Supporting Information for:

Temperature Sensitivity Trends and Multi-Stimuli Sensitive Behavior in Amphiphilic Oligomers

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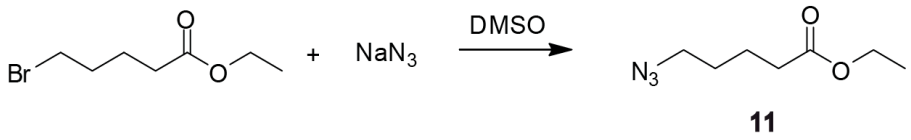
Materials and Methods :

All reagents and solvents were purchased from commercial sources and were used as received. ^{1}H NMR spectra were recorded on a 400 MHz Bruker NMR spectrometer using the residual proton resonance of the solvent as the internal standard. 13C NMR spectra were recorded on a 100 MHz Bruker spectrometer and 150 MHz Bruker spectrometer (for molecules **5**, **6**, **16** and **17**). Fluorescence spectra were recorded using a JASCO FP-6500 spectrofluorimeter. Size distribution of assemblies of oligomers were measured by a Malvern Nano Zetasizer. Turbidity studies were performed using the light scattering of a JASCO-720 spectropolarimeter.

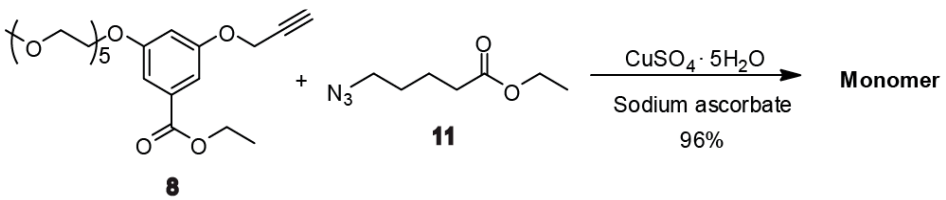
Syntheses of Oligomers:

Synthesis of ethyl-5-azidovalerate (**11**)

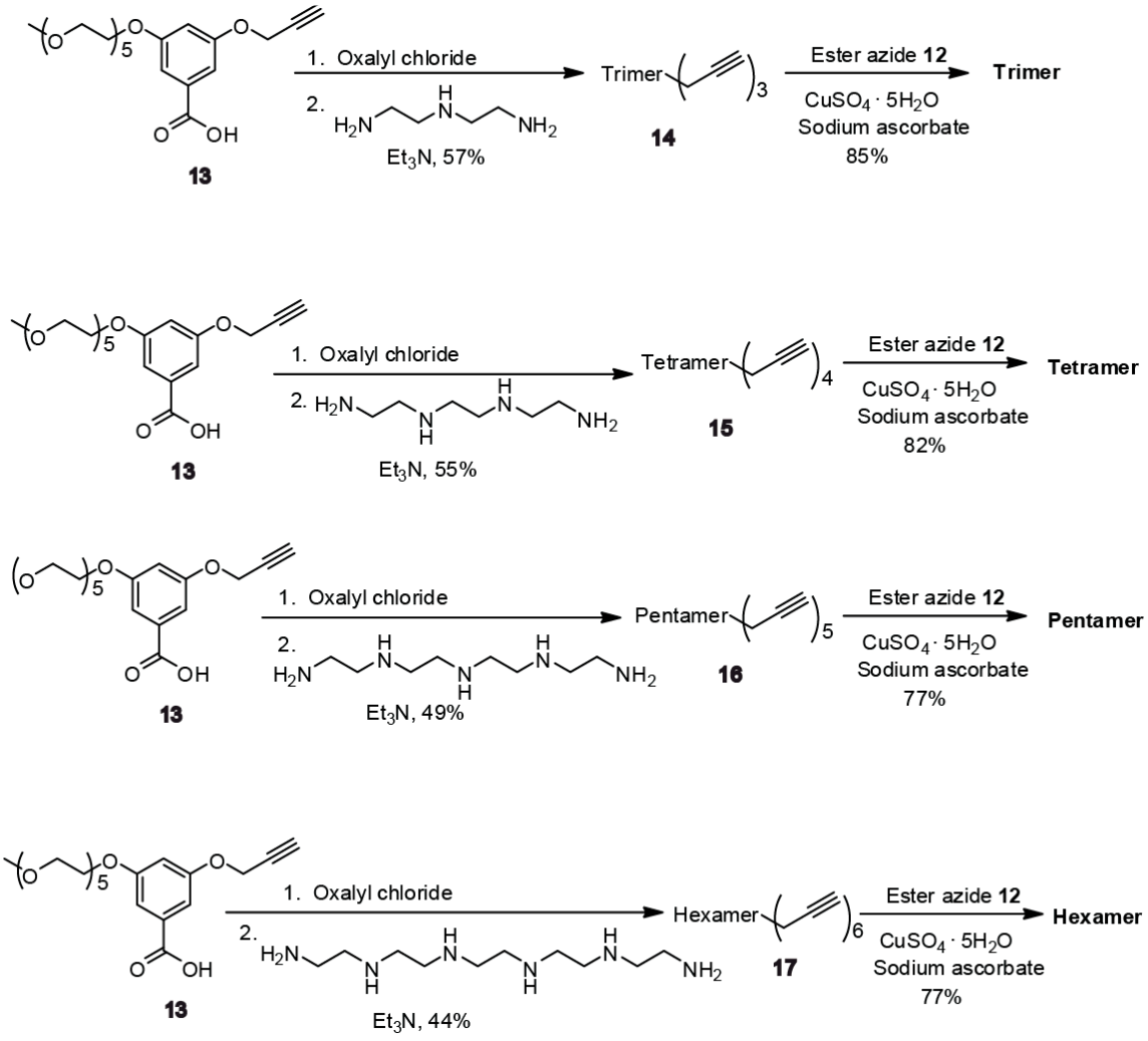


11

Synthesis of Momoner (**1**)



Syntheses of oligomers (**3**, **4**, **5** and **6**)



General procedure for the syntheses of the precursor oligomers before click reaction：

To a solution of acid monomer **13** in DCM, 2.0 equivalent of oxalyl chloride was added dropwise at 0°C. The reaction mixture was stirred at RT for 6 hours under argon atmosphere. After that, the solvent and other residues were removed out by vacuum, and the crude product was used directly in the next reaction.

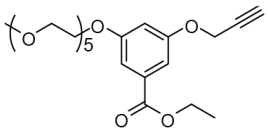
To the solution of 1.0 equivalent amine (ethylene diamine for dimer **10**, diethylene triamine for trimer **14**, and so on) in DCM, acid chloride monomer (2.5, 4.0, 6.0, 8.0 and 10.0 equiv. for dimer **10**, trimer **14**, tetramer **15**, pentamer **16** and hexamer **17** respectively) and triethyl amine (4.0, 6.0, 8.0, 10.0, 12.0 equiv. for dimer, trimer, tetramer, pentamer and hexamer respectively) were added. The mixture were

stirred under argon atmosphere for 1-7 days (1day for dimer **10**, 3days for trimer **14**, 5days for tetramer **15**, 7days for pentamer **16** and hexamer **17**). After completion of the reaction, the crude mixture was concentrated and purified by the Silica Flash Column on CombiFlash to get the product.

General procedure for the click reaction:

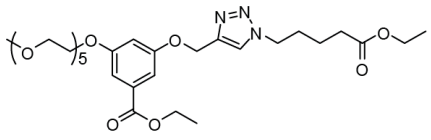
To a solution of the corresponding acetylene compounds (1.0 equiv.) in THF/H_2O (1:1), were added ethyl-5-azidovalerate (**11**) (3.0, 5.0, 8.0, 12.0 and 14.0 equiv. for dimer **2**, trimer **3**, tetramer **4**, pentamer **5** and hexamer **6** respectively), $CuSO_4$. $5H_2O$ (0.2 equiv.) and sodium ascorbate (0.2 equiv.). The mixture was stirred at 50°C for 2-7days. After the completion of the reaction, the crude mixture was quenched with saturated NH_4Cl aqueous solution, extracted with ethyl acetate (3 times) and purified by the Silica Flash Column on CombiFlash to get the product.

Characterization of molecules:



8

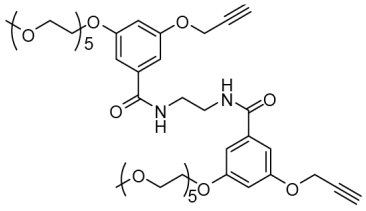
^{1}H NMR ($CDCl_3$): δ (ppm): 7.24-7.22 (m, 2H), 6.73 (s, 1H), 4.69(m, 2H), 4.35-4.33 (m, 2H), 4.14 (t, 2H), 3.85 (t, 2H), 3.68-3.61 (m, 14H), 3.53 (m, 2H), 3.35 (s, 3H), 2.54 (m, 1H), 1.36 (t, 3H). ^{13}C NMR ($CDCl_3$): δ (ppm): 166.00, 159.72, 158.41, 132.34, 108.55, 108.27, 106.85, 78.06, 75.98, 71.87, 70.19, 70.57, 70.55, 70.53, 70.51, 70.45, 69.51, 67.72, 61.11, 58.95, 56.01, 14.26. ESI-MS m/z calculated for $C_{23}H_{34}O_9$+H^+ : 455.2; Found : 455.4. ESI-MS m/z calculated for $C_{23}H_{34}O_9$+ Na^+ : 477.2; Found : 477.3.



