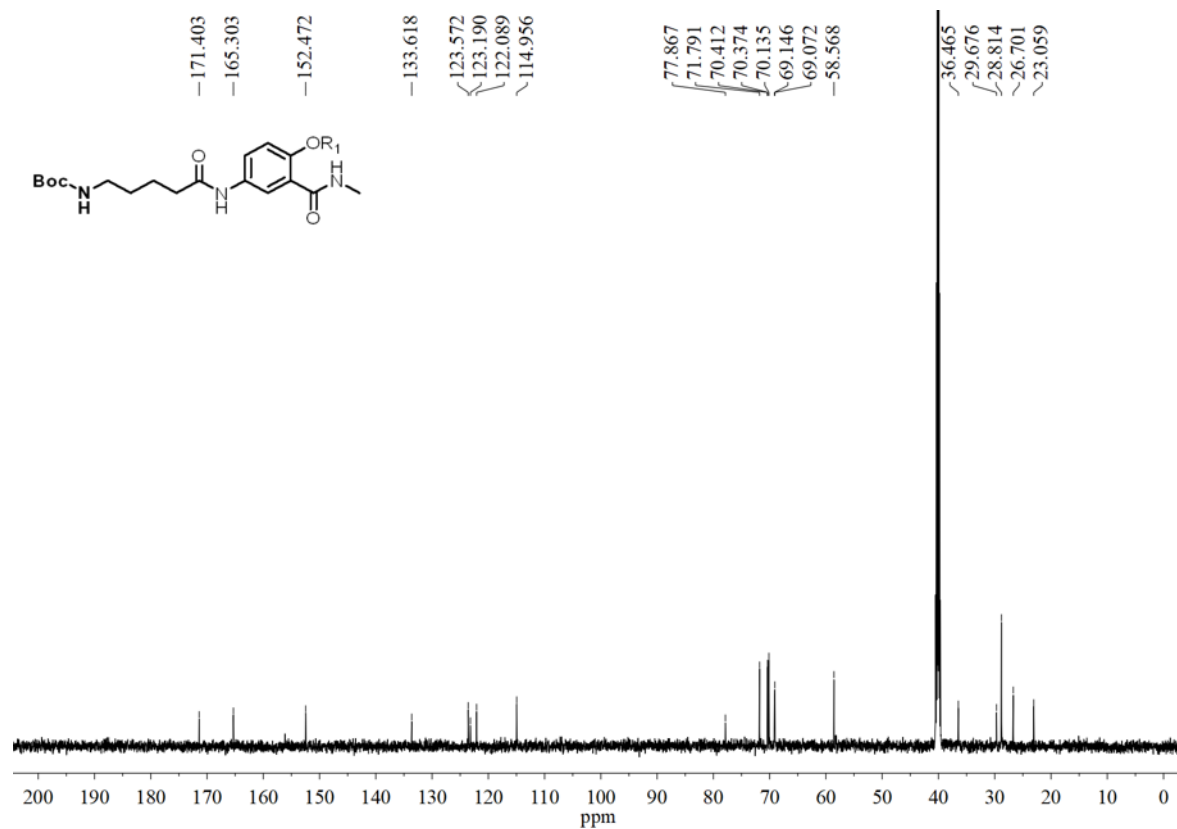
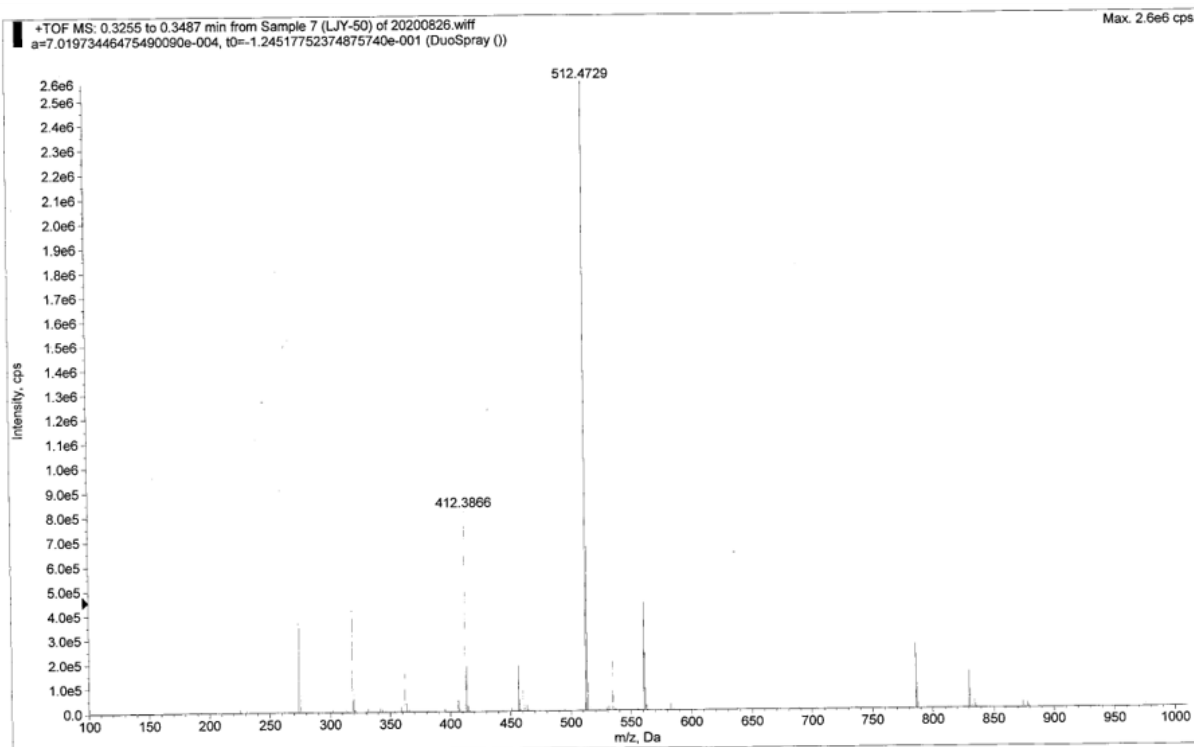
