

# Mixed pentafluorophenyl and *o*-fluorophenyl esters of aliphatic dicarboxylic acids: Efficient tools for peptide conjugation

## Supporting material:

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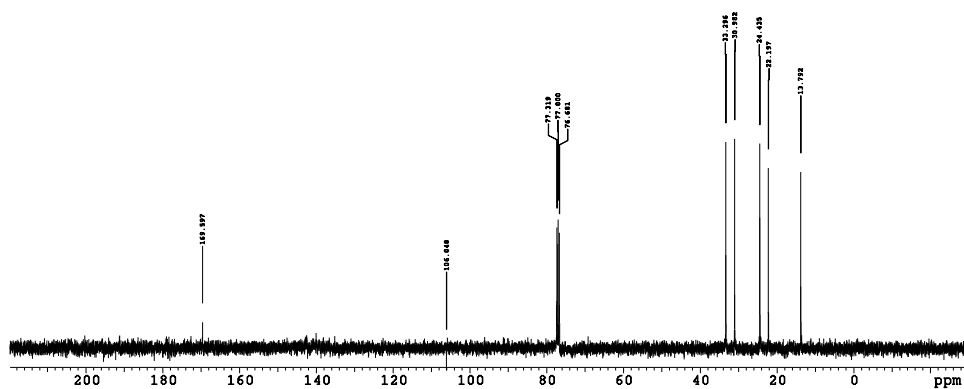
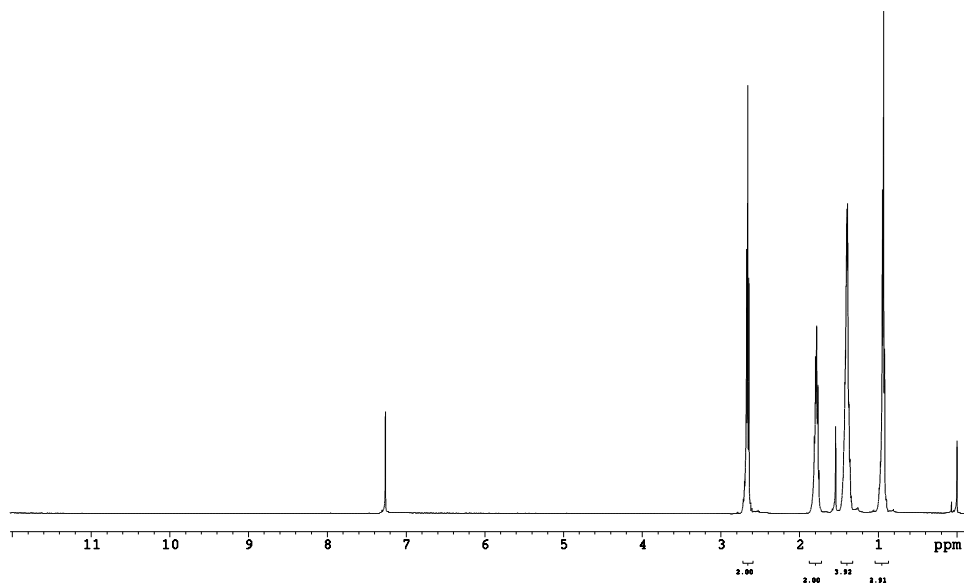
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**Pentafluorophenyl hexanoate (1a) (<sup>1</sup>H NMR, <sup>13</sup>C NMR):**

Yield: 214 mg (95%)

<sup>1</sup>H NMR (500 MHz, *CDCl*<sub>3</sub>) δ ppm 2.66 (m, 2H), 1.78 (m, 2H), 1.40 (m, 4H), 0.93 (m, 3H)

<sup>13</sup>C NMR (101 MHz, *CDCl*<sub>3</sub>) δ ppm 169.6, 33.3, 30.9, 24.4, 22.2, 13.8

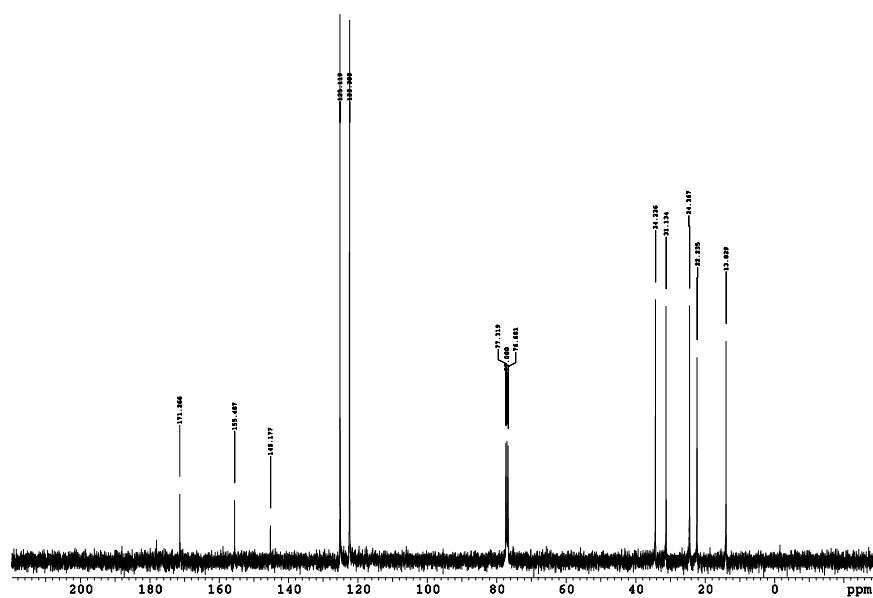
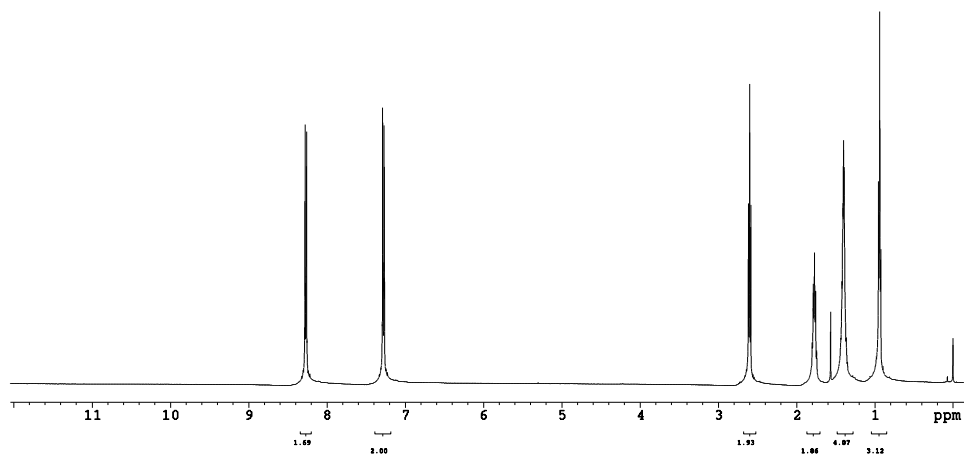


### p-Nitrophenyl hexanoate (**1b**) ( $^1\text{H}$ NMR, $^{13}\text{C}$ NMR)

Yield: 185 mg (98%)

$^1\text{H}$  NMR (500 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm 8.27 (m, 2H), 7.28 (m, 2H), 2.60 (m, 2H), 1.77 (m, 2H), 1.40 (m, 4H), 0.94 (m, 3H)

$^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm 171.3, 155.4, 145.2, 125.1, 122.3, 34.2, 31.1, 24.3, 22.2, 13.8

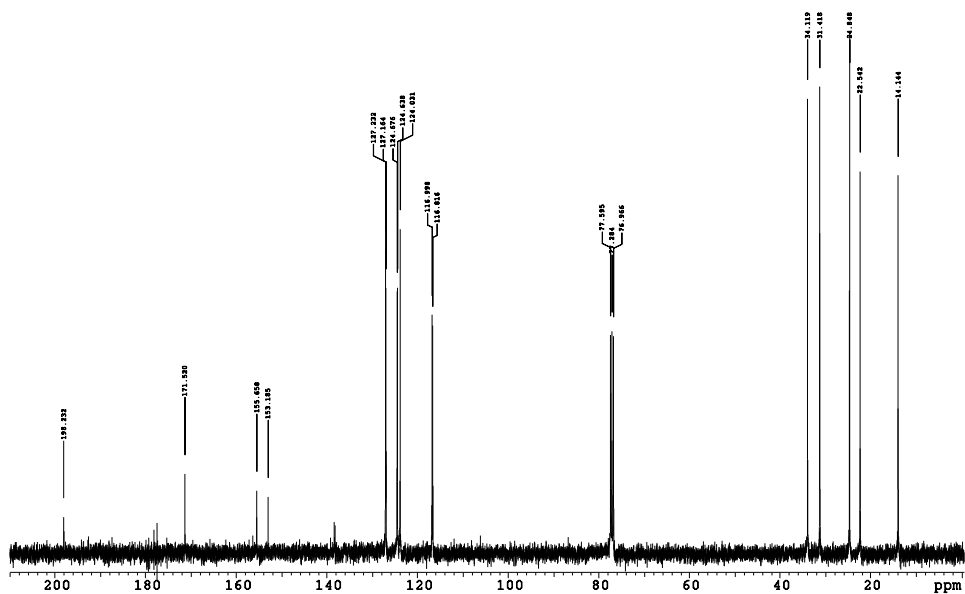
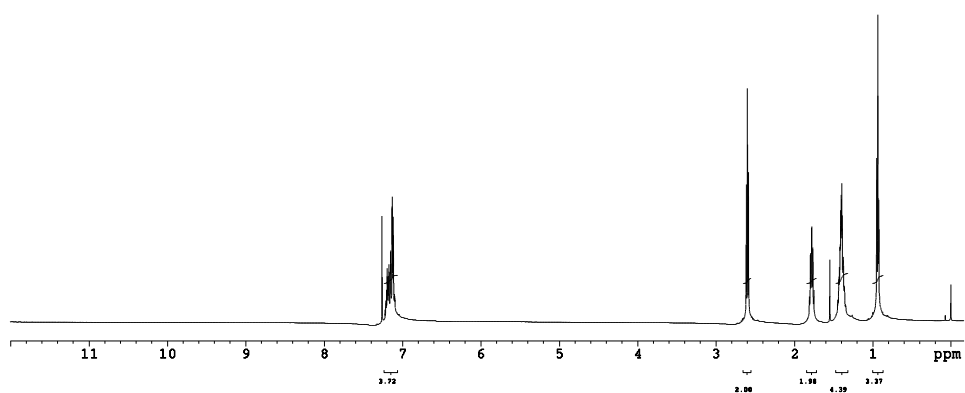


## **o-Fluorophenyl hexanoate (1c). (<sup>1</sup>H NMR, <sup>13</sup>C NMR)**

Yield: 161 mg (96%)

<sup>1</sup>H NMR (500 MHz, CDCl<sub>3</sub>) δ ppm 7.20 (m, 4H), 2.59 (m, 2H), 1.78 (m, 2H), 1.40 (m, 4H), 0.93 (m, 3H)

<sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>) δ ppm 198.8, 171.2, 155.3, 152.8, 126.9, 126.8, 124.3, 124.3, 123.7, 116.7, 116.5, 33.8, 31.1, 24.5, 22.2, 13.8