Monomer 1

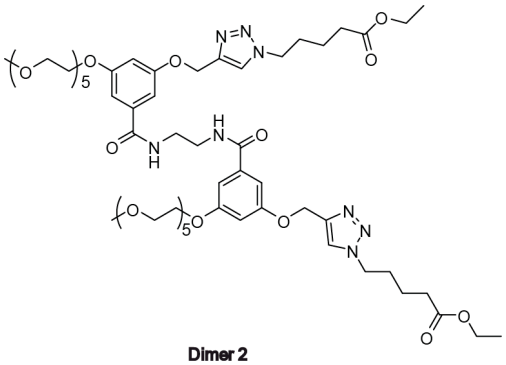
^{1}H NMR (CD_3OD): δ (ppm): 8.18 (s, 1H), 7.24 (s, 1H), 7.20 (s,1H), 6.87 (s, 1H), 5.21 (s, 2H), 4.44 (t, 2H), 4.36-4.34 (q, 2H), 4.15 (m, 2H), 4.11-4.09 (q, 2H), 3.85 (m, 2H), 3.70-3.58 (m, 14H), 3.52-3.50 (m, 2H), 3.33 (s, 3H), 2.35 (t, 2H), 1.96-1.92 (m, 2H), 1.60-1.56 (m, 2H), 1.40-1.36 (t, 3H), 1.24-1.21 (t, 3H). ^{13}C NMR (CD_3OD): δ (ppm): 173.30, 166.07, 160.06, 159.44, 132.26, 108.02, 107.95, 106.26, 71.56, 70.39, 70.21, 70.16, 70.14, 69.95, 69.31, 67.68, 60.98, 60.09, 57.73, 50.18, 32.80, 29.04, 21.50,

13.30, 13.22. ESI-MS m/z calculated for $C_{30}H_{47}N_3O_{11}+H^+$: 626.3; Found : 616.7. ESI-MS m/z calculated for $C_{30}H_{47}N_3O_{11}+ Na^+$: 648.3; Found : 648.5.



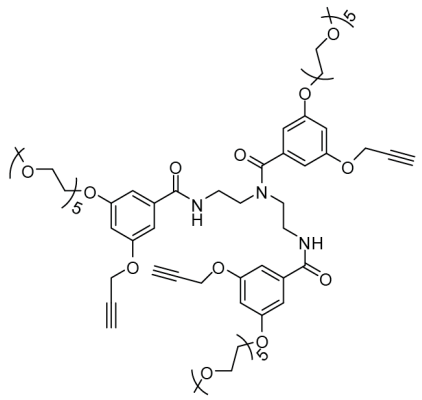
10

^{1}H NMR (CD_3OD): δ (ppm): 7.05-7.03 (m, 4H), 6.75 (m, 2H), 4.76 (m, 4H), 4.16 (m, 4H), 3.85 (m, 4H), 3.71-3.57 (m, 32H), 3.52-3.50 (m, 4H), 3.36 (m, 6H), 2.99-2.98 (m, 2H). ^{13}C NMR (CD_3OD): δ (ppm): 168.61,160.03, 158.86, 136.28, 106.32, 106.04, 104.85, 78.20, 75.99, 71.55, 70.37, 70.20, 70.18, 70.15, 70.13, 69.94, 69.33, 67.61, 57.73, 55.58, 39.56. ESI-MS m/z calculated for $C_{44}H_{64}N_2O_{16}+Na^+$: 899.9; Found : 899.5.



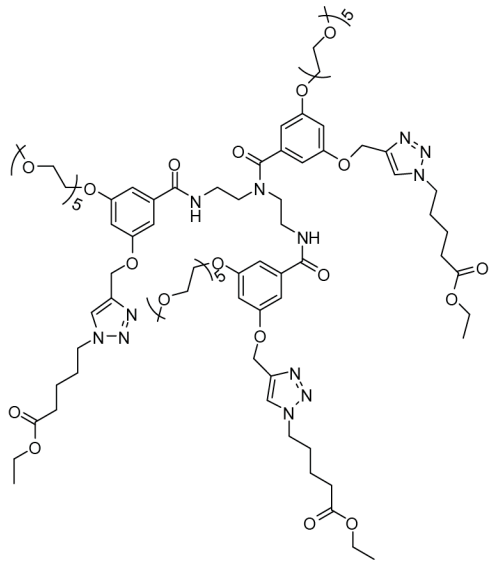
Dimer 2

^{1}H NMR (CD_3OD): δ (ppm): 8.08 (s, 2H), 7.07 (s, 2H), 7.01 (s, 2H), 6.74 (s, 2H), 5.16 (s, 4H), 4.43 (t, 4H), 4.12-4.09 (m, 8H), 3.82 (m, 4H), 3.67-3.50 (m, 36H), 3.31(m, 6H), 2.38-2.33 (t, 4H), 1.95-1.92 (m, 4H), 1.60-1.57 (m, 4H), 1.24-1.20 (t, 6H). ^{13}C NMR (CD_3OD): δ (ppm): 173.40, 168.71, 160.11, 159.47, 136.40, 106.09, 105.88, 104.84, 71.53, 70.35, 70.17, 70.13, 70.11, 69.92, 69.30, 67.59, 61.24, 60.10, 57.69, 49.64, 39.54, 32.80, 29.19, 21.45, 13.16. ESI-MS m/z calculated for $C_{58}H_{90}N_8O_{20}+Na^+$: 1242.3; Found : 1241.8.



14

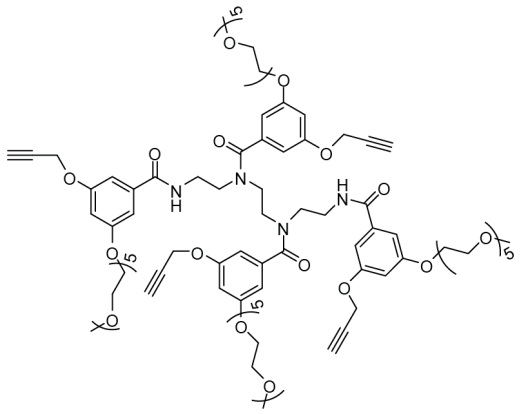
^{1}H NMR (CD_3OD): δ (ppm): 7.04-6.96 (m, 4H), 6.74 (m, 2H), 6.51 (m, 1H), 6.39 (s, 1H), 6.34 (s, 1H), 4.75 (m, 4H), 4.14-4.11 (m, 4H), 3.82- 3.49 (m, 66H), 3.33-3.32 (m, 9H), 3.00-2.94 (m, 3H). ^{13}C NMR (CD_3OD): δ (ppm): 172.79, 168.36, 167.84, 160.12, 160.02, 158.93, 158.79, 138.05, 136.41, 135.93, 106.39, 106.04, 105.42, 105.01, 104.93, 104.86, 102.98, 78.20, 78.16, 76.11, 72.27, 70.35, 69.32, 69.23, 67.67, 67.37, 57.74, 55.63, 55.44, 44.26, 37.70, 37.34. ESI-MS m/z calculated for $C_{67}H_{97}N_3O_{24}$+Na^+ : 1351.5; Found : 1350.8.



Trimer 3

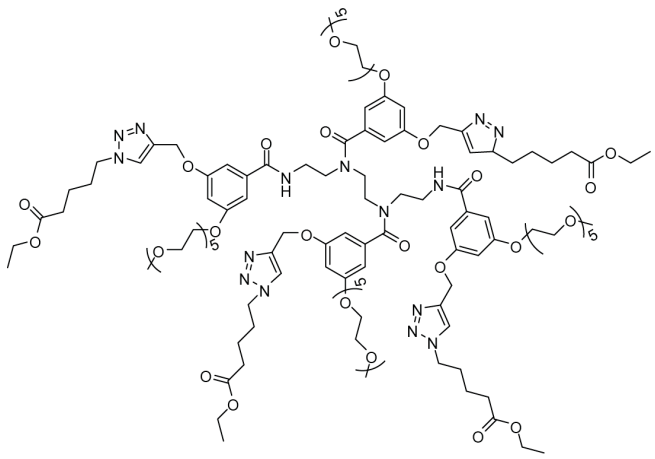
^{1}H NMR (CD_3OD): δ (ppm): 8.11-8.07 (m, 2H), 8.00 (s, 1H), 7.11-6.98 (m, 4H), 6.72 (s, 2H), 6.51-6.35 (m, 3H), 5.18-5.15 (m, 4H), 4.41-4.40 (m, 6H), 4.11-4.10 (m, 10H), 3.92-3.51 (m, 66H), 3.32(m, 9H), 2.35 (m, 6H), 1.93 (m, 6H), 1.59 (m, 6H), 1.22 (m, 9H). ^{13}C NMR (CD_3OD): δ (ppm): 173.27, 172.83,

168.39. 167.95, 160.15, 160.02, 159.50, 159.37, 143.13, 143.06, 138.19, 136.50, 136.11, 124.16, 124.05, 106.17, 105.84, 105.08, 104.94, 104.85, 103.11, 71.52, 70.33, 70.28, 70.14, 70.12, 70.08, 69.89, 69.28, 69.14, 67.59, 67.29, 61.17, 60.98, 60.10, 57.72, 49.61, 44.04, 37.68, 37.36, 32.81, 29.19, 21.50, 21.46, 13.19. ESI-MS m/z calculated for $C_{88}H_{136}N_{12}O_{30}+Na^{+}$: 1865.0; Found : 1865.0. ESI-MS m/z calculated for $C_{88}H_{136}N_{12}O_{30}+Na_2^{2+}$: 944.0; Found : 943.9.