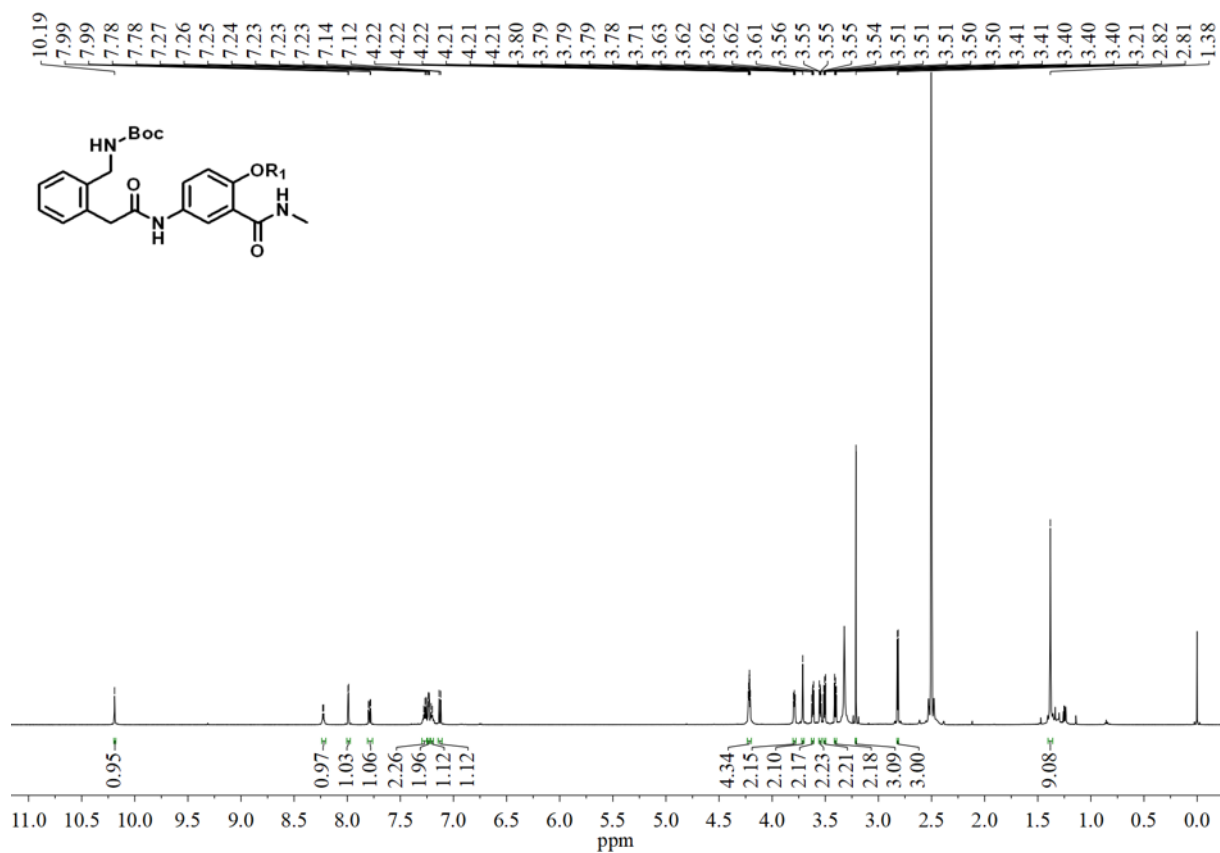