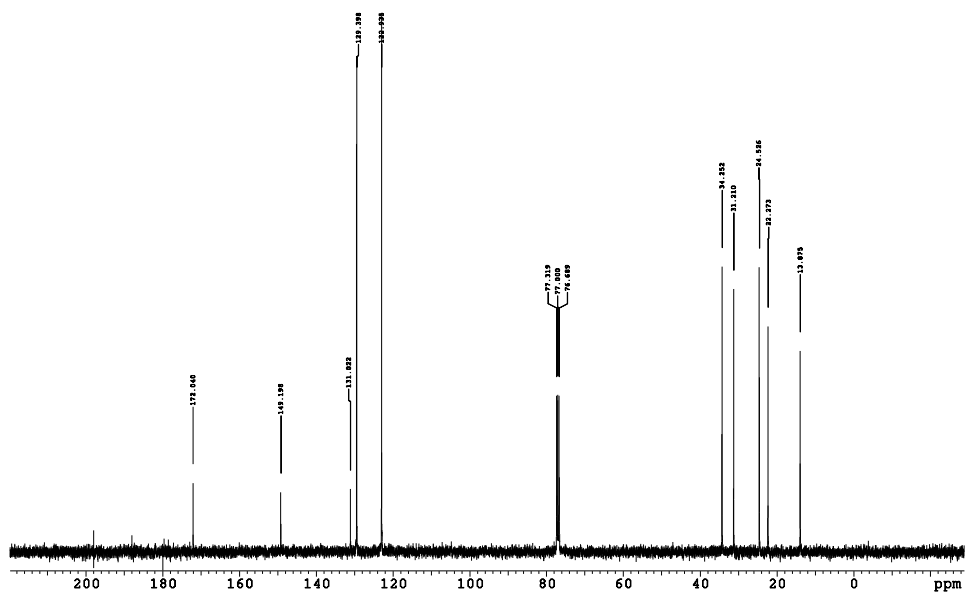
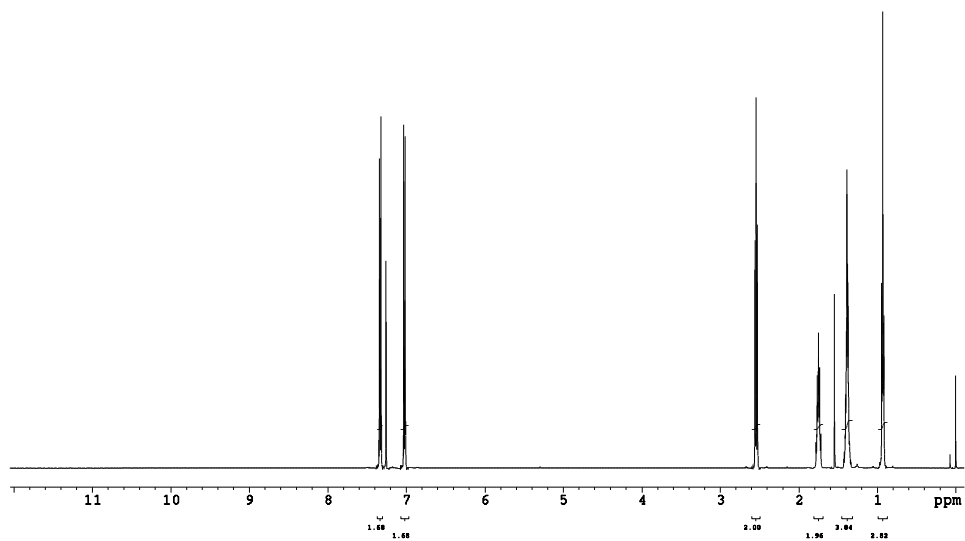


**p-Chlorophenyl hexanoate (1d).** ( $^1\text{H}$  NMR,  $^{13}\text{C}$  NMR,  $^{19}\text{F}$  NMR)

Yield: 170 mg (94%)

$^1\text{H}$  NMR (500 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm 7.33 (m, 2H), 7.02 (m, 2H), 2.54 (m, 2H), 1.75 (m, 2H), 1.38 (m, 4H), 0.93 (m, 3H)

$^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm 197.9, 172.0, 131.0, 129.3, 122.9, 34.2, 31.2, 24.5, 22.2, 13.8

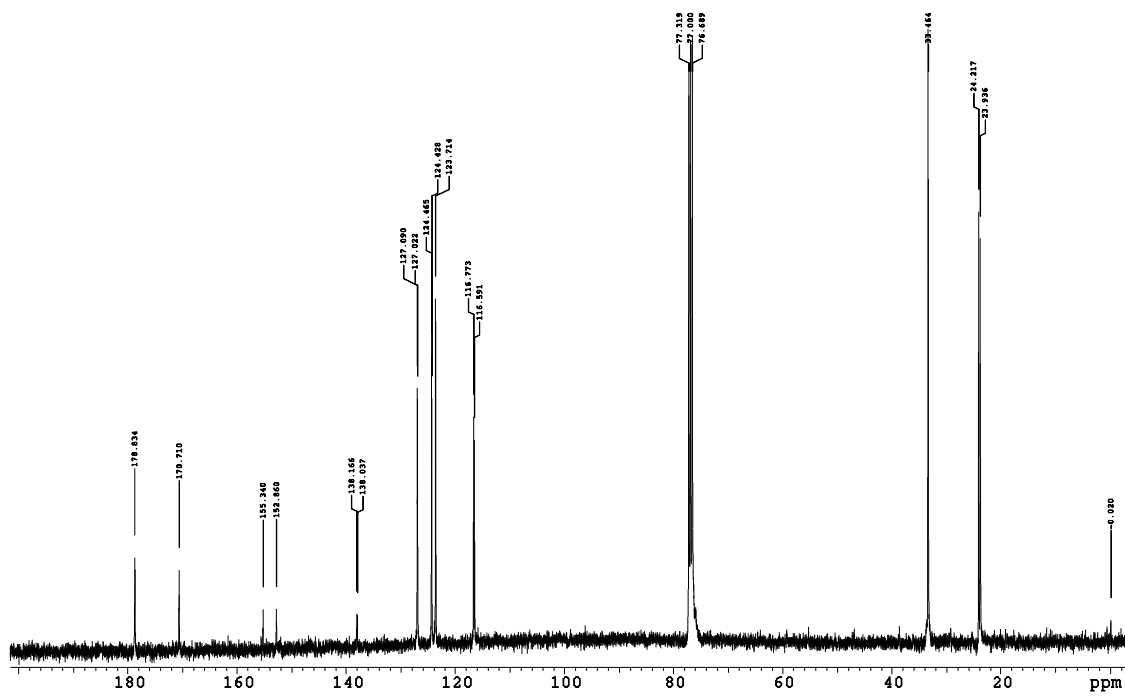
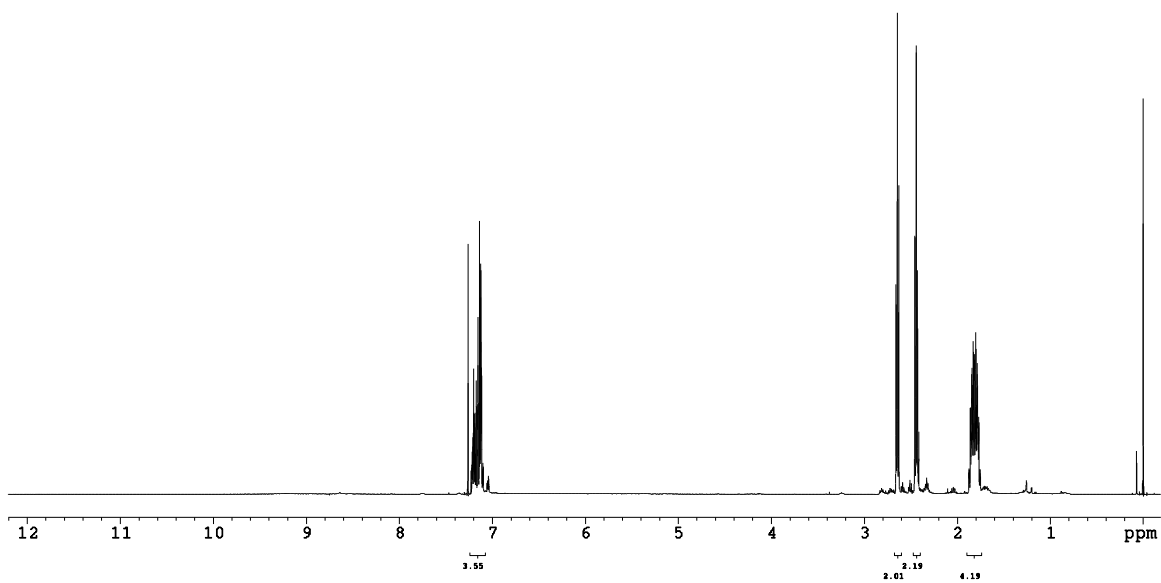


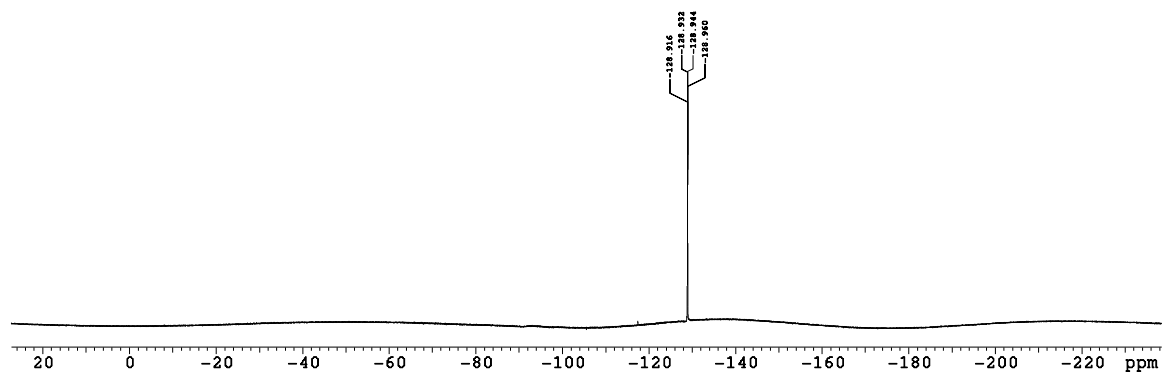
**Mono-*o*-fluorophenyl adipate (2a) (<sup>1</sup>H NMR, <sup>13</sup>C NMR, <sup>19</sup>F NMR):**

<sup>1</sup>H NMR (500 MHz, CDCl<sub>3</sub>) δ ppm 7.16 (m, 4H), 2.64 (t, *J* = 7.1 Hz, 2H), 2.43 (t, *J* = 6.9 Hz, 2H), 1.81 (m, 4H)

<sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>) δ ppm 178.87, 170.70, 155.33, 152.85, 138.16, 127.4, 124.4, 123.71, 116.6, 33.47, 24.22, 23.94

<sup>19</sup>F NMR (376 MHz, CDCl<sub>3</sub>) δ ppm -128.9 (m, 1F)