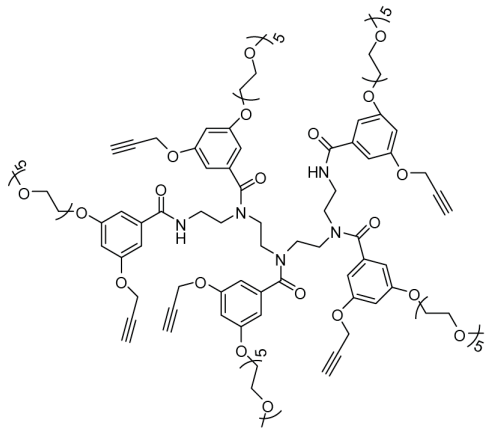
15

^{1}H NMR (CD_3OD): δ (ppm): 7.25-7.23 (m, 1H), 7.04 (m, 3H), 6.76- 6.37 (m, 8H), 4.78-4.76 (m, 5H), 4.17-4.14 (m, 5H), 3.82-3.52 (m, 90H), 3.34 (m, 12H), 3.04-2.98 (m, 4H). ^{13}C NMR (CD_3OD): δ (ppm): 172.65, 172.29, 168.29, 167.73, 160.14, 159.98, 158.93, 158.85, 138.04, 137.56, 136.33, 135.87, 106.38, 106.06, 105.52, 105.20, 104.87, 103.32, 103.04, 78.23, 76.19, 76.12, 76.08, 71.56, 70.36, 70.27, 70.18, 70.16, 70.14, 69.94, 69.30, 69.14, 67.69, 67.61, 67.32, 57.74, 55.64, 55.42, 42.31, 37.61. ESI-MS m/z calculated for $C_{90}H_{130}N_4O_{32}+Na^{+}$: 1802.9; Found : 1802.8. ESI-MS m/z calculated for $C_{90}H_{130}N_4O_{32}+Na_2^{2+}$: 912.9; Found : 912.9.



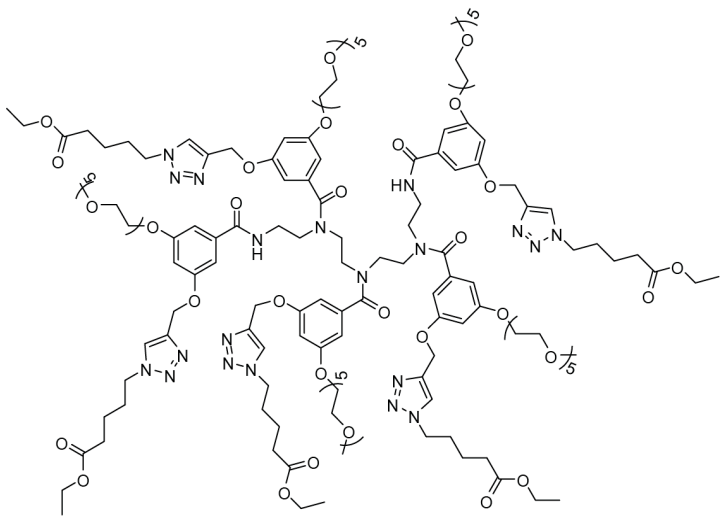
Tetramer 4

^{1}H NMR (CD_3OD): δ (ppm): 8.09-7.96 (m, 4H), 7.26-6.34 (m, 12H), 5.20-5.12 (m, 5H), 5.04 (m, 1H), 4.73 (s, 1H), 4.46-4.35 (m, 8H), 4.17-4.07 (m, 14H), 3.95-3.50 (m, 87H), 3.33-3.30 (m, 12H), 2.36-2.34 (m, 8H), 1.92-1.91 (m, 8H), 1.60-1.57 (m, 8H), 1.23-1.22 (m, 12H). ^{13}C NMR (CD_3OD): δ (ppm): 173.35, 172.65, 172.37, 168.33, 167.76, 160.18, 160.12, 160.02, 159.50, 159.41, 143.13, 138.18, 137.78, 136.47, 136.02, 124.17, 124.02, 106.15, 105.78, 105.71, 105.48, 105.25, 105.13, 104.95, 104.67, 103.26, 103.01, 71.56, 70.35, 70.26, 70.17, 70.15, 70.13, 69.94, 69.30, 69.11, 67.61, 67.30, 61.20, 61.04, 60.10, 57.73, 49.62, 43.48, 42.41, 37.63, 36.82, 32.82, 29.21, 21.50, 13.22. ESI-MS m/z calculated for $C_{118}H_{182}N_{16}O_{40}+Na_2^{2+}$: 1255.3; Found : 1255.0. ESI-MS m/z calculated for $C_{118}H_{182}N_{16}O_{40}+Na_3^{3+}$: 844.5; Found : 844.5.



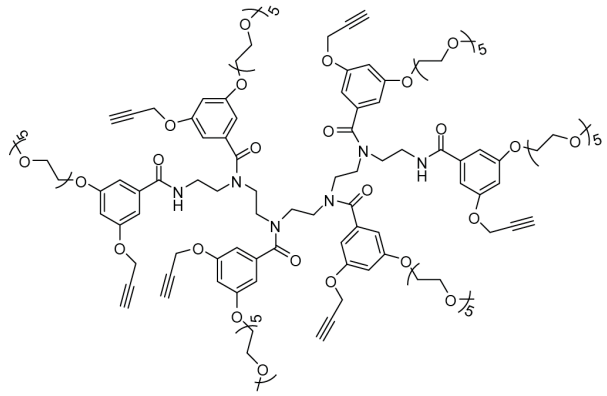
16

^{1}H NMR (CD_3OD): δ (ppm): 7.24- 6.97 (m, 5H), 6.76- 6.41 (m, 10H), 4.76 (m, 7H), 4.15 (m,4H), 4.05-3.51 (m, 115H), 3.34 (m, 15H), 3.03-2.97 (m, 5H). ^{13}C NMR (CD_3OD): δ (ppm): 172.48, 172.34, 172.14, 172.06, 168.13, 167.61, 167.50, 160.12, 159.98, 159.93, 159.58, 158.91, 158.85, 158.73, 158.41, 138.04, 137.92, 137.71, 137.60, 136.39, 135.89, 133.77, 131.92, 108.33, 108.25, 108.22, 108.07, 107.84, 106.54, 106.44, 106.11, 105.99, 105.56, 105.35, 105.11, 104.87, 103.39, 103.28, 103.00, 78.39, 76.48, 76.44, 76.35, 71.59, 70.38, 70.30, 70.19, 70.16, 69.97, 69.33, 69.21, 67.74, 67.68, 57.82, 55.72, 43.71, 41.78, 37.73, 36.87. ESI-MS m/z calculated for $C_{113}H_{163}N_5O_{40}+Na_2^{2+}$: 1138.7; Found : 1138.4. ESI-MS m/z calculated for $C_{113}H_{163}N_5O_{40}+Na_3^{3+}$: 766.8; Found : 766.8.



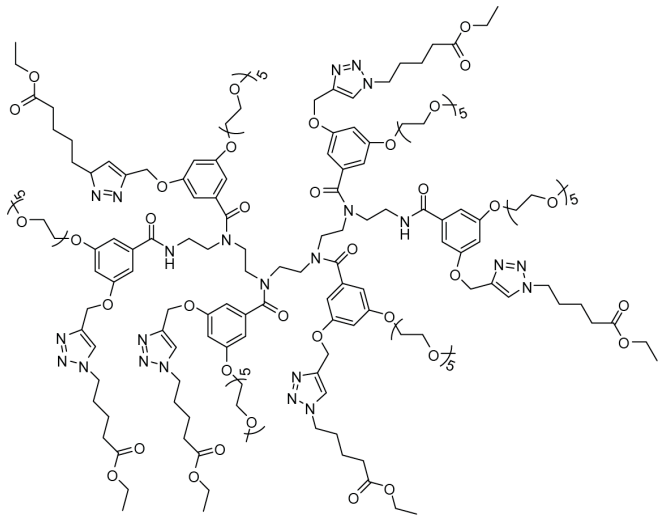
Pentamer 5

^{1}H NMR (CD_3OD): δ (ppm): 8.10-7.96 (m, 5H), 7.10-7.00 (m, 5H), 6.81-6.37 (m, 10H), 5.20-4.92 (m, 10H), 4.44-4.35 (m, 8H), 4.15-4.06 (m, 15H), 3.95-3.50 (m, 113H), 3.33 (m, 15H), 2.35-2.33 (m, 10H), 1.94-1.91 (m, 10H), 1.60-1.55 (m, 10H), 1.24-1.20 (m, 15H). ^{13}C NMR (CD_3OD): δ (ppm): 173.44, 173.38, 172.62, 172.37, 172.25, 168.38, 167.68, 160.23, 160.20, 160.16, 160.05, 159.58, 159.50, 159.42, 143.16, 143.08, 138.07, 137.81, 137.67, 136.49, 136.00, 124.16, 123.99, 108.09, 106.26, 106.16, 105.95, 105.86, 105.73, 105.40, 105.18, 105.01, 103.23, 103.11, 71.56, 70.36, 70.27, 70.19, 70.17, 70.14, 69.95, 69.33, 69.30, 69.27, 69.24, 69.19, 67.63, 67.60, 67.55, 61.29, 61.21, 61.10, 60.10, 57.72, 57.68, 49.63, 49.60, 43.43, 41.75, 39.04, 37.73, 36.76, 32.82, 29.21, 21.50, 21.47, 13.21, 13.29. ESI-MS m/z calculated for $C_{113}H_{163}N_5O_{40}+Na_2^{2+}$: 1566.75; Found : 1566.2. ESI-MS m/z calculated for $C_{113}H_{163}N_5O_{40}+Na_3^{3+}$: 1052.16; Found : 1051.6.