Figure S27.  $^{13}\text{C}$  NMR spectrum of compound 16 (DMSO, 298 K, 151 MHz)



**Figure S28.** MS spectrum of compound **16**



**Figure S29.** <sup>1</sup>H NMR spectrum of compound **18** (DMSO, 298 K, 600 MHz)

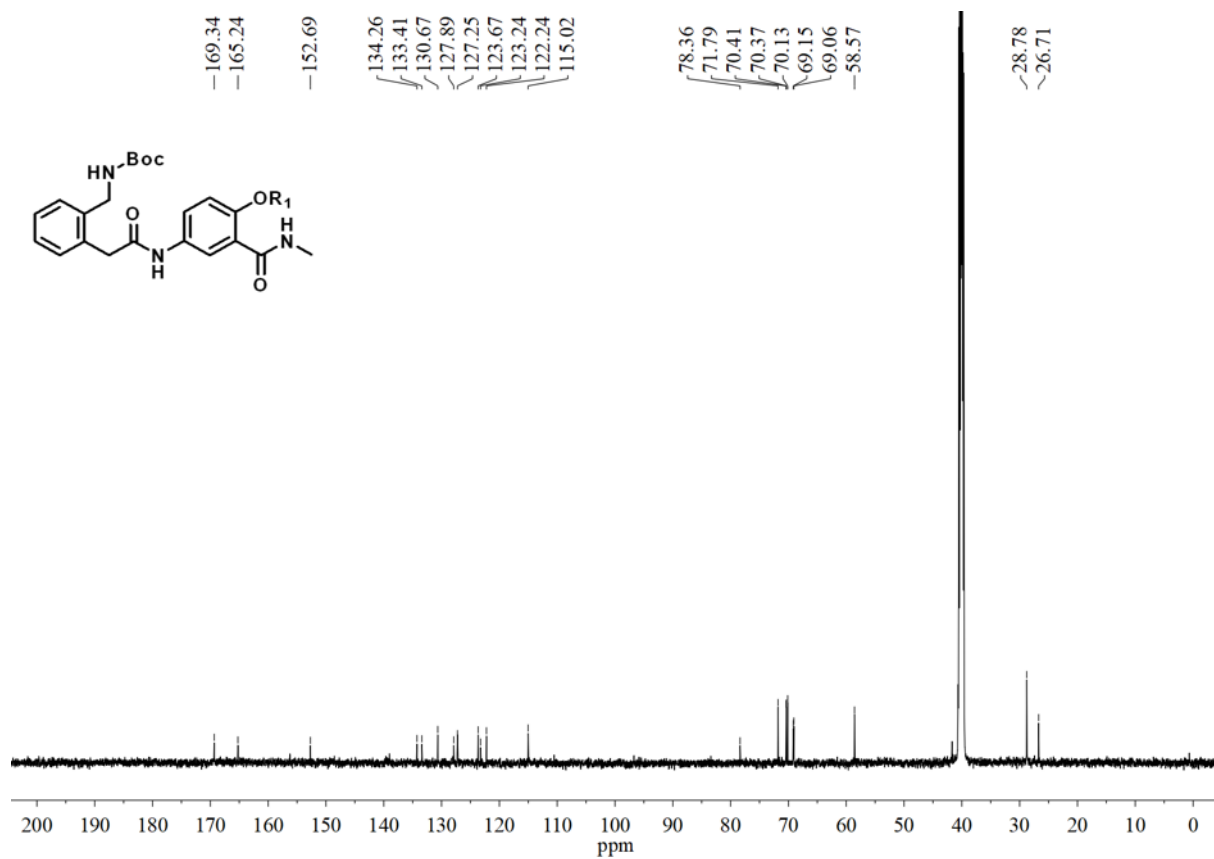


Figure S30. <sup>13</sup>C NMR spectrum of compound 18 (DMSO, 298 K, 151 MHz)