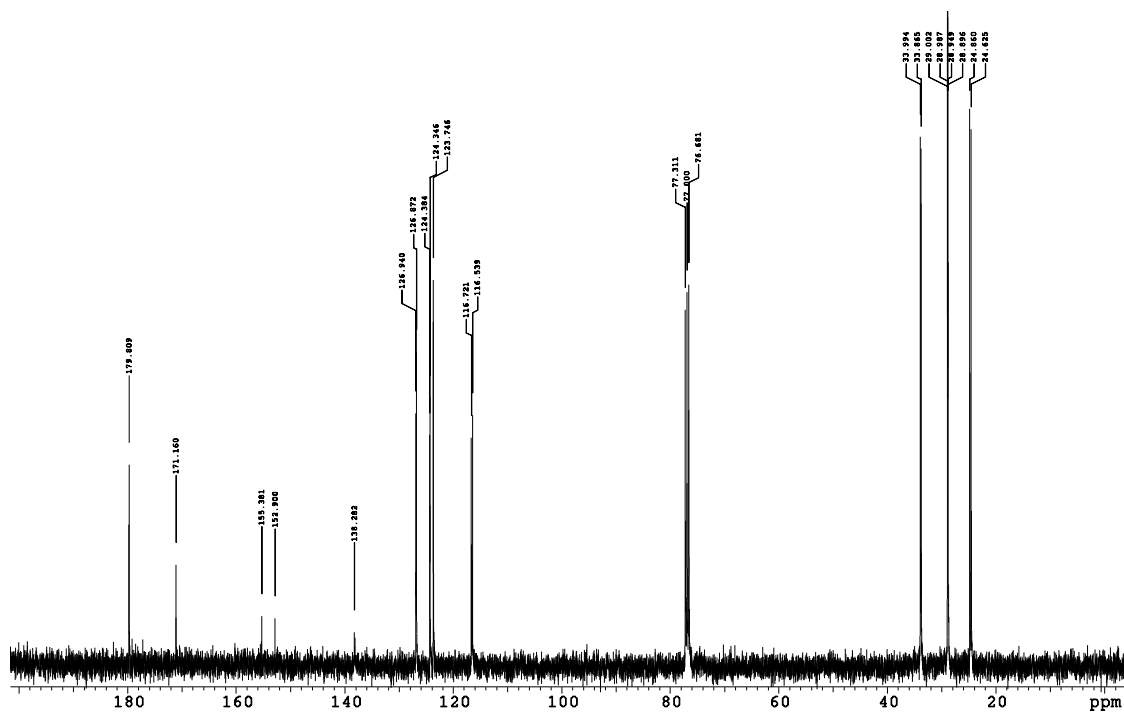
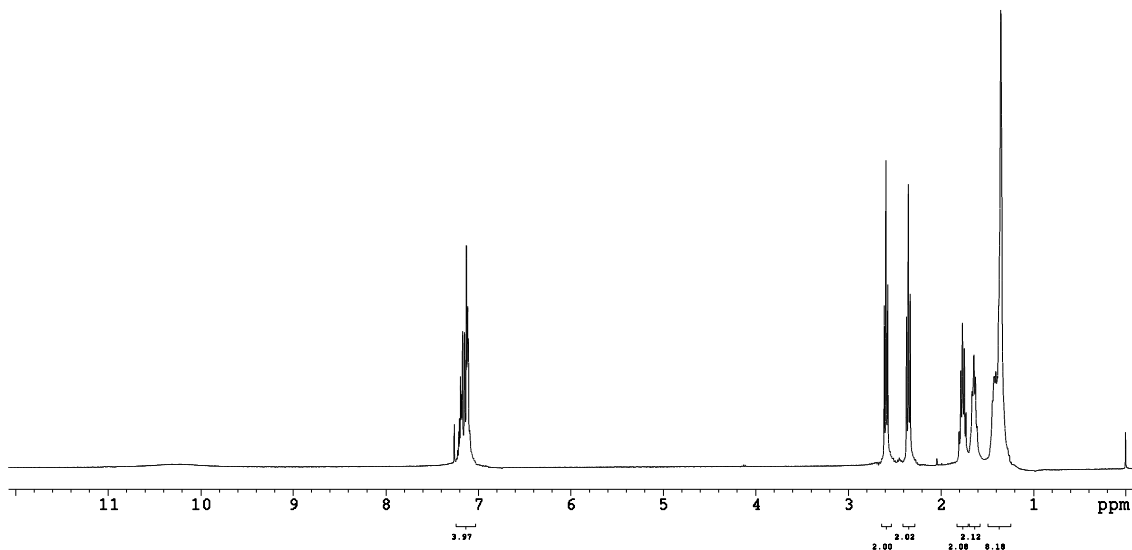


**Mono-*o*-fluorophenyl sebacate (2b) ( $^1\text{H}$  NMR,  $^{13}\text{C}$  NMR,  $^{19}\text{F}$  NMR):**

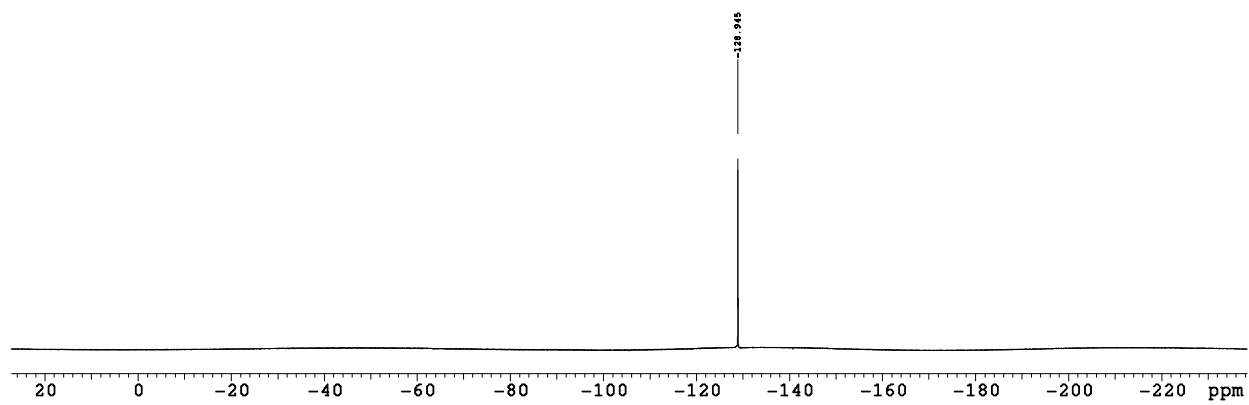
$^1\text{H}$  NMR (500 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm 7.14 (m, 4H), 2.59 (t,  $J = 7.43$  Hz, 2H), 2.35 (t,  $J = 7.5$  Hz, 2H), 1.76 (m, 2H), 1.64 (m, 2H), 1.47-1.31 (m, 8H)

$^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm 179.7, 171.1, 155.3, 152.8, 138.2, 126.9, 126.8, 124.3, 124.3, 123.7, 116.7, 116.5, 33.9, 33.8, 29.0, 28.9, 28.9, 28.9, 24.8, 24.6

$^{19}\text{F}$  NMR (376 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm -128.9 (m, 1F)





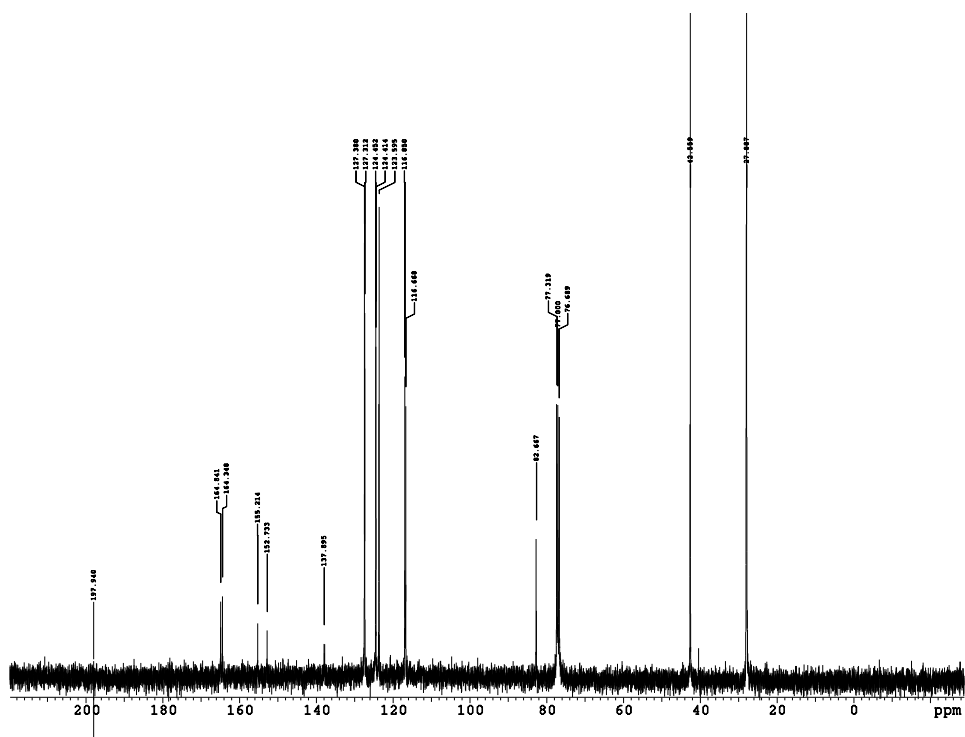
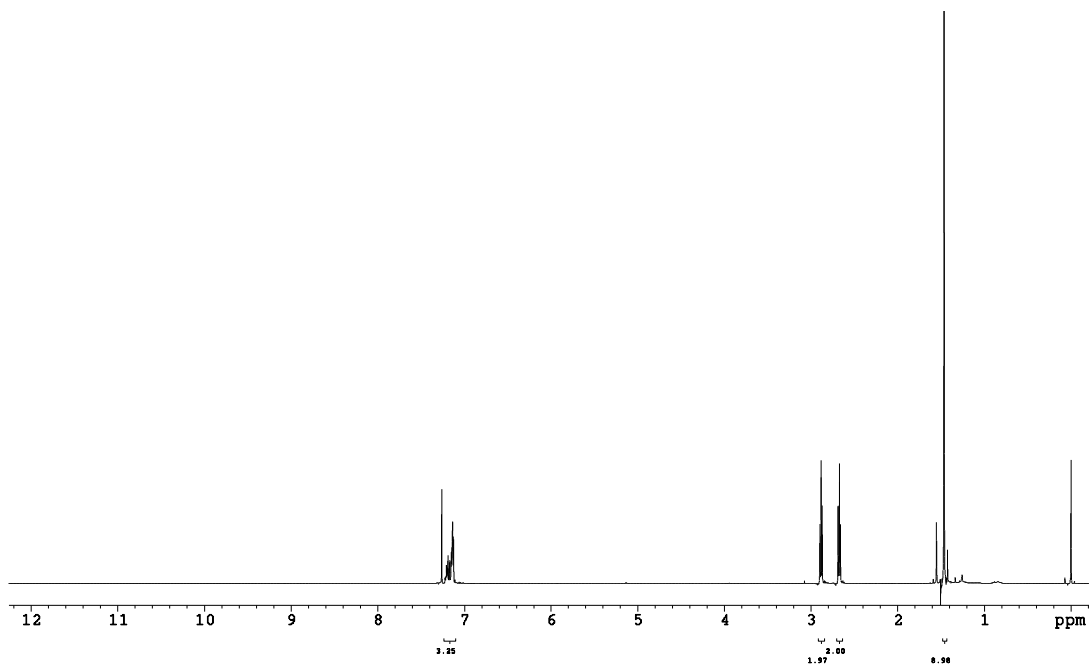


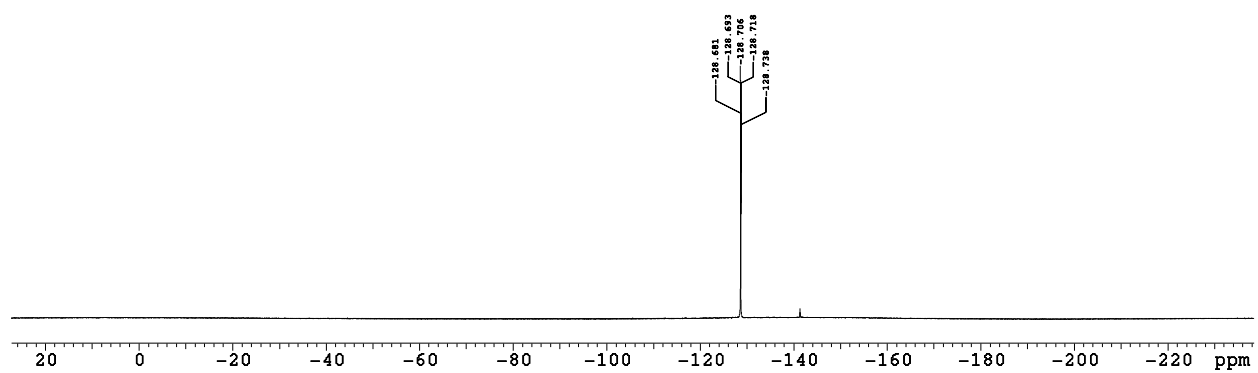
**tert-Butyl o-fluorophenyl succinate (5) ( $^1\text{H}$  NMR,  $^{13}\text{C}$  NMR,  $^{19}\text{F}$  NMR):**

$^1\text{H}$  NMR (500 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm 7.17 (m, 4H), 2.88 (t,  $J = 6.9$  Hz, 2H), 2.67 (t,  $J = 6.9$  Hz, 2H), 1.46 (s, 9H)

$^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm 197.9, 164.5 (d,  $J = 49.7$  Hz), 153.9 (d,  $J = 249.8$  Hz), 127.3 (d,  $J = 7.0$  Hz), 124.4 (d,  $J = 3.9$  Hz), 123.5, 116.7 (d,  $J = 18.4$  Hz), 82.6, 42.5, 27.8

$^{19}\text{F}$  NMR (376 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm -128.7 (m).