17

^{1}H NMR (CD_3OD): δ (ppm): 7.23-7.00 (m, 6H), 6.76-6.39 (m, 12H), 4.76-4.67 (m, 10H), 4.16-4.13 (m, 7H), 4.00-3.50 (m, 133H), 3.34-3.31(m, 20H), 3.04-2.97 (m, 6H), ^{13}C NMR (CD_3OD): δ (ppm):172.55, 172.14, 172.08, 168.26, 168.16, 167.54, 160.15, 159.99, 159.96, 158.93, 158.87, 158.76, 138.03, 137.63, 136.39, 135.89, 133.45, 108.37, 108.27, 106.46, 106.10, 105.57, 105.49, 105.30, 105.15, 104.88, 103.34, 103.01, 78.34, 78.21, 76.40, 76.24, 76.02, 71.59, 71.57, 70.40, 70.38, 70.30, 70.20, 70.15, 69.96, 69.93, 69.34, 67.75, 67.70, 57.80, 57.79, 57.74, 55.72, 55.60, 43.75, 41.87, 41.76, 37.70, 36.85. ESI-MS m/z calculated for $C_{136}H_{196}N_6O_{48}+Na_2^{2+}$: 1364.7; Found : 1364.1. ESI-MS m/z calculated for $C_{136}H_{196}N_6O_{48}+Na_3^{3+}$: 917.3; Found : 917.3.



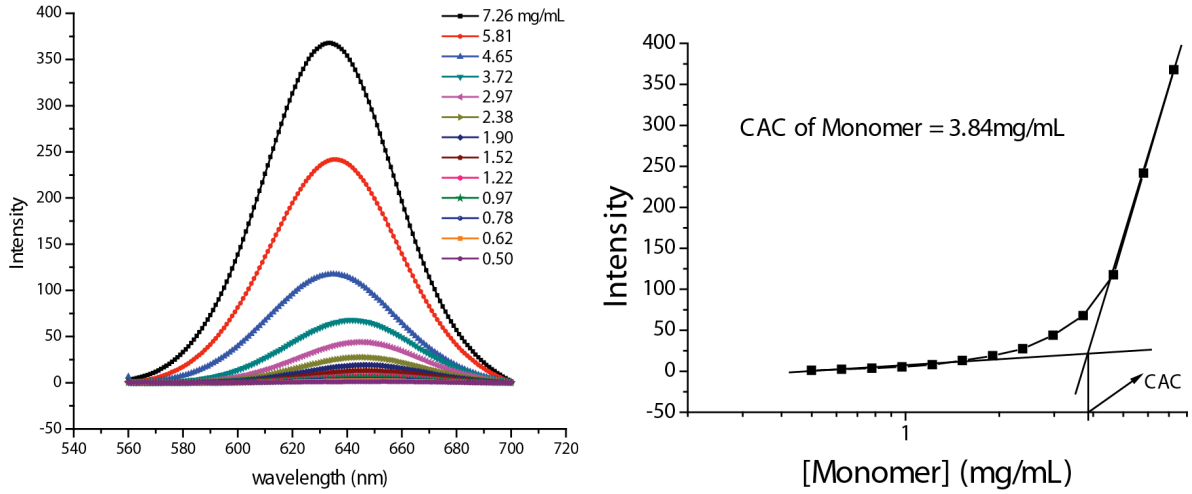
Hexamer 6

^{1}H NMR (CD_3OD): δ (ppm): 8.11-7.96 (m, 6H), 7.11-6.31 (m, 18H), 5.15-4.95 (m, 9H), 4.77 (m,1H), 4.42-4.37 (m, 10H), 4.11-3.41 (m, 156H), 3.33-3.30 (m, 18H), 2.36-2.32 (m, 12H), 1.95-1.89 (m, 12H), 1.59-1.56 (m, 12H), 1.24-1.20 (m, 18H). ^{13}C NMR (CD_3OD): δ (ppm):173.36, 172.40, 168.44, 167.60, 160.19, 160.12, 159.61, 159.50, 143.32, 138.21, 137.79, 136.65, 136.02, 124.24, 108.50, 106.17, 105.19, 104.92, 103.19, 71.57, 70.36, 70.28, 70.14, 69.95, 69.31, 67.63, 61.35, 61.25, 60.10, 57.72, 49.65, 43.28, 42.82, 41.77, 37.79, 36.78, 32.82, 29.19, 21.50, 21.44, 13.20. ESI-MS m/z calculated for $C_{178}H_{274}N_{24}O_{60}+Na_2^{2+}$: 1878.0; Found : 1878.5. ESI-MS m/z calculated for $C_{178}H_{274}N_{24}O_{60}+Na_3^{3+}$: 1259.7; Found : 1259.7.

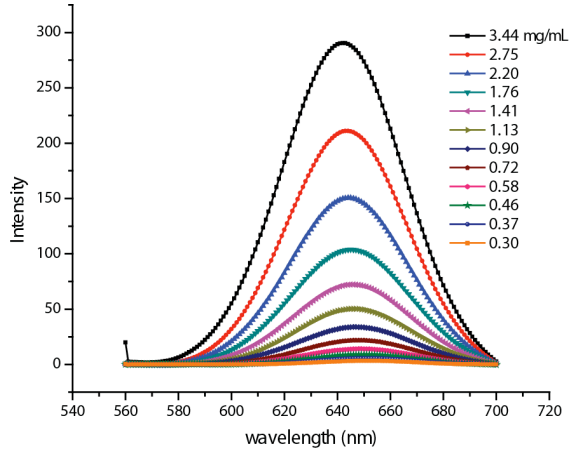
CAC measurement:

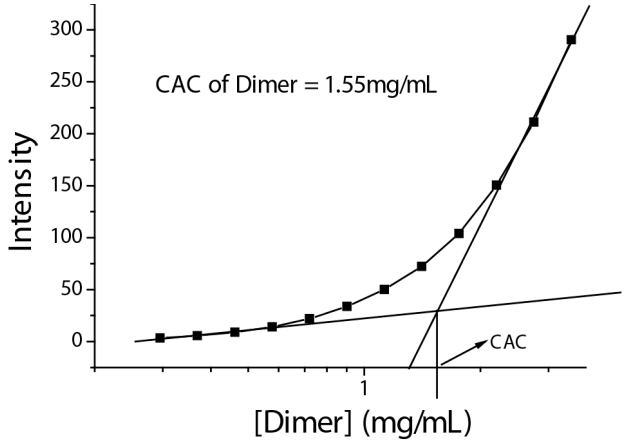
Nile red was added into oligomer solution in MilliQ water, and the mixture solution was stirred overnight at low temperature to encapsulate Nile red. Then the solution was filtered with a 0.22 um syringe filter to remove the residual Nile red. For CAC measurement, solutions with encapsulated Nile red were successively diluted by MilliQ water, and the fluorescence from Nile red was monitored. Upon each dilution, the emission intensity was plotted against the oligomer solution concentration to generate a sigmoidal curve, and the inflection point was reported as the CAC.