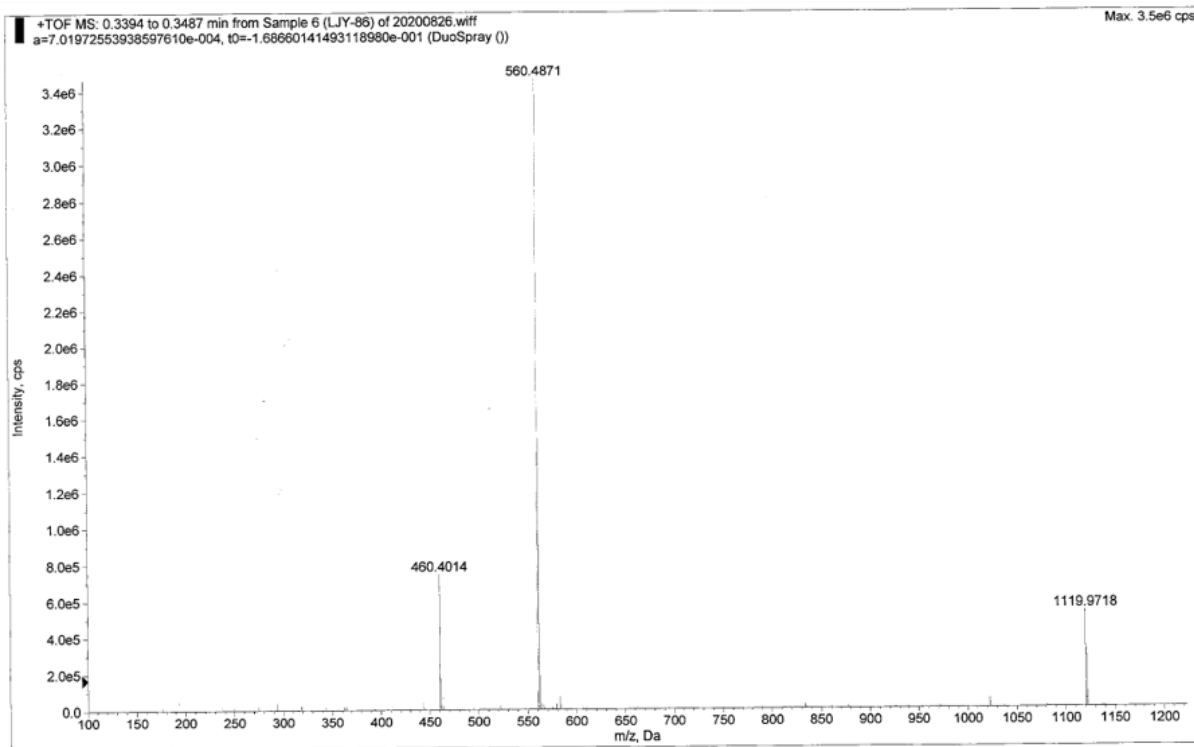
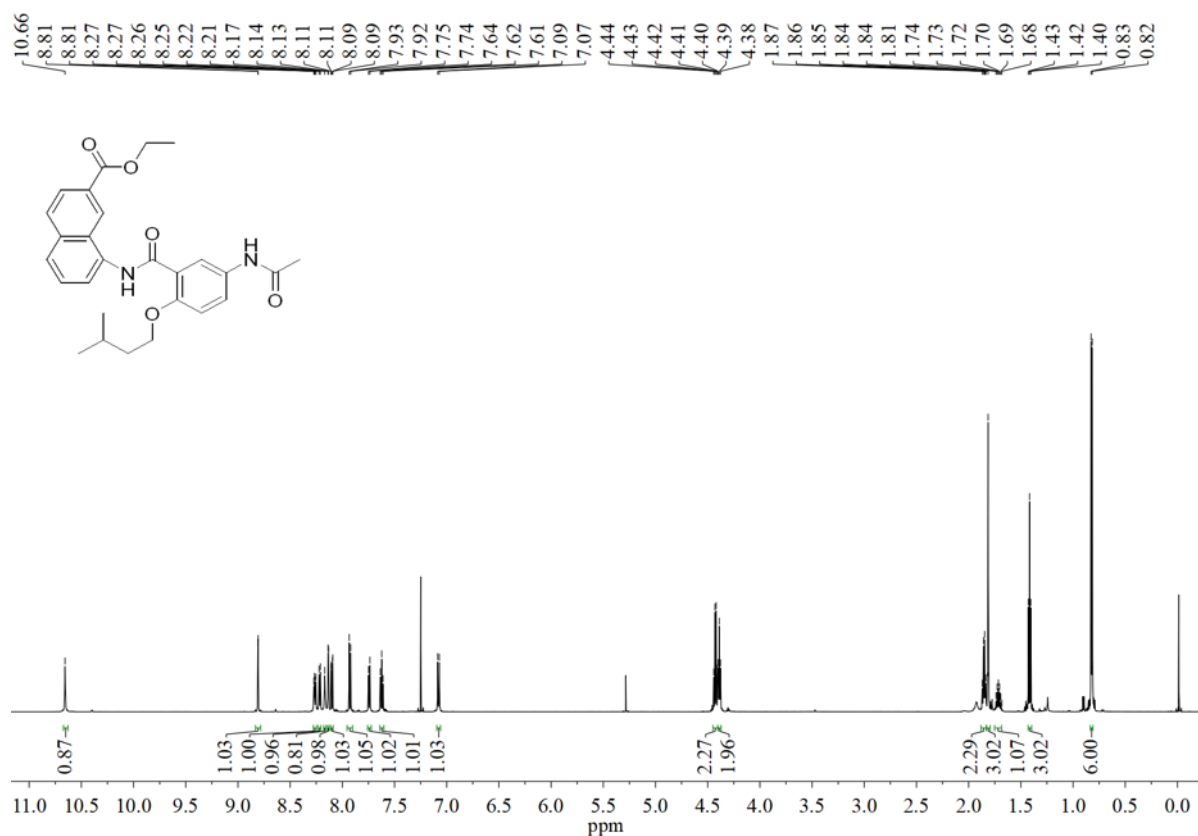