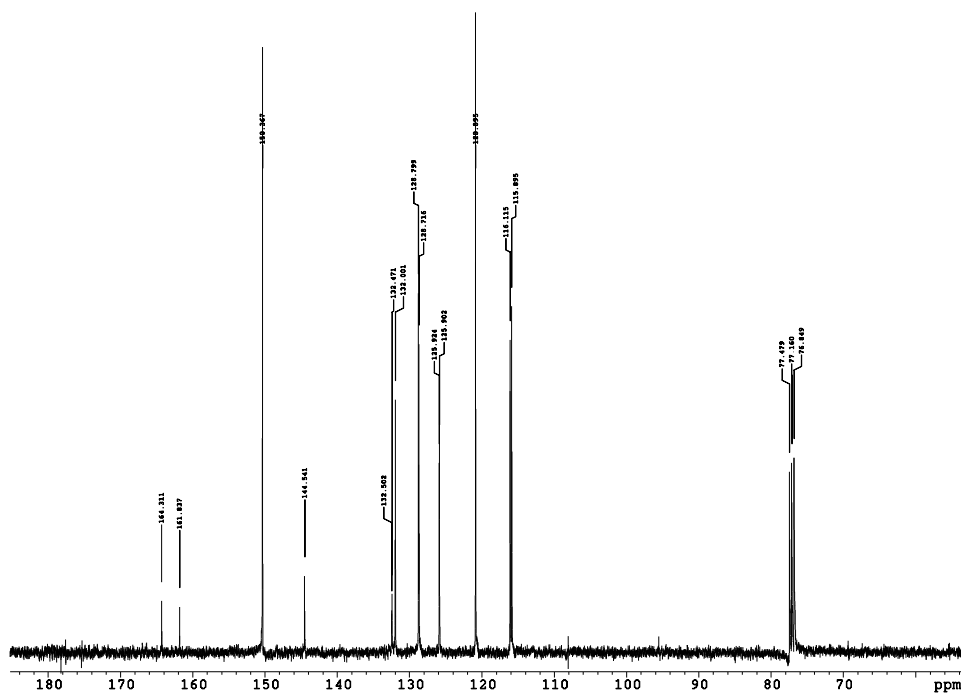
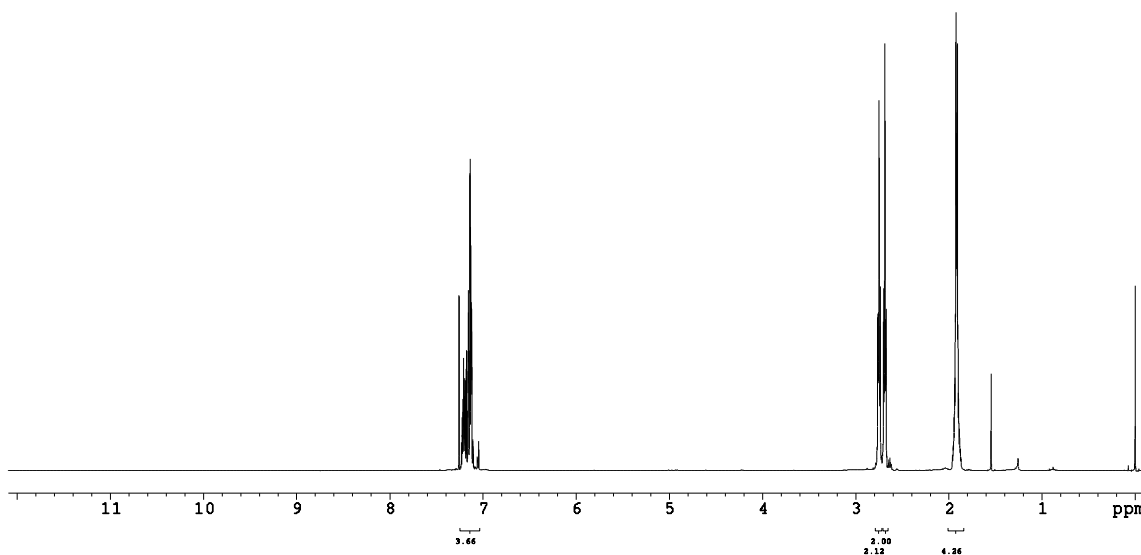


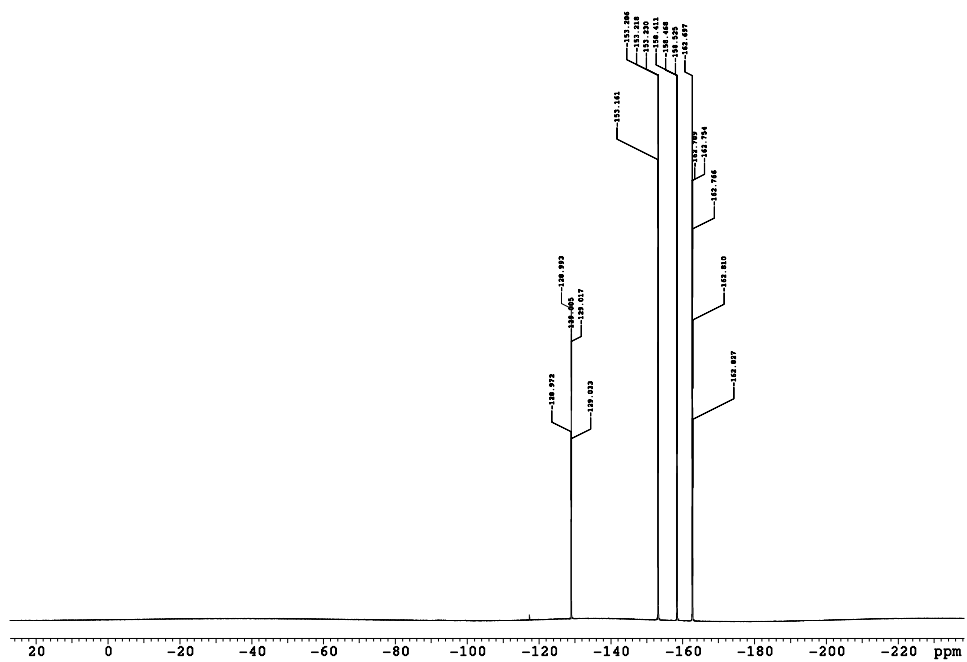
### Adipic acid *o*-fluorophenol, 2,3,4,5,6-pentafluorophenol diester (3a) (<sup>1</sup>H NMR, <sup>13</sup>C NMR, <sup>19</sup>F NMR, HRMS)

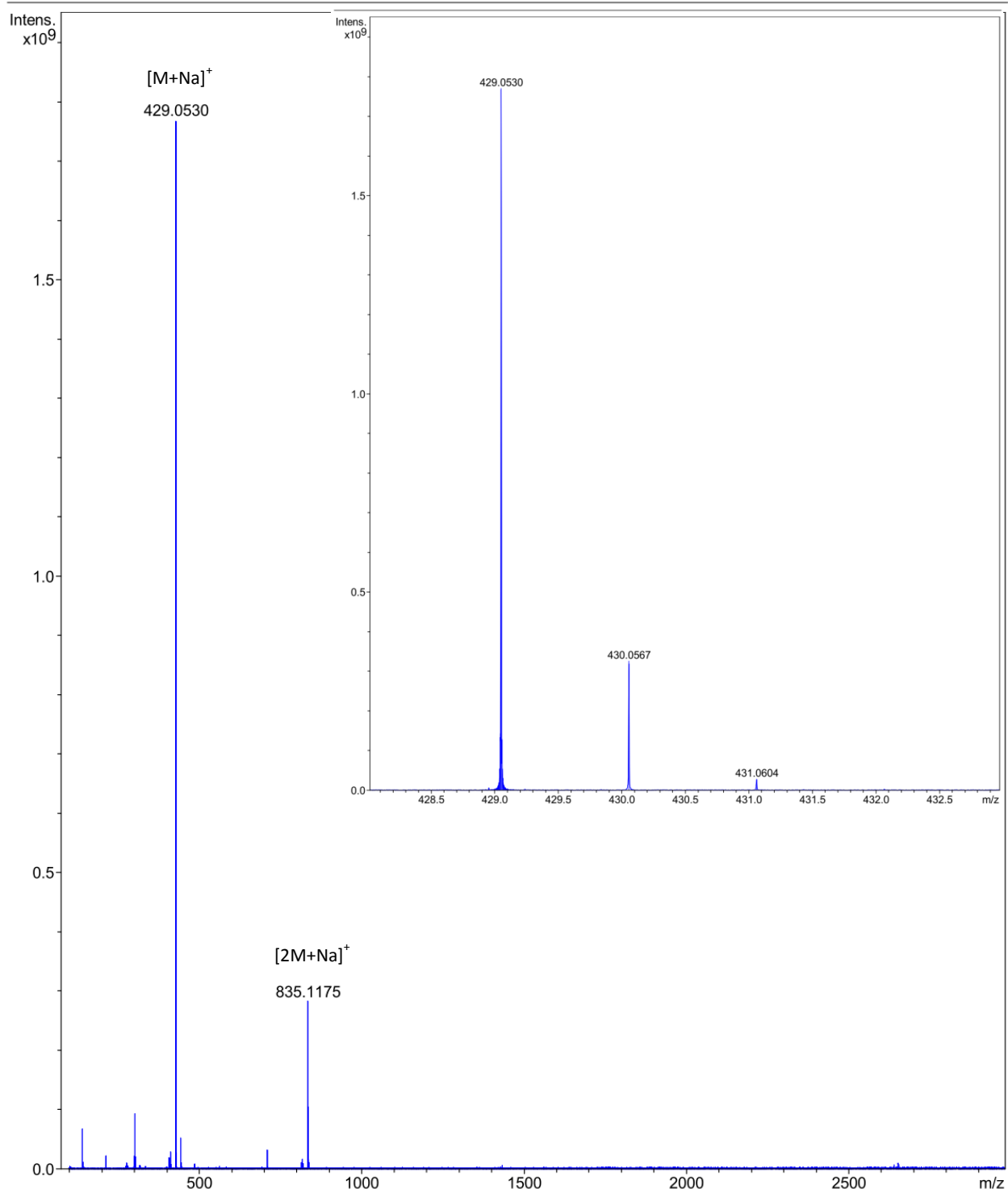
<sup>1</sup>H NMR (500 MHz, CDCl<sub>3</sub>) δ ppm 7.17 (m, 4H), 2.75 (m, 2H), 2.69 (m, 2H), 1.97 – 1.87 (m, 4H).

<sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>) δ ppm 170.4, 169.0, 155.3, 152.8, 127.1 (d, *J* = 7.2 Hz), 124.4 (d, *J* = 3.8 Hz) 123.6, 116.6 (d, *J* = 18.6 Hz), 33.3, 32.9, 24.1, 23.1.

<sup>19</sup>F NMR (376 MHz, CDCl<sub>3</sub>) δ ppm -129.2 (m, 1 F), -153.2 (m, 2 F), -158.5 (m, 1 F), -162.8 (m, 2 F).