Monomer **1**:

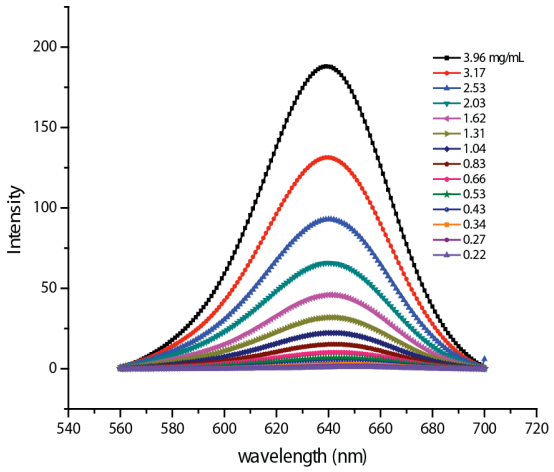


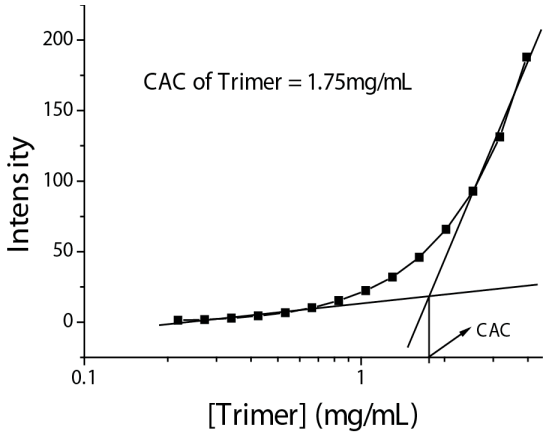
Dimer **2**:



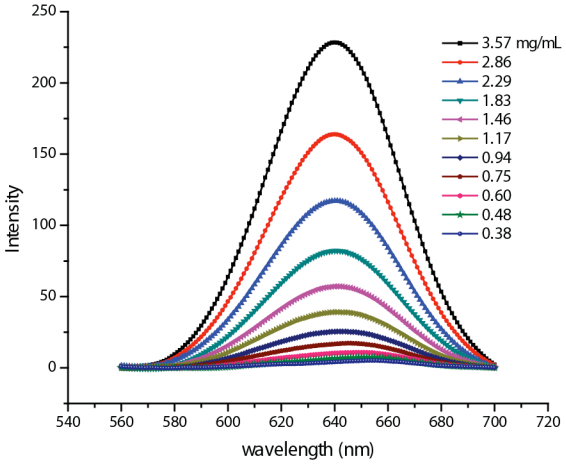


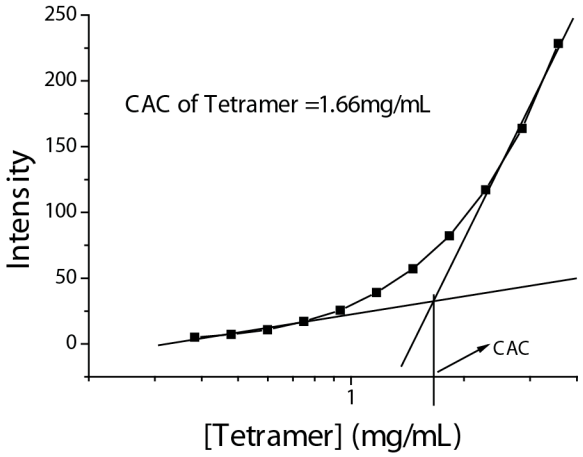
Trimer **3**:



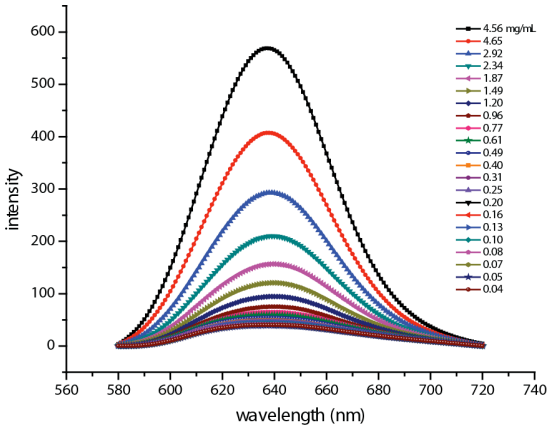


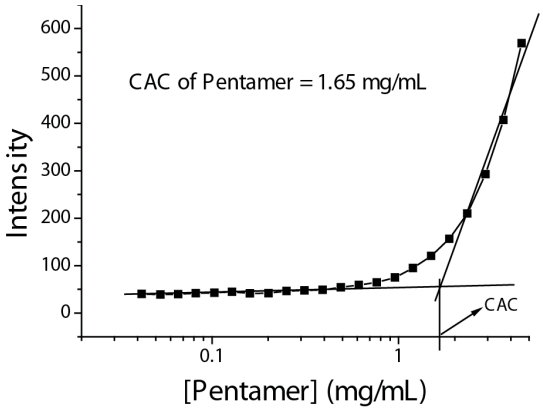
Tetramer **4**:



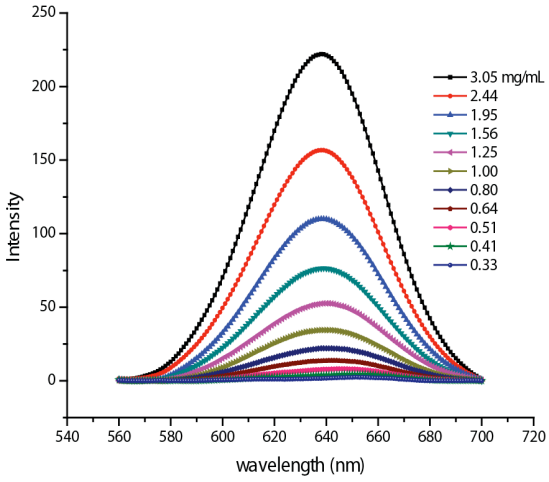


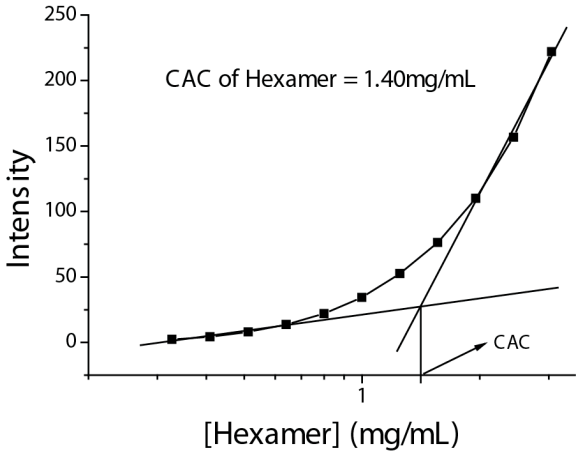
Pentamer **5**:



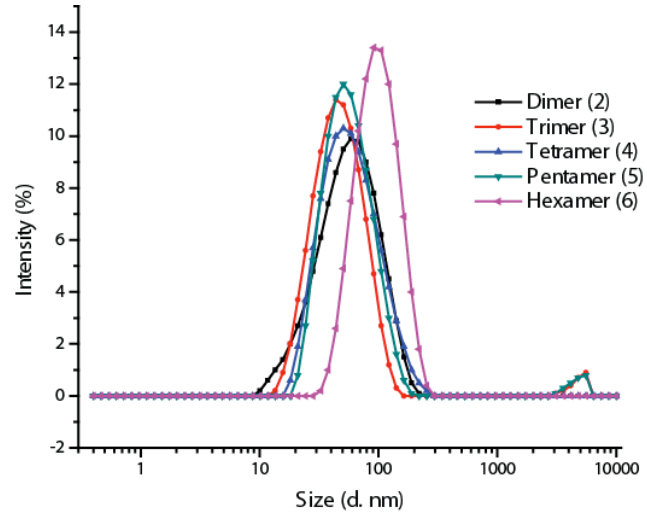


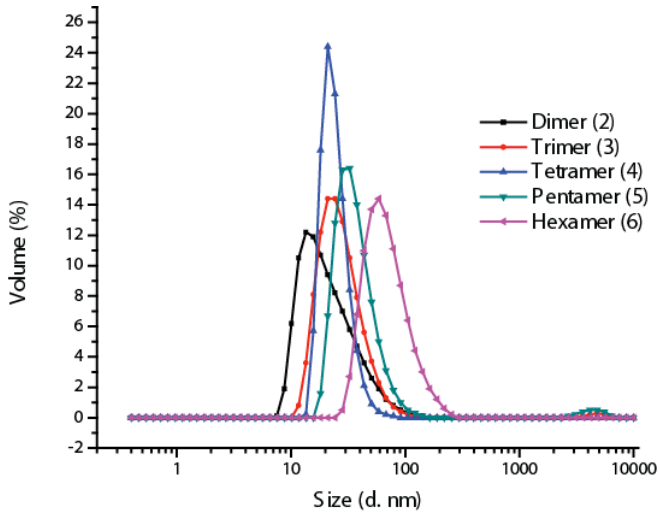
Hexamer **6**:





Assemblies size by DLS:

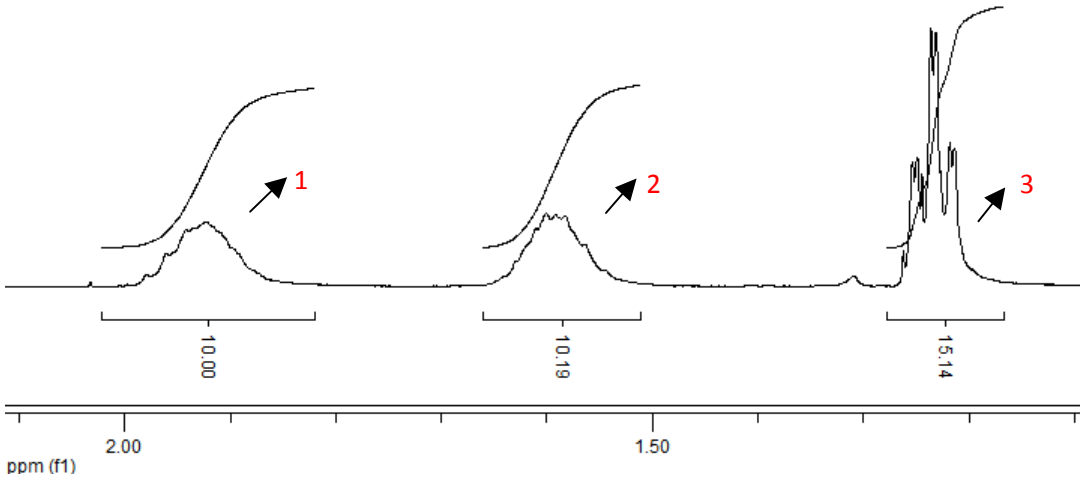




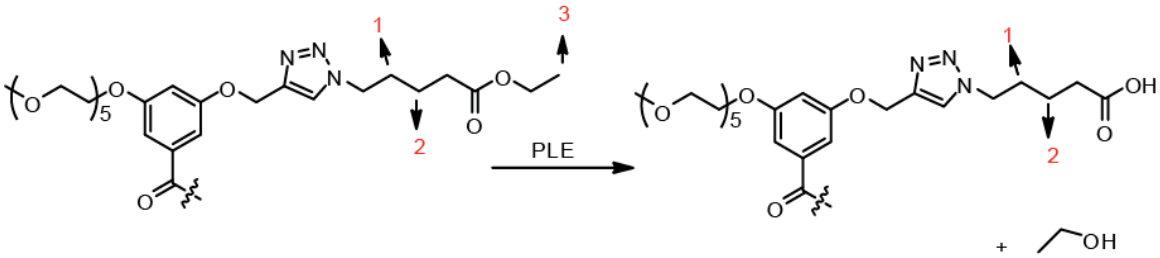
Polydispersity index (PDI) value for assemblies of these oligomers were calculated by DLS based on volume percentage to be 0.237 for dimer, 0.256 for trimer, 0.236 for tetramer, 0.235 for pentamer and 0.189 for hexamer.

Generation of the carboxylic acid functionality after esterase reaction for Pentamer **5**:

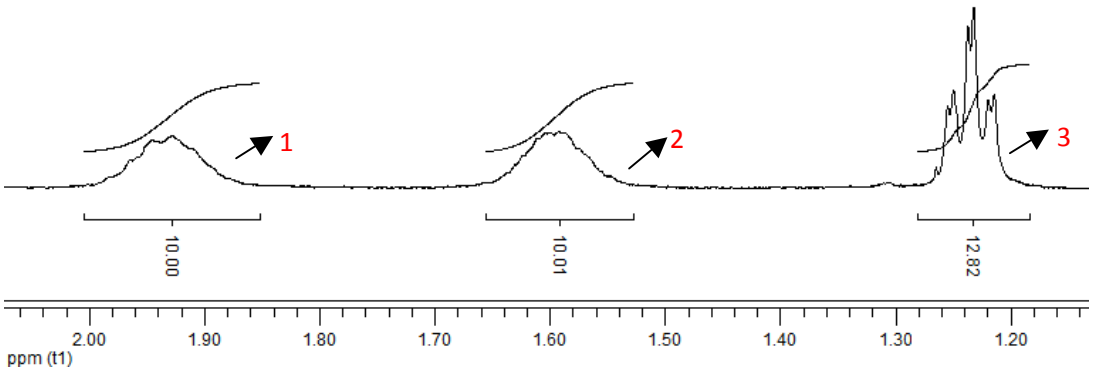
^{1}H NMR before esterase reaction



After esterase reaction, the ratio between 3^{H} and 1^{H} or 2^{H} decreased, and 15% of the ester groups in pentamer **5** are hydrolyzed at this concentration of the enzyme.