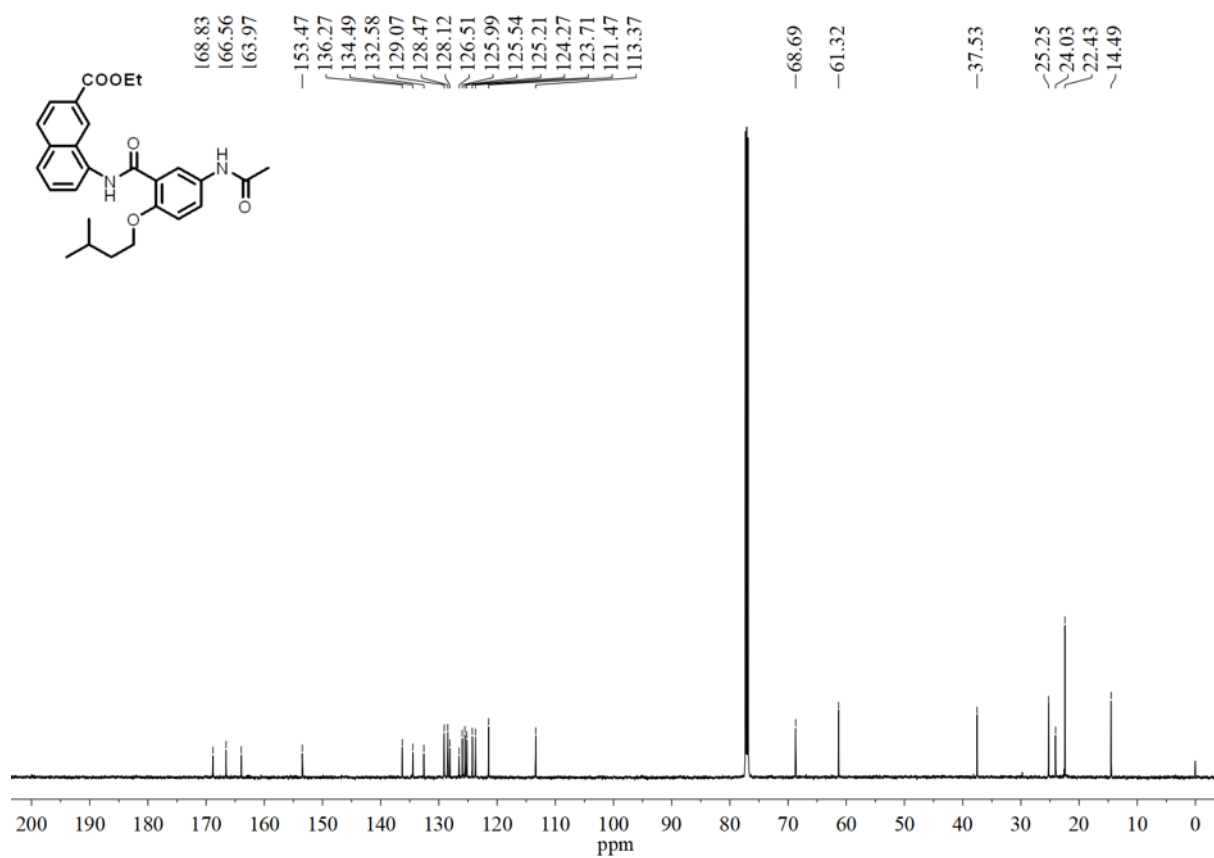