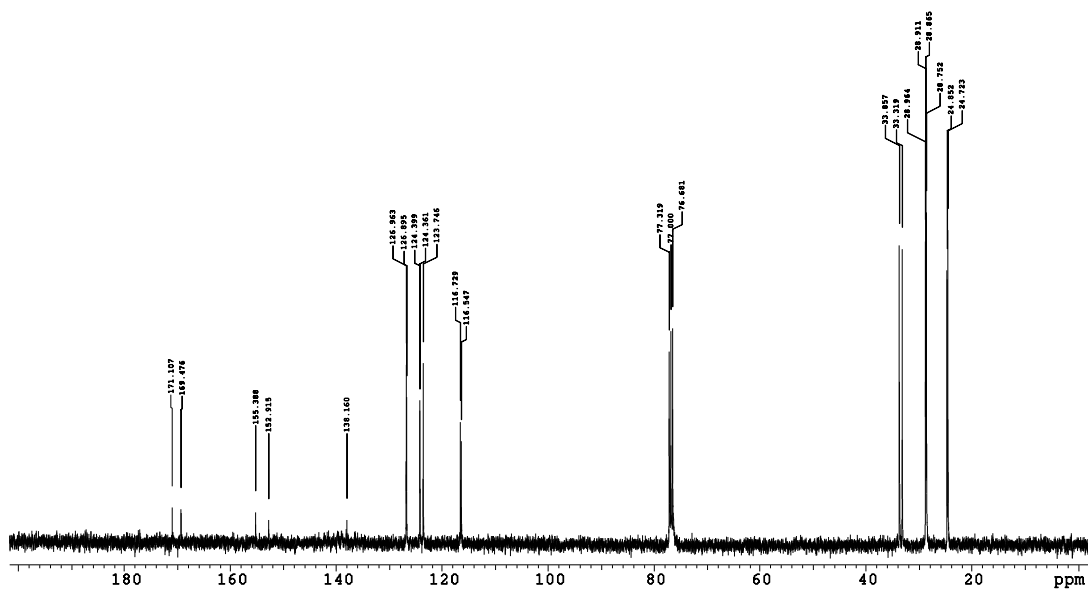
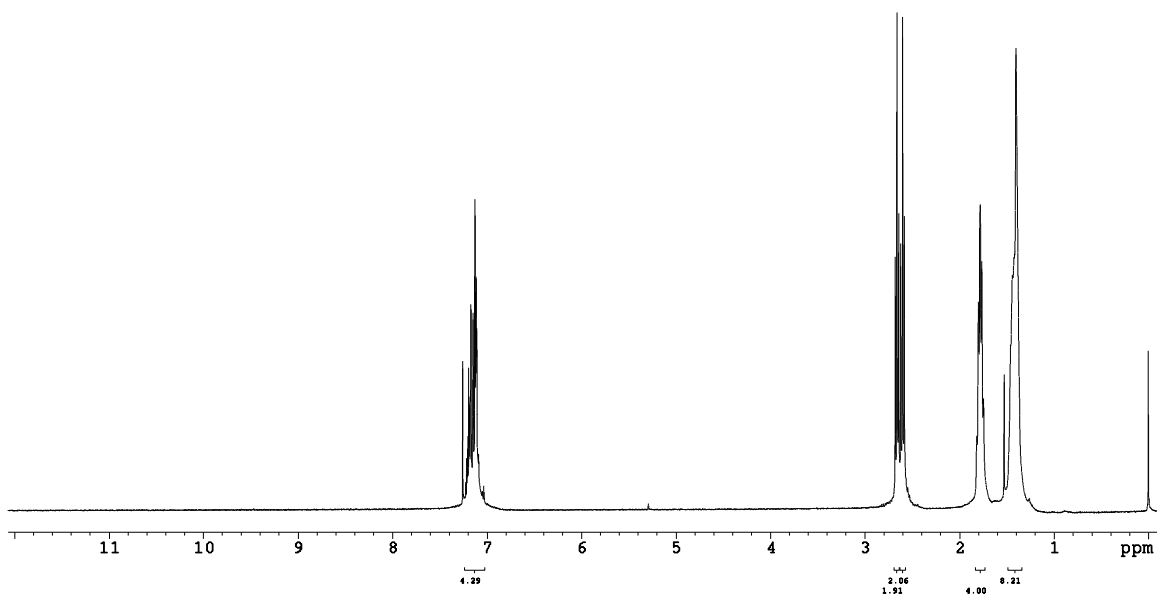


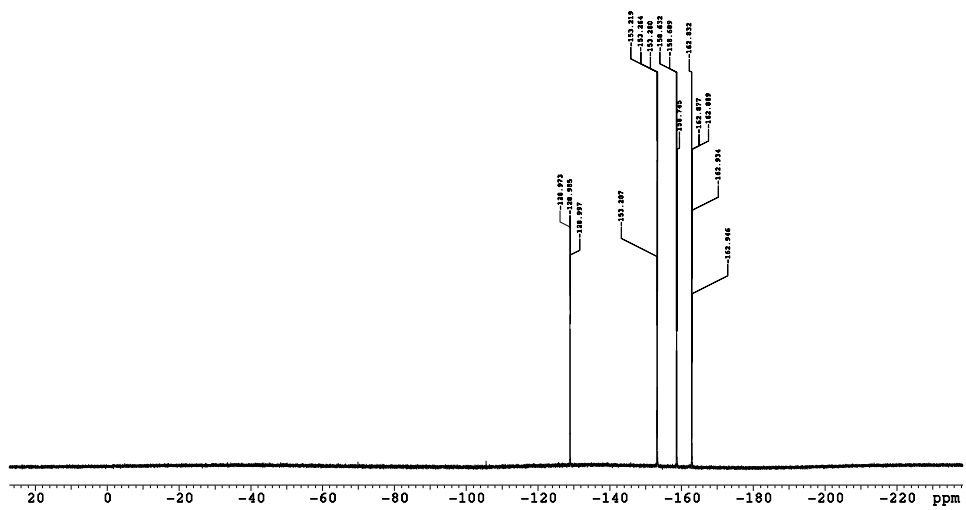
### Sebacic acid *o*-fluorophenol, 2,3,4,5,6-pentafluorophenol diester (**3b**) ( $^1\text{H}$ NMR, $^{13}\text{C}$ NMR, $^{19}\text{F}$ NMR, HRMS)

$^1\text{H}$  NMR (500 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm 7.16 (m, 4 H), 2.63 (m, 4 H), 1.79 (m, 4 H), 1.42 (m, 8 H)

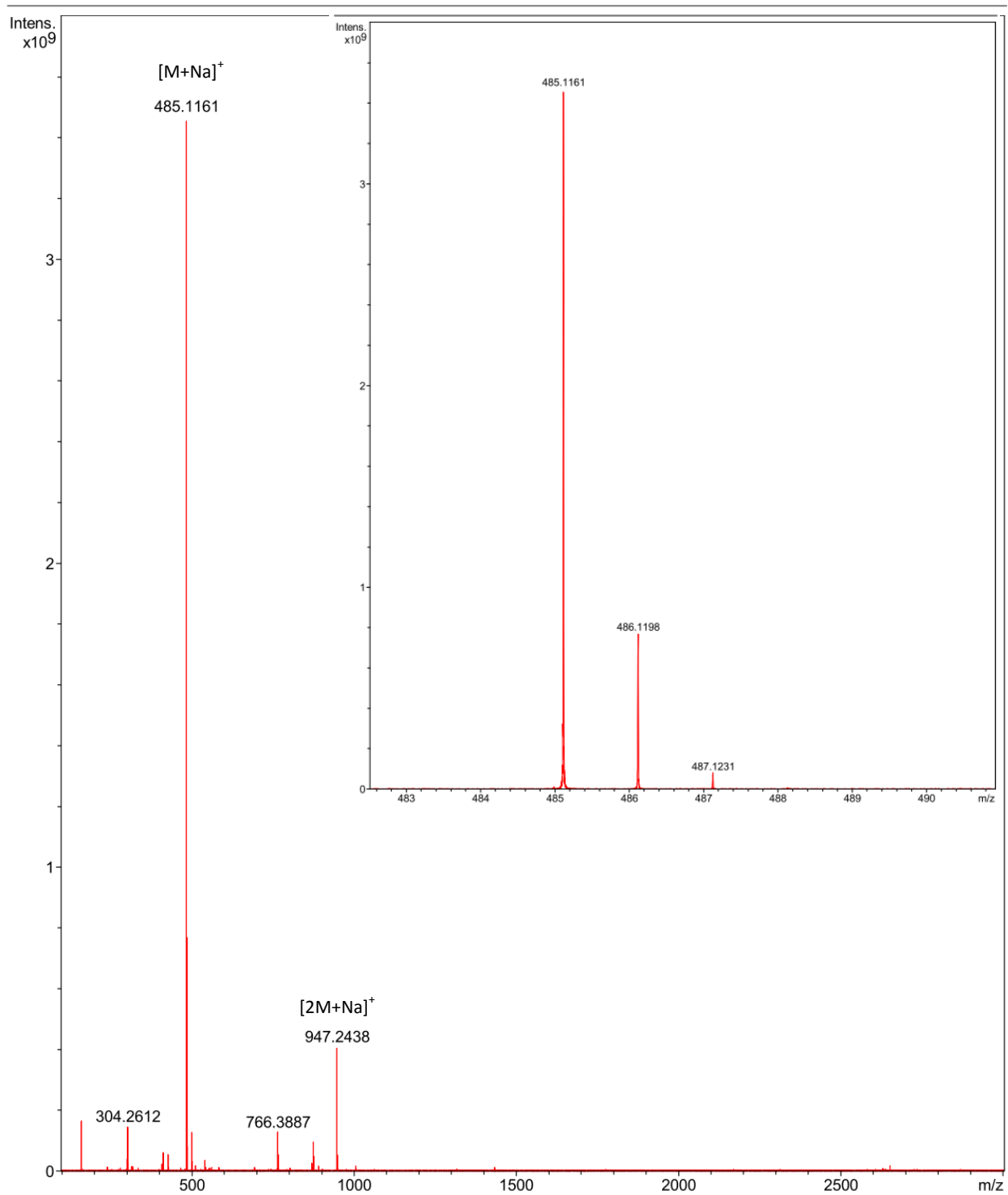
$^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm 171.3, 169.7, 155.6, 153.1, 138.3, 127.1 (d,  $J = 7.2$  Hz), 124.6 (d,  $J = 3.8$  Hz), 123.9, 116.87 (d,  $J = 18.6$  Hz), 34.1, 33.55, 29.2, 29.15, 29.1, 28.99, 25.1, 24.9.

$^{19}\text{F}$  NMR (376 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm -129.0 (m, 1 F), -153.26 (m, 2 F), -158.70 (m, 1 F), -162.9 (m, 2 F).







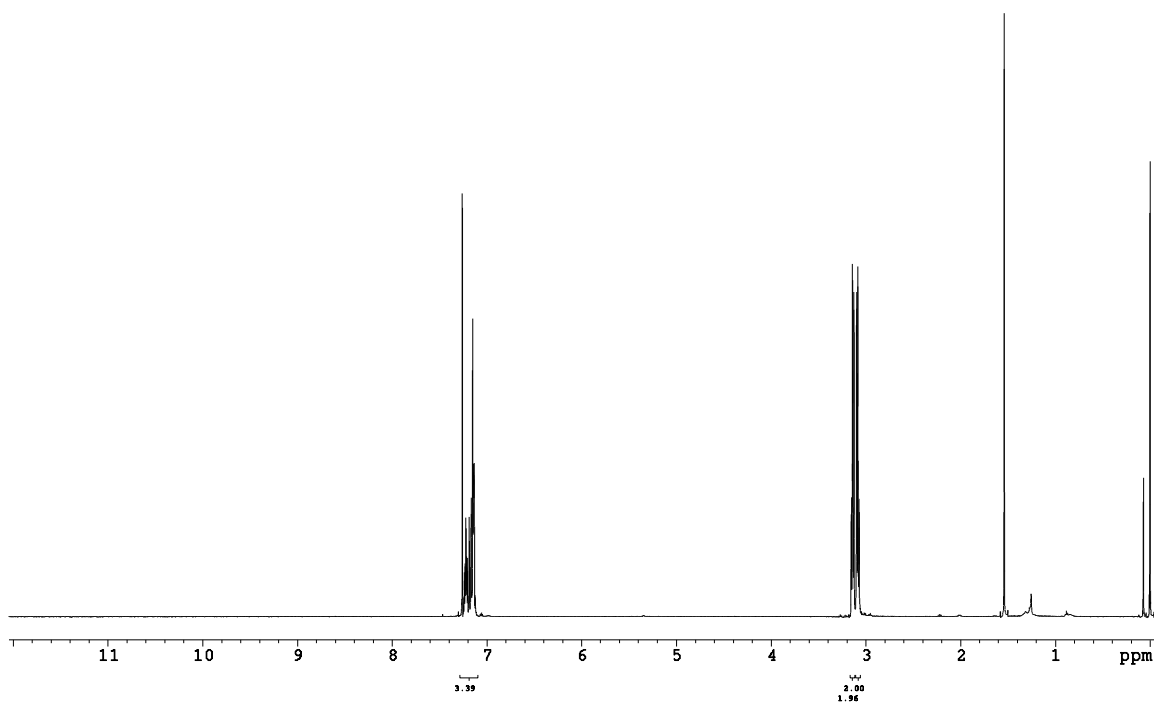


**Succinic Acid, o-fluorophenol, 2,3,4,5,6-pentafluorophenol diester (3c) (<sup>1</sup>H NMR, <sup>13</sup>C NMR, <sup>19</sup>F NMR, HRMS)**