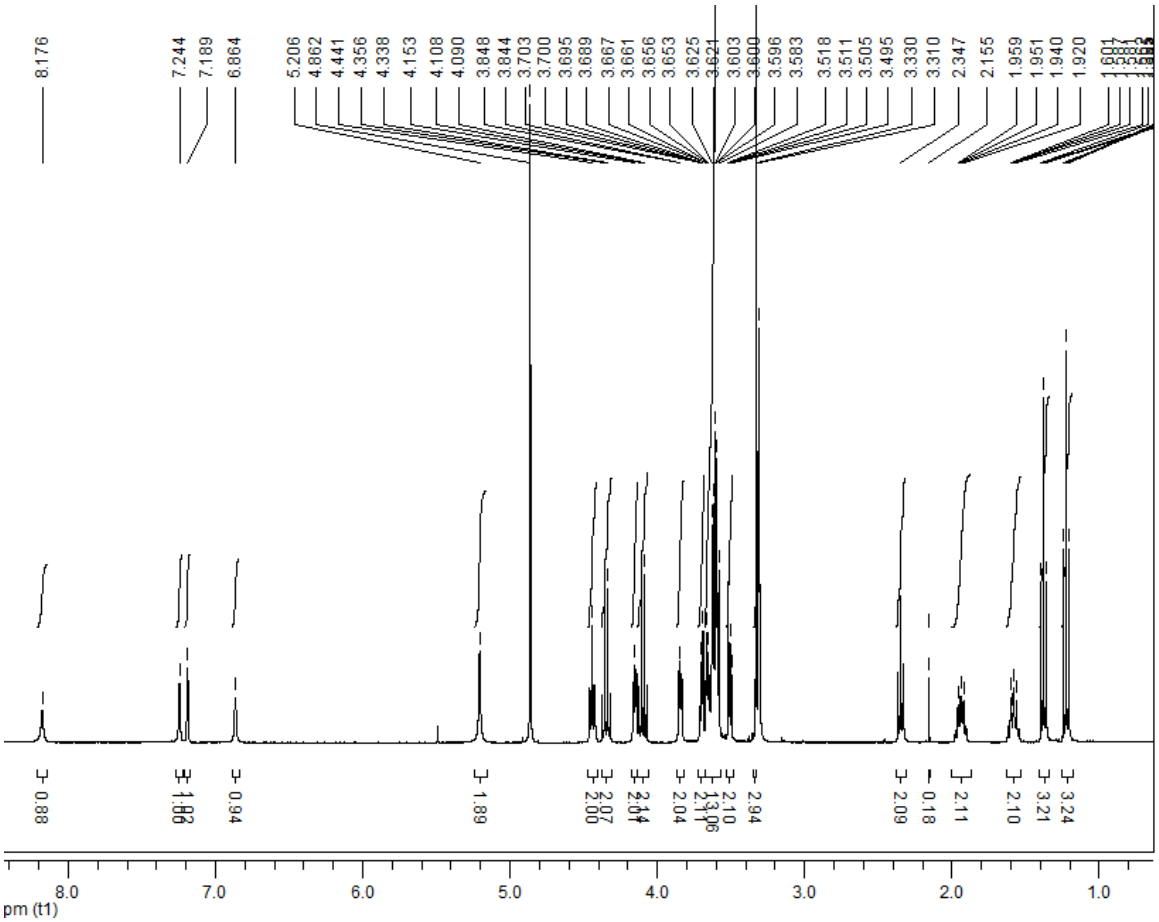


^{1}H NMR after esterase reaction

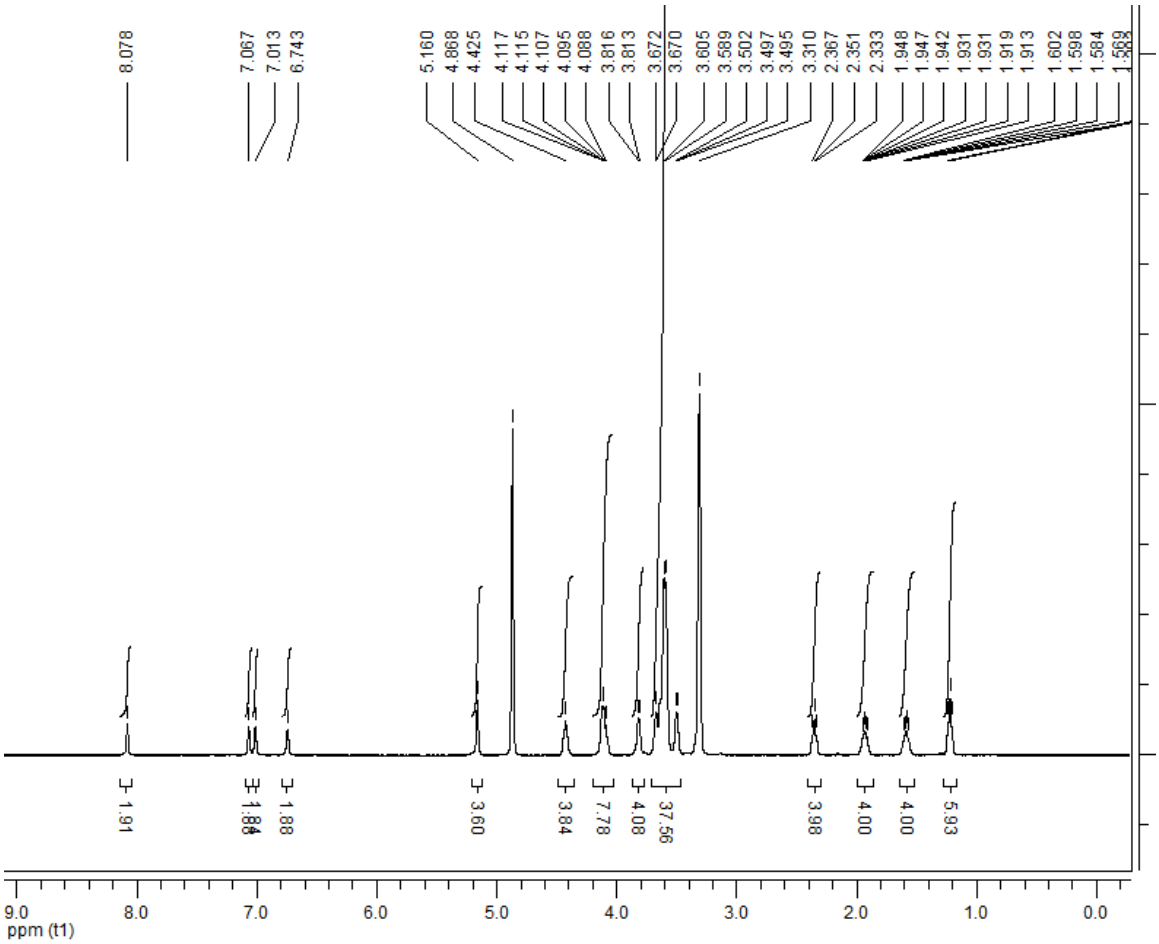


^{1}H NMR

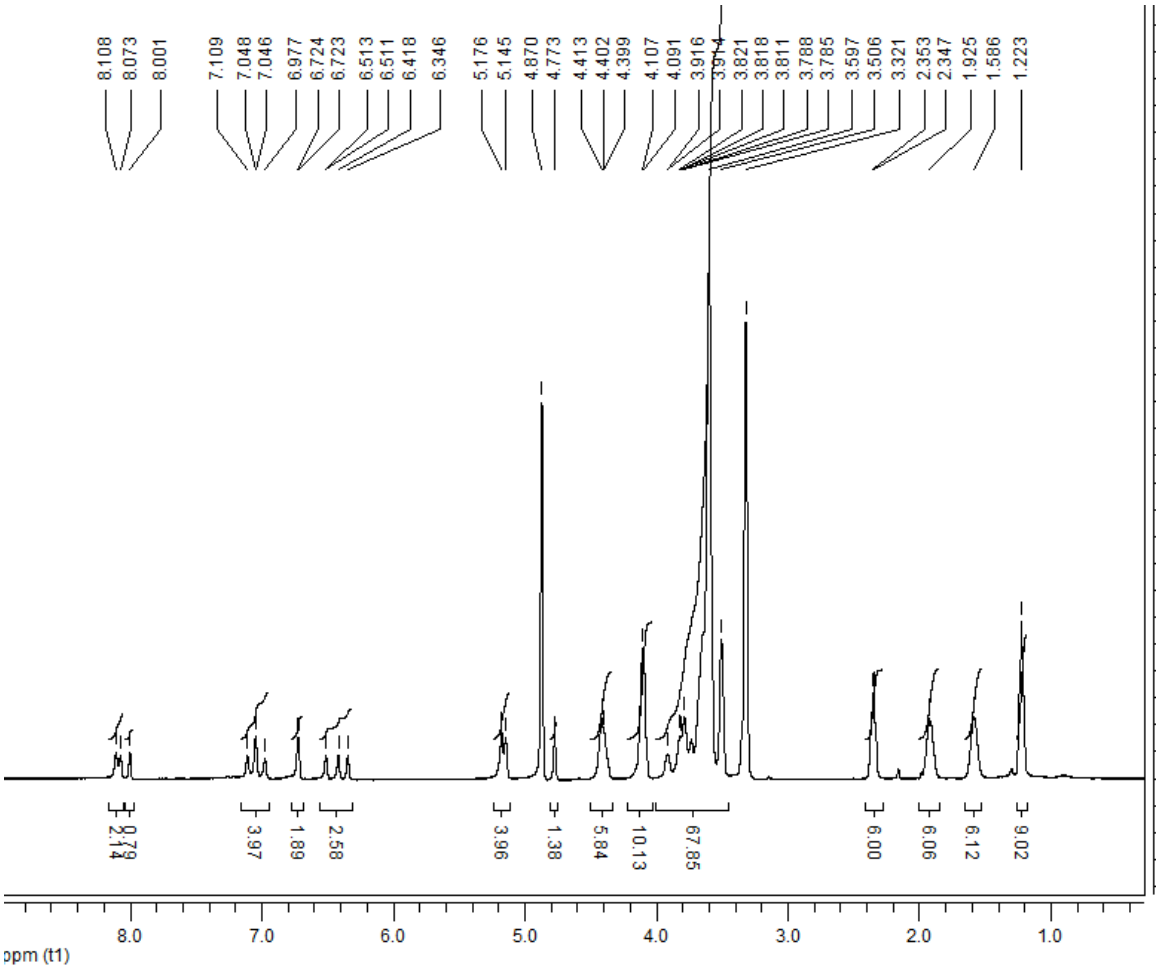
Monomer **1**:



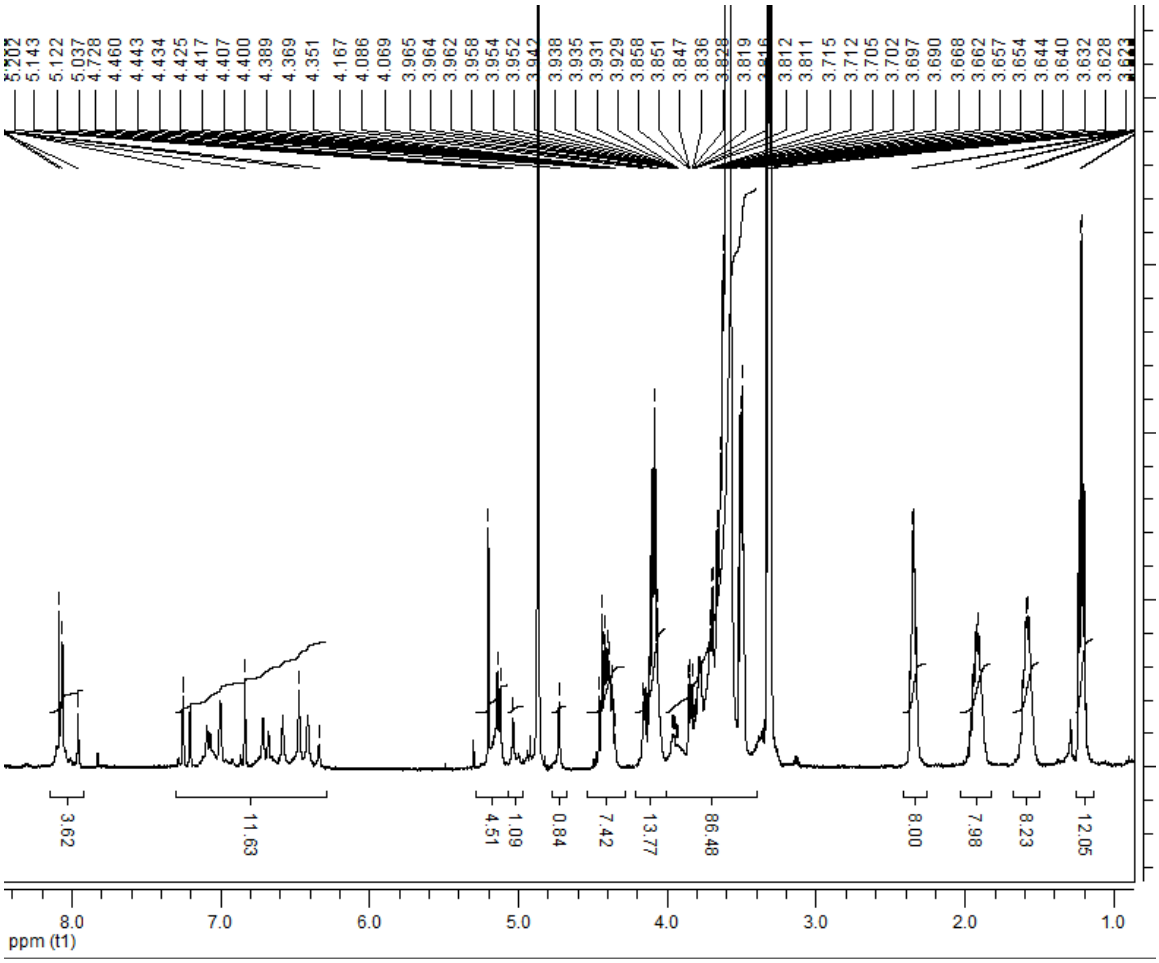
Dimer **2**:


8.078
7.067
7.013
6.743
5.160
4.868
4.425
4.117
4.115
4.107
4.095
4.088
3.816
3.813
3.672
3.670
3.605
3.589
3.502
3.497
3.495
3.310
2.367
2.351
2.333
1.948
1.947
1.942
1.931
1.931
1.919
1.913
1.602
1.598
1.584
1.569
1.91
1.84
1.88
3.90
3.84
7.78
4.08
37.56
3.98
4.00
4.00
5.93
9.0
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
ppm (t1)