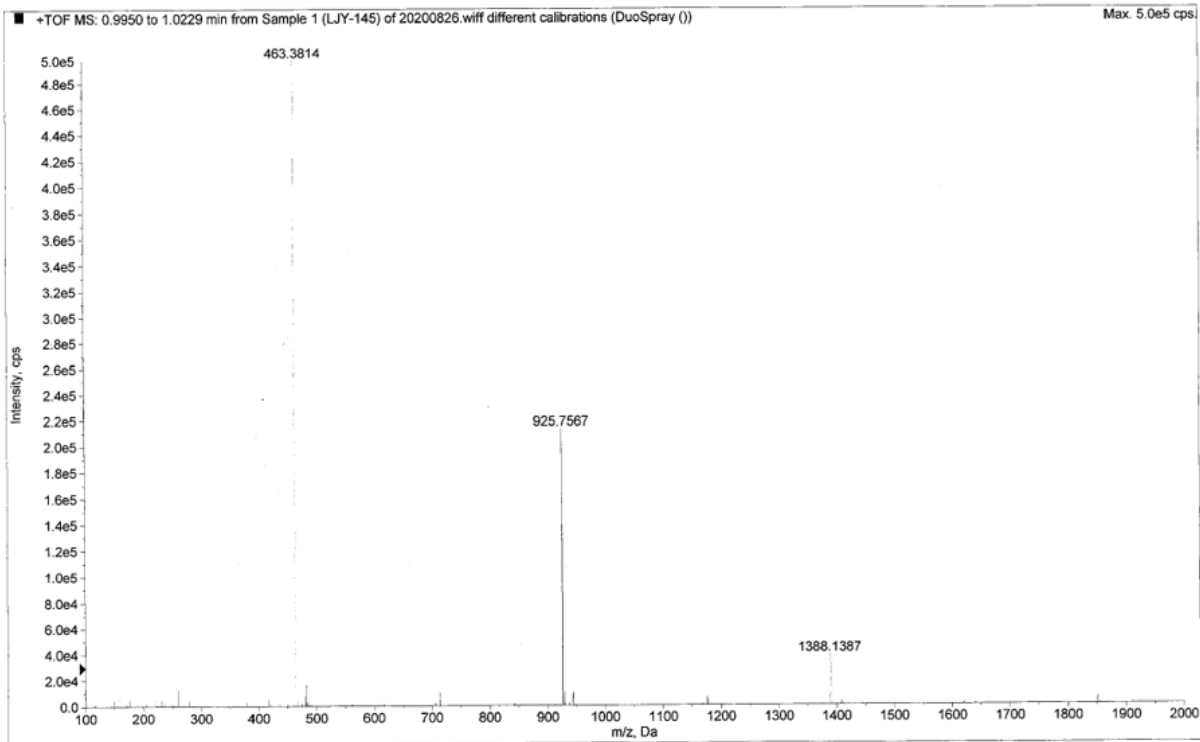
Figure S31. MS spectrum of compound 18



**Figure S32.** <sup>1</sup>H NMR spectrum of compound **21** (CDCl<sub>3</sub>, 298 K, 600 MHz)



**Figure S33.** <sup>13</sup>C NMR spectrum of compound **21** (CDCl<sub>3</sub>, 298 K, 151 MHz)



*Figure S34.* MS spectrum of compound **21**

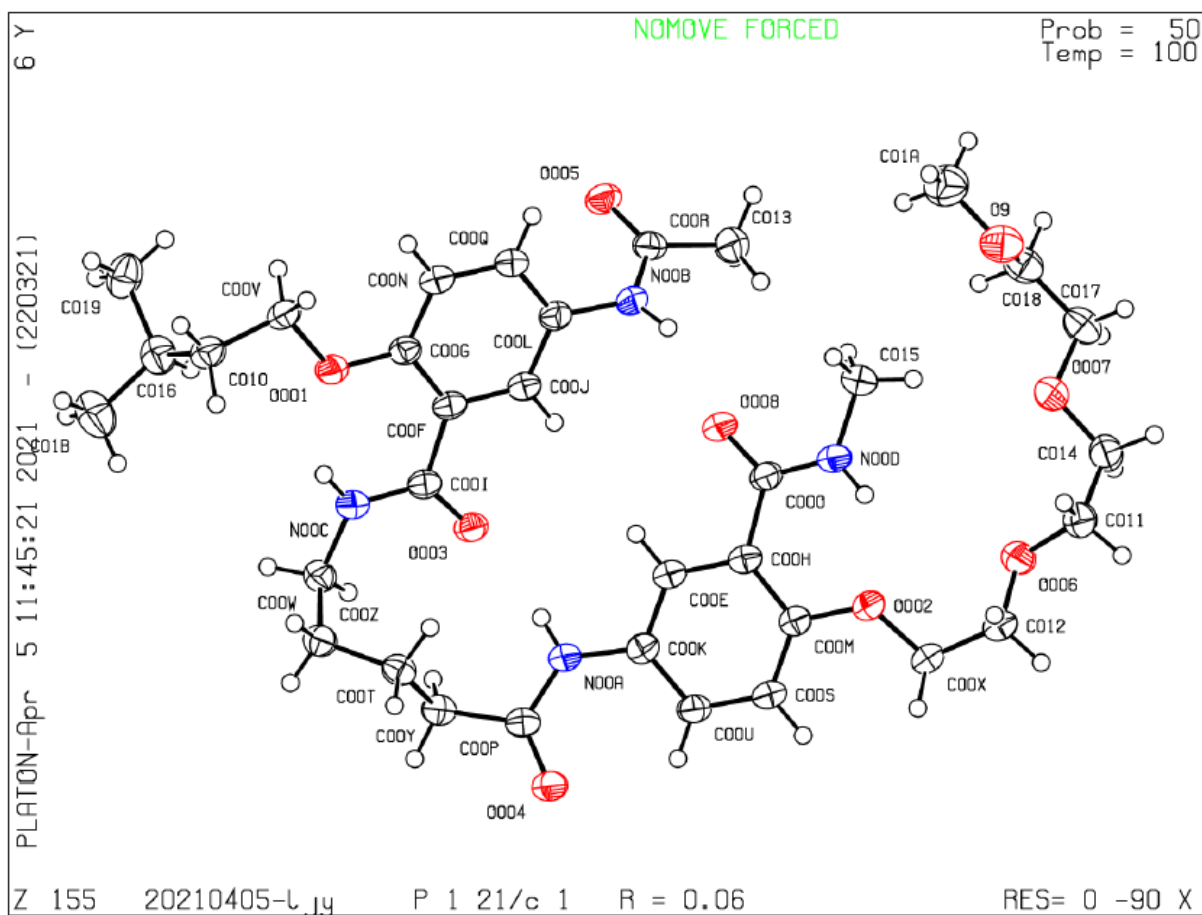
### III. X-ray crystallography

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|   |   |                    |
|---|---|--------------------|
| Bond precision:   | C-C = 0.0025 Å                                  | Wavelength=1.54184 |
| Cell:   | a=17.8519(6)      b=7.8840(2)      c=24.4993(7) |                    |
|   | alpha=90      beta=92.698(3)      gamma=90      |                    |
| Temperature: 100 K  |   |                    |
|   | Calculated                                      | Reported           |
| Volume  | 3444.32(18)                                     | 3444.32(18)        |
| Space group   | P 21/c  | P 1 21/c 1         |
| Hall group  | -P 2ybc   | -P 2ybc            |