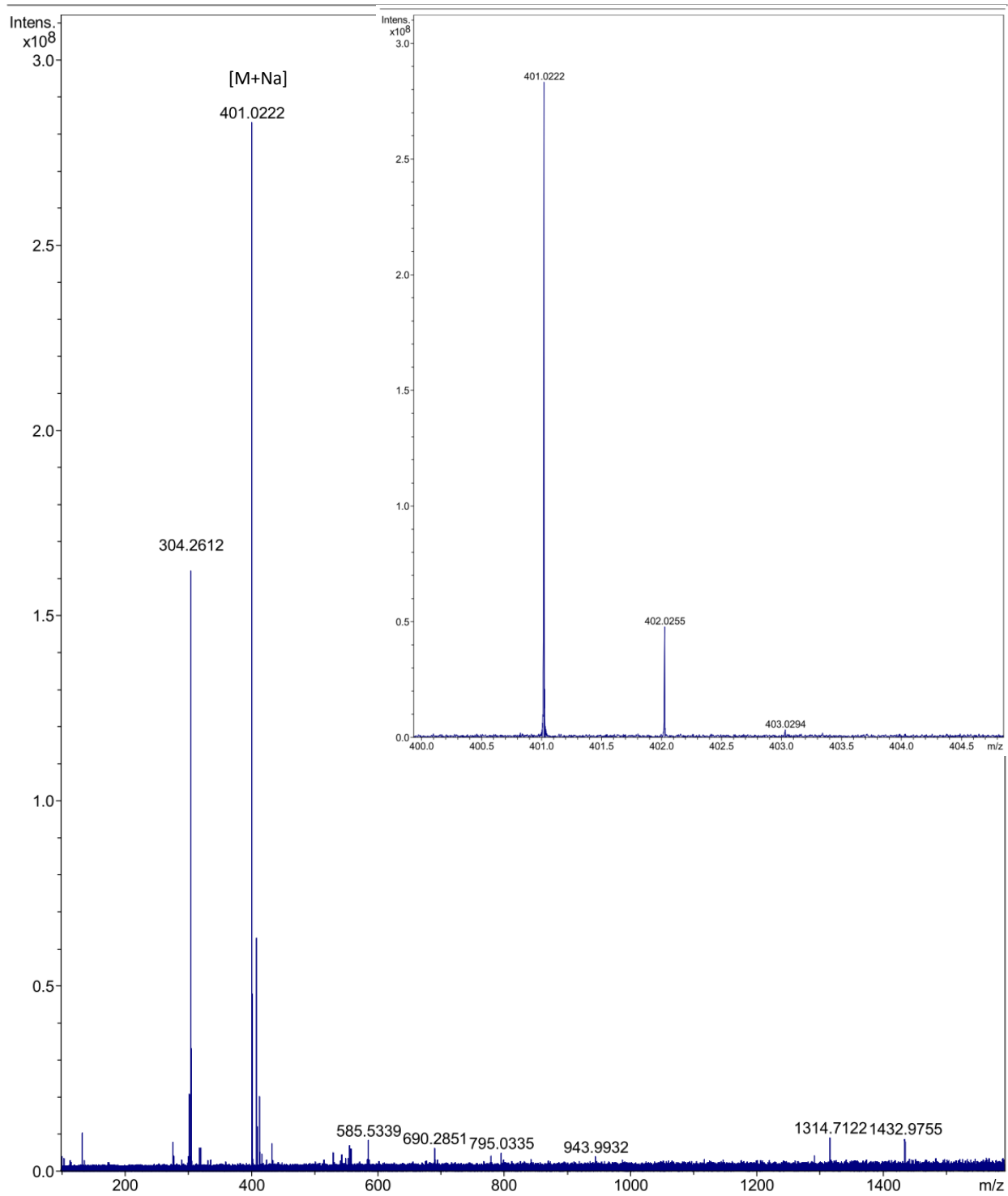
<sup>1</sup>H NMR (500 MHz, CDCl<sub>3</sub>) δ ppm 7.19 (m, 4H), 3.14 (m, 2H), 3.09 (m, 2H).

<sup>13</sup>C NMR (101 MHz, CDCl<sub>3</sub>) δ ppm 178.1, 169.0, 168.0, 155.2, 152.7, 142.8, 127.4, 127.3, 124.5, 124.4, 123.6, 116.8, 116.7, 28.6, 28.4.

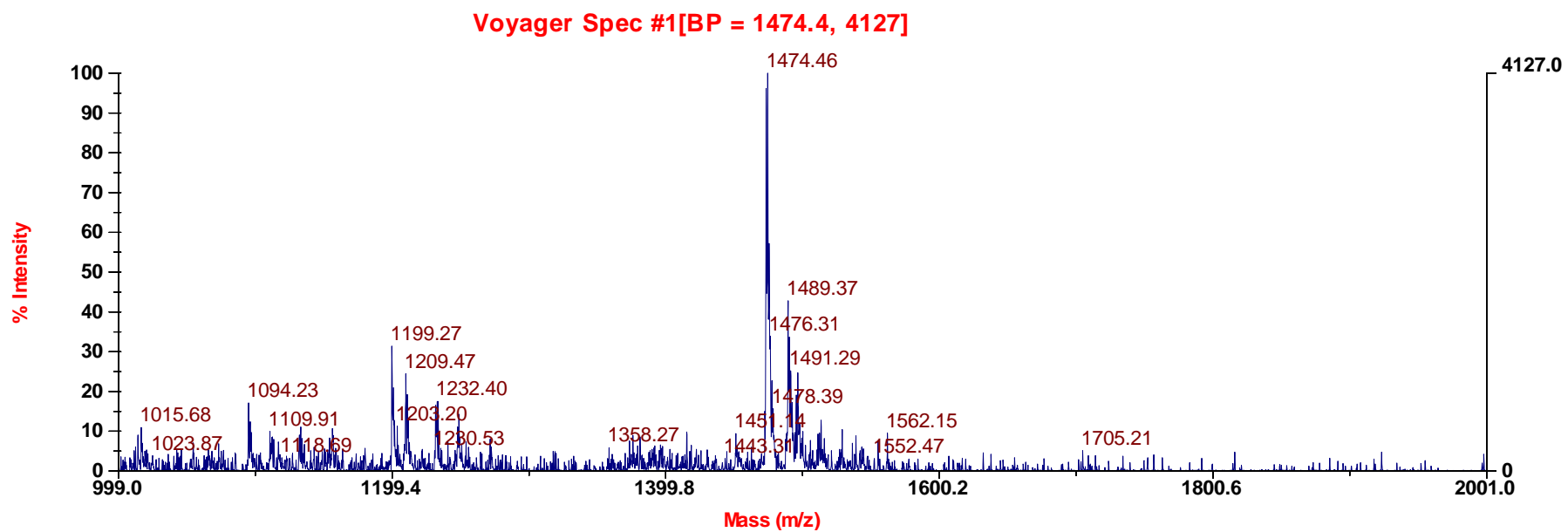
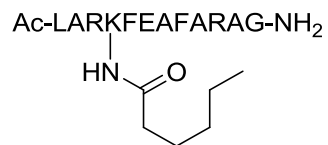
<sup>19</sup>F NMR (376 MHz, CDCl<sub>3</sub>) δ ppm -129.1 (m), -152.9 (m), -158.1 (m), -162.6 (m).





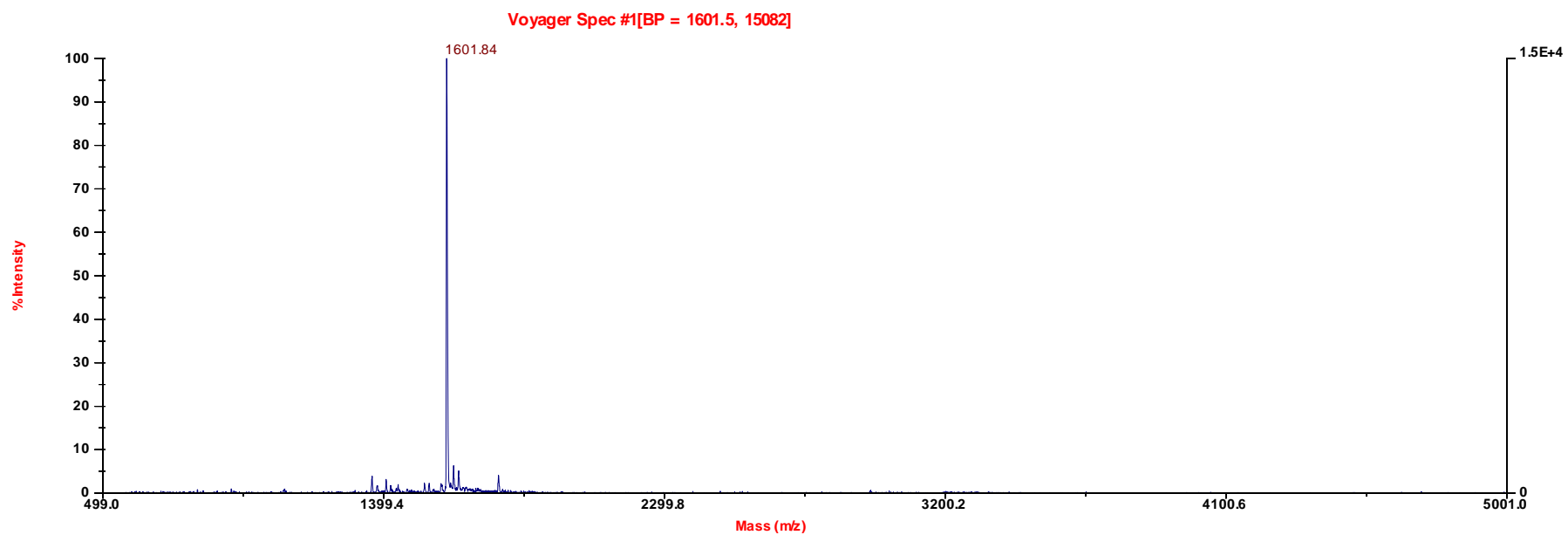
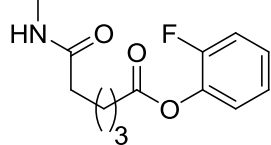


### Dodecapeptide conjugate with hexanoic acid (MALDI) form 2 and 1a-d



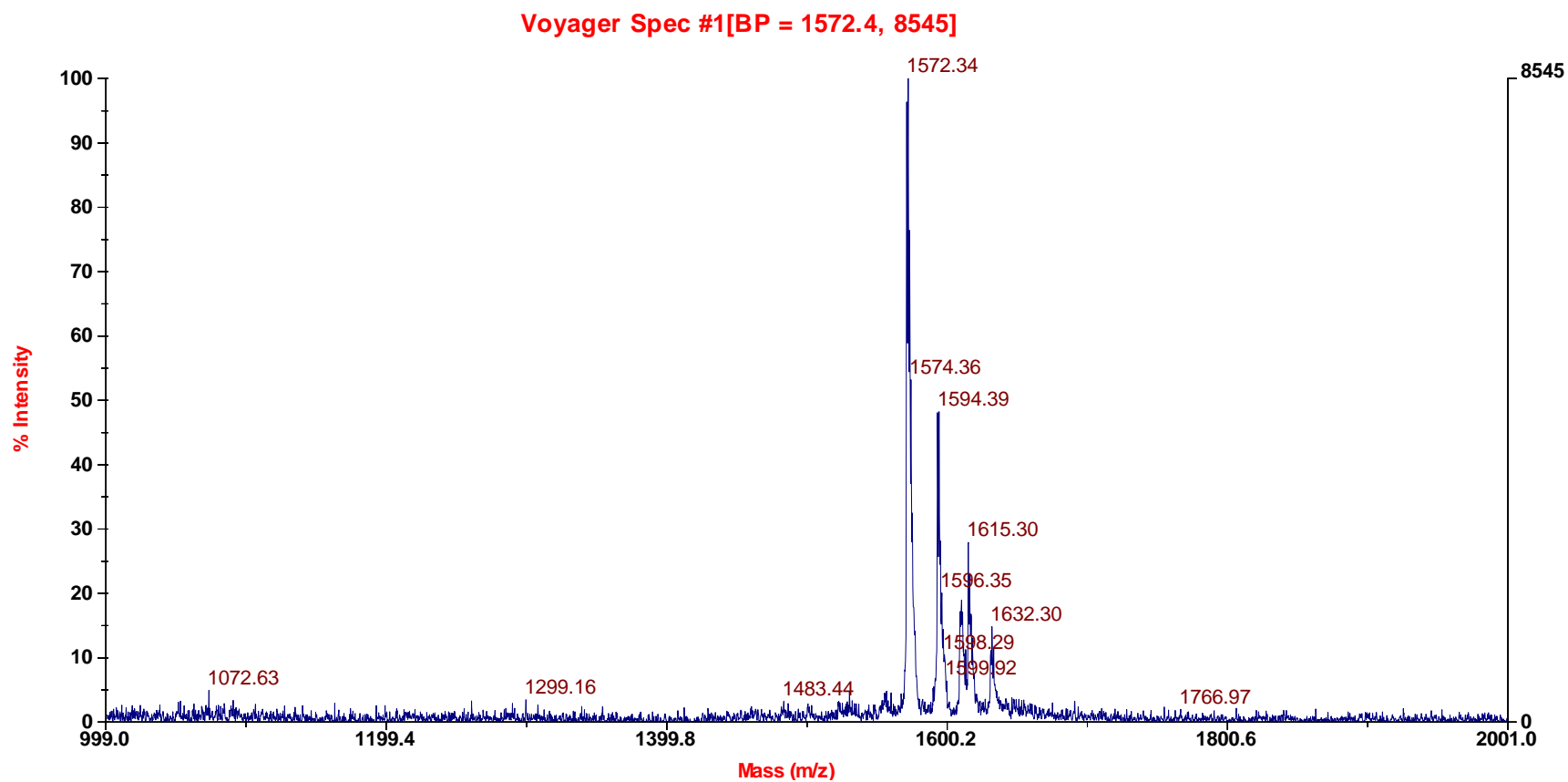
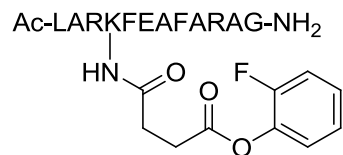
### Compound 6a (MALDI)