| Moiety formula  | C34 H50 N4 O9                                   | C34 H50 N4 O9      |
| Sum formula   | C34 H50 N4 O9                                   | C34 H46 N4 O9      |
| Mr  | 658.78  | 654.75             |
| Dx, g cm <sup>-3</sup>  | 1.270   | 1.263              |
| Z   | 4   | 4                  |
| Mu (mm <sup>-1</sup> )  | 0.757   | 0.757              |
| F000  | 1416.0  | 1400.0             |
| F000'   | 1420.51   |                    |
| h,k,lmax  | 22,9,30   | 22,9,30            |
| Nref  | 7211  | 6875               |
| Tmin,Tmax   | 0.834,0.927                                     | 0.452,1.000        |
| Tmin'   | 0.828   |                    |
| Correction method= # Reported T Limits: Tmin=0.452 Tmax=1.000 AbsCorr = |   |                    |
| MULTI-SCAN  |   |                    |
| Data completeness= 0.953  | Theta(max)= 76.232                              |                    |
| R(reflections)= 0.0616( 5765)   | wR2(reflections)= 0.1799( 6875)                 |                    |
| S = 1.360   | Npar= 429                                       |                    |

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**Figure S35.** Crystal data and structure refinement of **1b**



**Figure S36.** Ortep representation as obtained from the CrysAlisPro (Rigaku OD) software of the **1b**



## IV. Computational Study

The molecular modeling and geometric producing were obtained by using Gaussian view 5.0.8 program. Geometries of **1b-1e** models were optimized at the level of B3LYP/6-311++G\*\*, and the energies of the best optimized geometry were calculated at the same level of theory.

**Table S2.** The energy and H-bond distance of hybrid peptides **1b-1e**.

| Compound  | Energy (a.u.) | H <sub>b</sub> ···O (Å) | H <sub>d</sub> ···O (Å) |
|-----------|---------------|-------------------------|-------------------------|
| <b>1b</b> | -1603.5051    | 1.934                   | 2.103                   |
| <b>1c</b> | -1642.8217    | 2.592                   | 2.267                   |
| <b>1d</b> | -1755.9688    | 1.878                   | 2.005                   |
| <b>1e</b> | -1830.9648    | 3.548                   | 4.423                   |

## V. References

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