Ac-LARKFEAFARAG-NH<sub>2</sub>



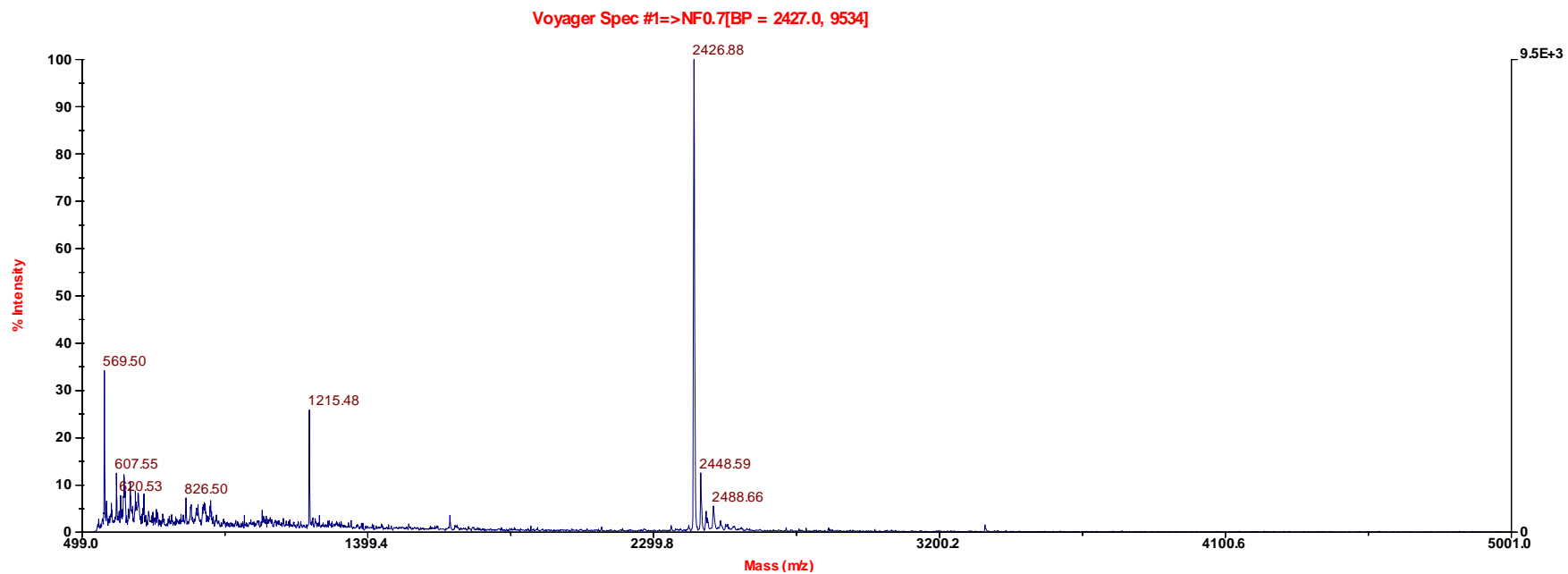
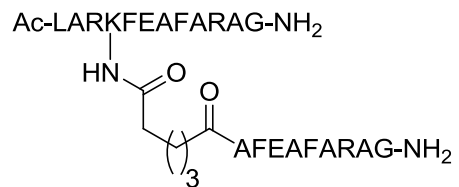


## Peptide-linker conjugate (6c) (MALDI)



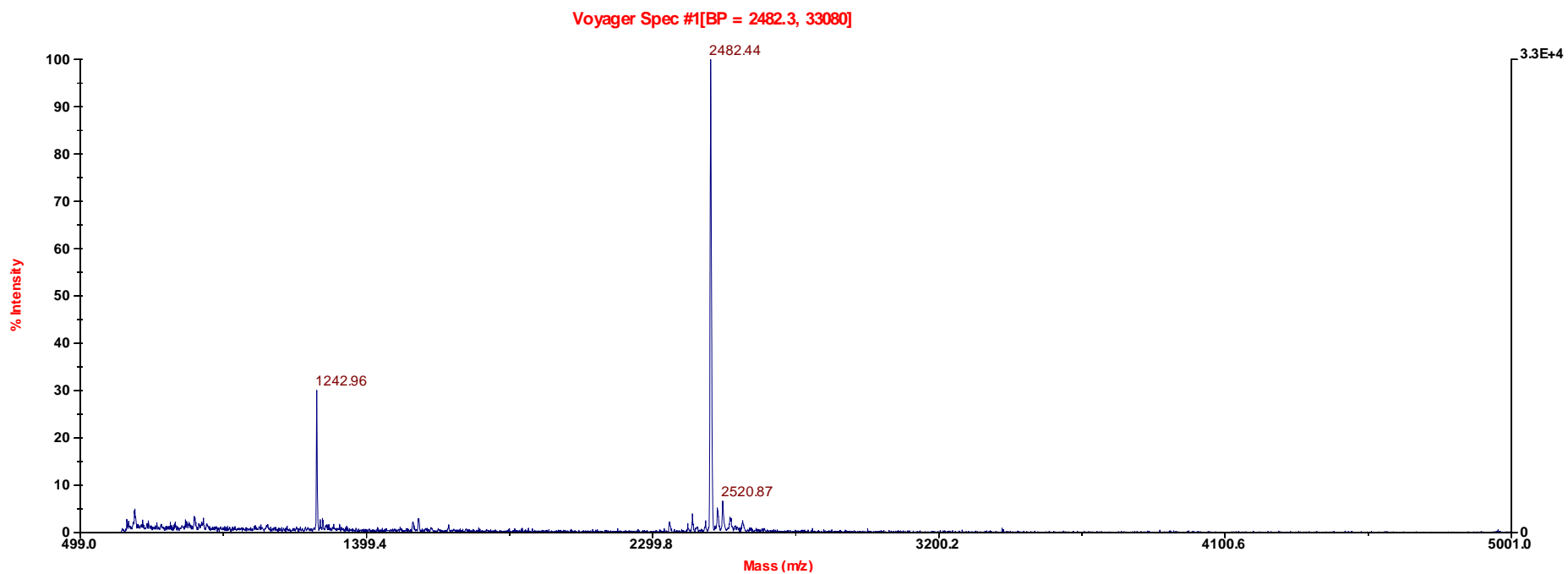
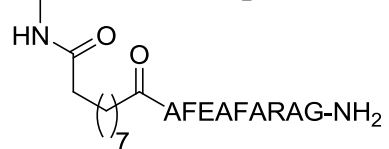


## Peptide-peptide conjugate (8a) (MALDI)



### Peptide-peptide conjugate (8b) (MALDI)

Ac-LARKFEAFARAG-NH<sub>2</sub>

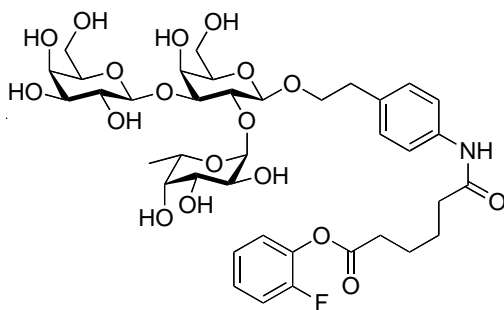


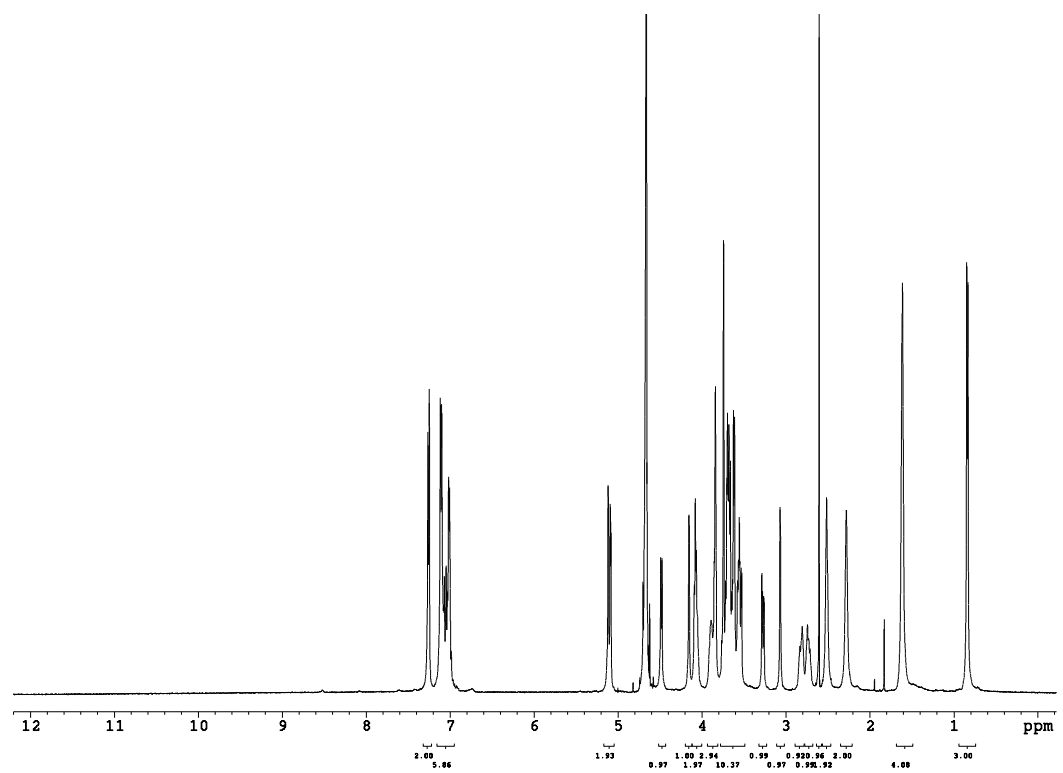
### Sugar-linker conjugate (**11**) ( $^1\text{H}$ NMR, $^{13}\text{C}$ NMR, $^{19}\text{F}$ NMR, MALDI)

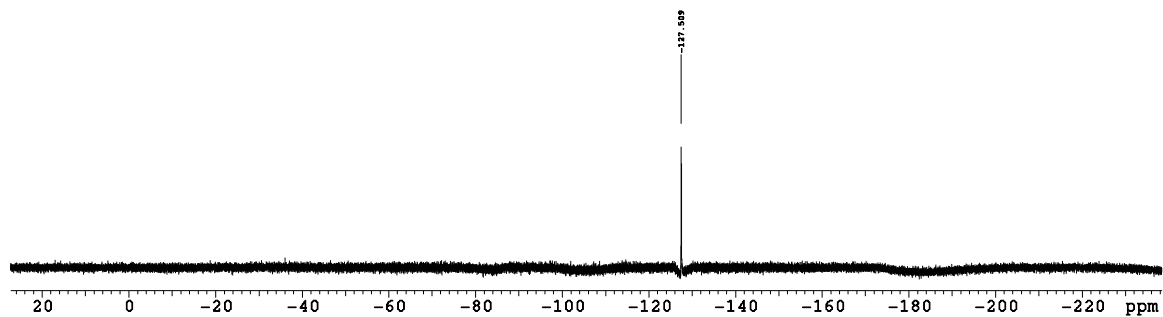
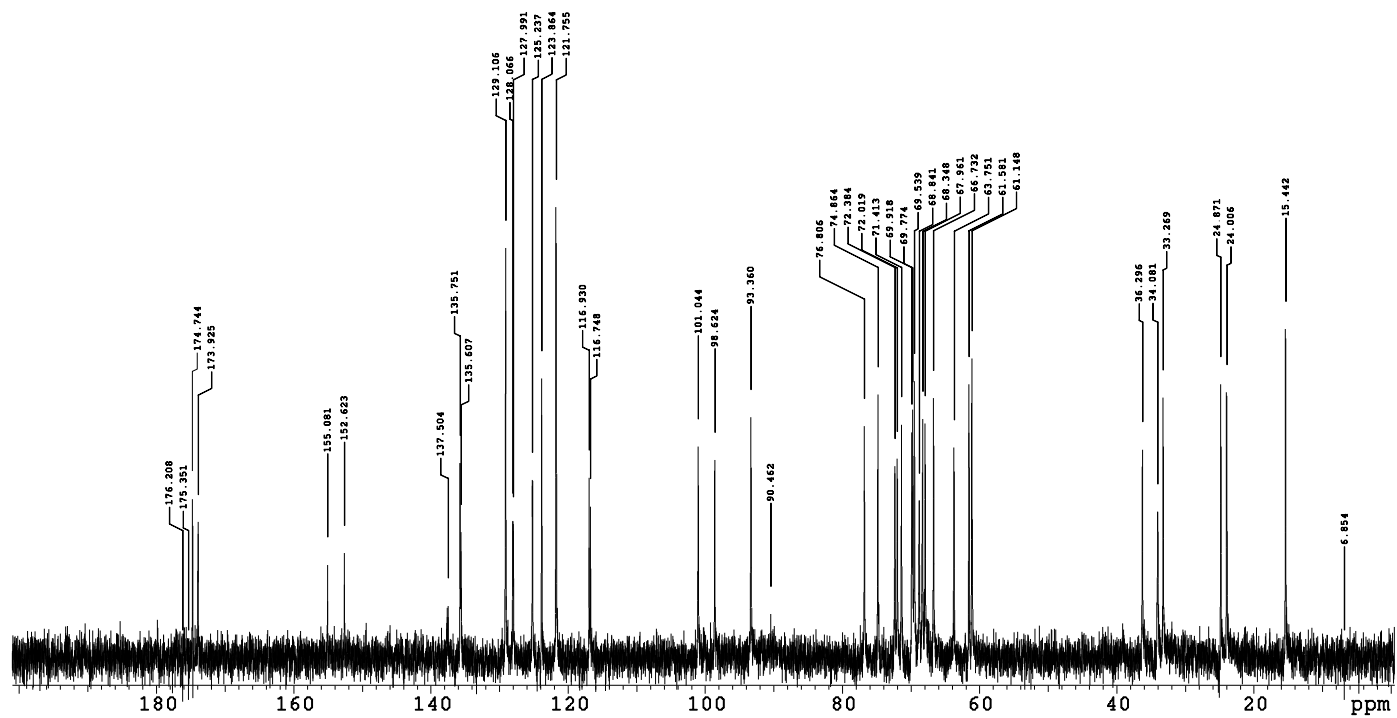
$^1\text{H}$  NMR (500 MHz,  $\text{D}_2\text{O}$ ,  $\delta$  HOD = 4.68)  $\delta$  ppm 7.28 (d,  $J$  = 8.2 Hz, 2H), 7.13 (d,  $J$  = 8.2 Hz, 2H), 7.01-7.12 (m, 4H), 5.14 (d,  $J$  = 2.1 Hz, 1H), 5.11 (d,  $J$  = 4.0 Hz, 1H), 4.50 (d,  $J$  = 7.8 Hz, 1H), 3.54 - 4.20 (m, 16H), 3.29 (dd,  $J$  = 3.0, 10.0 Hz, 1H), 3.09 (d,  $J$  = 2.7 Hz, 1H), 2.84 (m, 1H), 2.75 (m, 1H), 2.53 (m, 2H), 2.39 (m, 2H), 1.63 (m, 4H), 0.86 (d,  $J$  = 6.4 Hz, 3H)

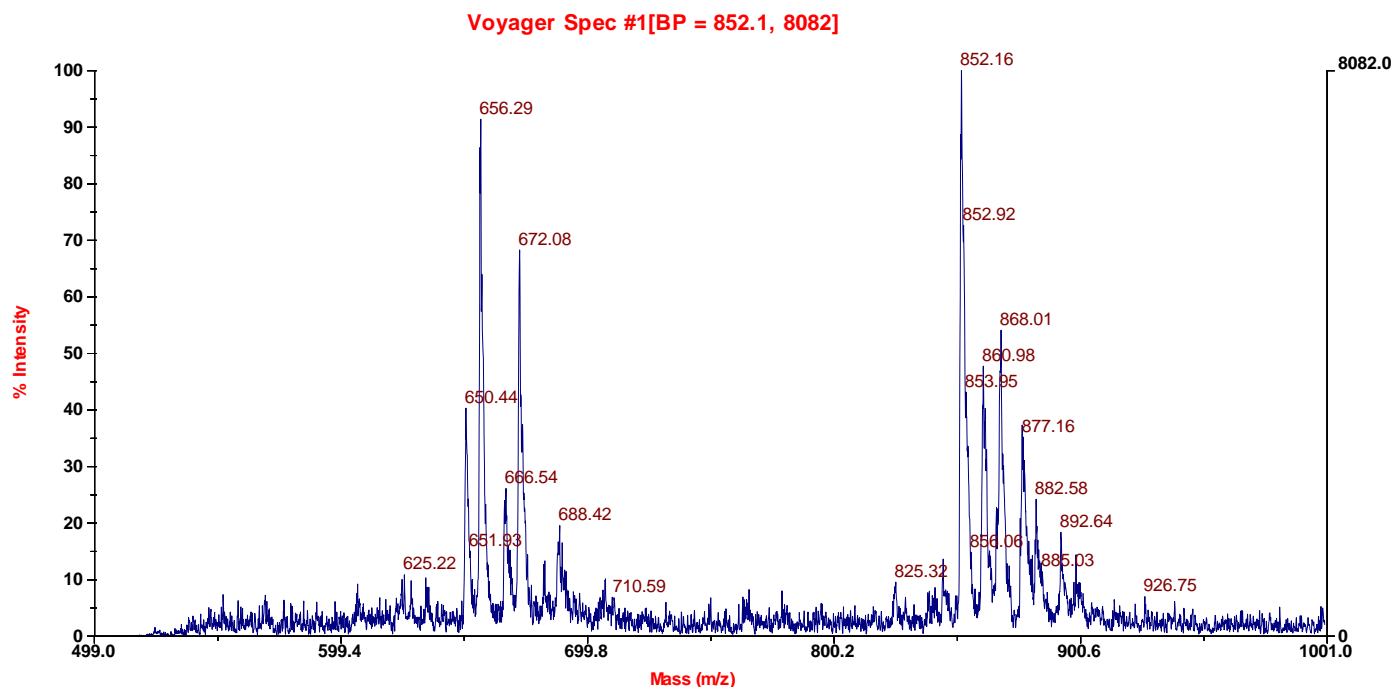
$^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm 174.7, 173.9, 155.0, 152.6, 137.6, 137.4, 135.7, 135.6, 135.5, 129.0, 128.9, 128.9, 128.8, 128.0, 127.9, 125.2, 125.1, 125.1, 123.8, 123.7, 121.7, 121.6, 121.6, 121.5, 121.5, 116.9, 116.8, 116.7, 101.0, 98.6, 93.3, 76.8, 74.8, 72.3, 72.0, 71.4, 69.9, 69.7, 69.5, 68.8, 68.3, 67.9, 66.7, 63.79, 61.5, 61.1, 36.2, 34.0, 34.0, 33.2, 24.8, 24.0, 15.4

$^{19}\text{F}$  NMR (376 MHz,  $\text{CDCl}_3$ )  $\delta$  ppm -127.5

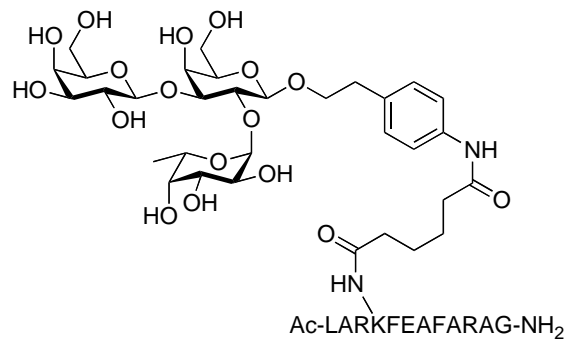




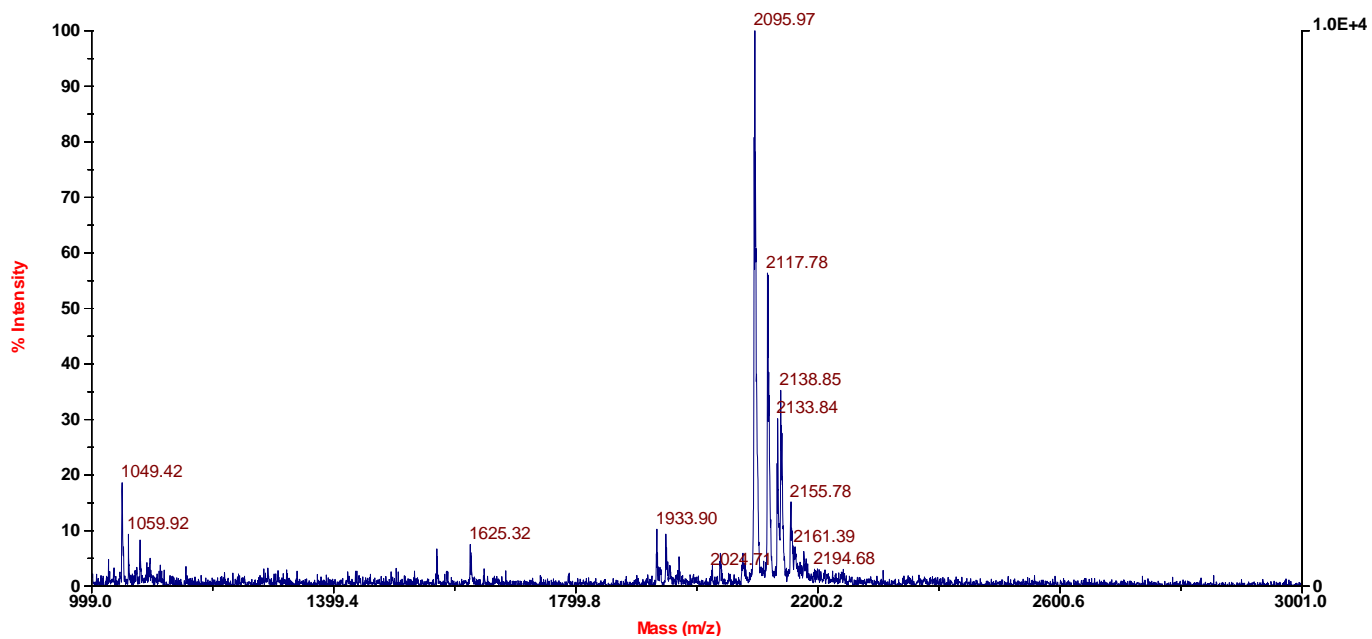




### Sugar-peptide conjugate (12) (MALDI)



Voyager Spec #1 [BP = 2096.0, 10398]



## Peptide-peptide conjugate (14) (MALDI)