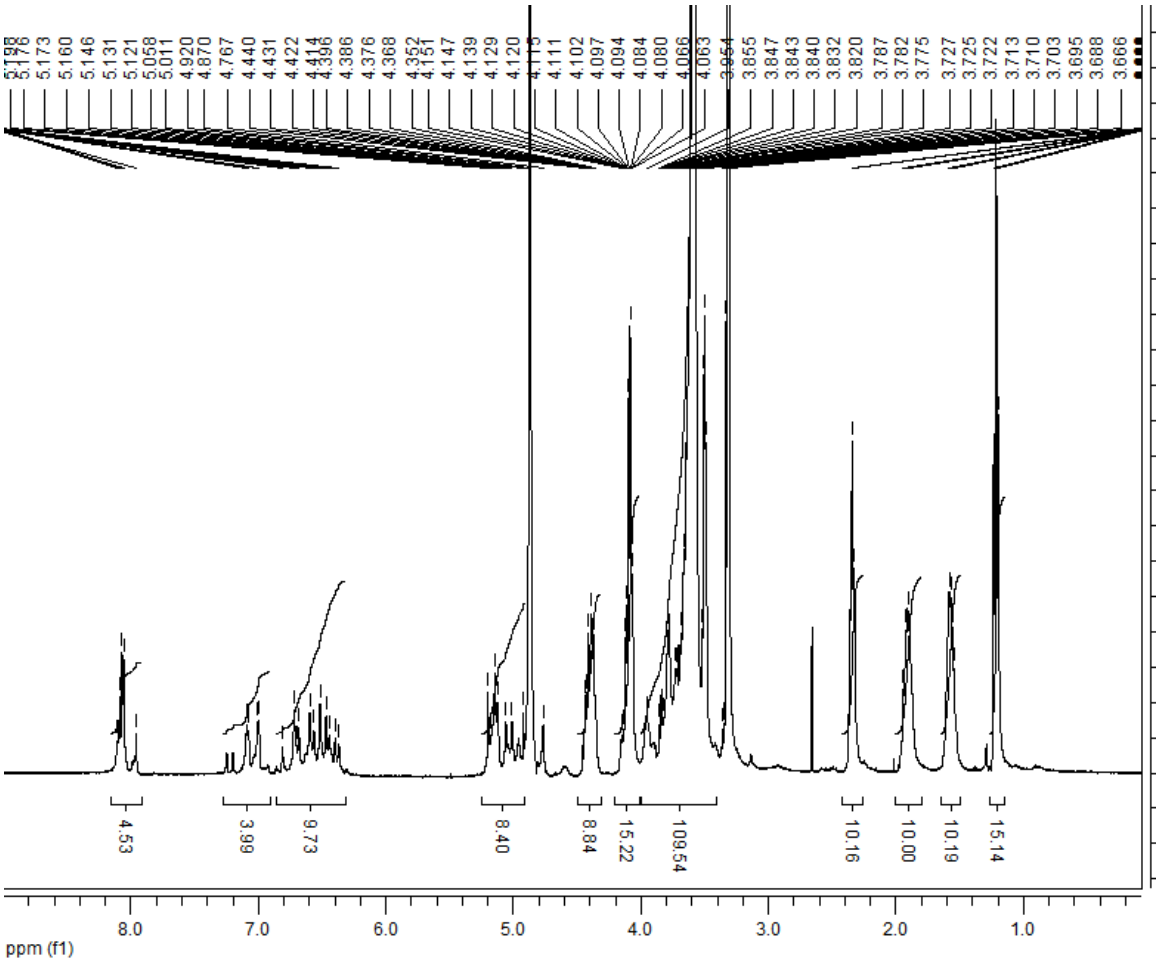
Trimer **3**:


8.108
8.073
8.001
7.109
7.048
7.046
6.977
6.724
6.723
6.513
6.511
6.418
6.346
5.176
5.145
4.870
4.773
4.413
4.402
4.399
4.107
4.091
3.916
3.914
3.821
3.818
3.811
3.788
3.785
3.597
3.506
3.321
2.353
2.347
1.925
1.586
1.223
2.14
0.79
3.97
1.89
2.58
3.96
1.38
5.84
10.13
67.85
9.00
6.06
6.12
9.02
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (t1)

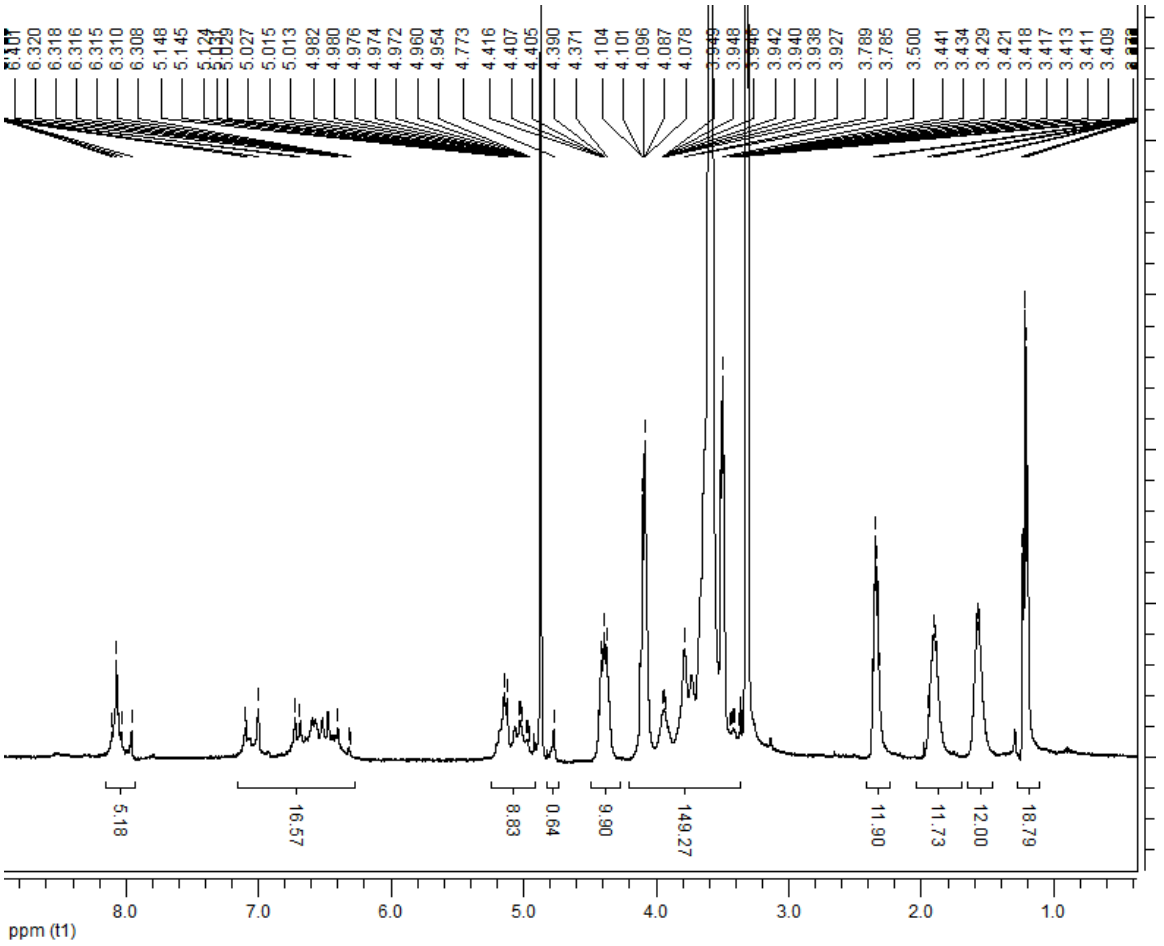
Tetramer **4**:


5.202
5.143
5.122
5.037
4.728
4.460
4.443
4.434
4.425
4.417
4.407
4.400
4.389
4.369
4.351
4.167
4.086
4.069
3.965
3.964
3.962
3.958
3.954
3.952
3.942
3.938
3.935
3.931
3.929
3.858
3.851
3.847
3.836
3.828
3.819
3.816
3.812
3.811
3.715
3.712
3.705
3.702
3.697
3.690
3.668
3.662
3.657
3.654
3.644
3.640
3.632
3.628
3.623
3.62
11.63
4.51
1.09
0.84
7.42
13.77
86.48
8.00
7.98
8.23
12.05
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (t1)

Pentamer **5**:


5.188
5.178
5.173
5.160
5.146
5.131
5.121
5.058
5.011
4.920
4.870
4.767
4.440
4.431
4.422
4.414
4.396
4.386
4.376
4.368
4.352
4.151
4.147
4.139
4.129
4.120
4.115
4.111
4.102
4.097
4.094
4.084
4.080
4.066
4.063
3.954
3.855
3.847
3.843
3.840
3.832
3.820
3.787
3.782
3.775
3.727
3.725
3.722
3.713
3.710
3.703
3.695
3.688
3.666
4.53
3.99
9.73
8.40
8.84
15.22
109.54
10.16
10.00
10.19
15.14
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (f1)

Hexamer **6**:


6.320
6.318
6.316
6.315
6.310
6.308
5.148
5.145
5.027
5.015
5.013
4.982
4.980
4.976
4.974
4.972
4.960
4.954
4.773
4.416
4.407
4.405
4.390
4.371
4.104
4.101
4.096
4.087
4.078
3.942
3.940
3.938
3.927
3.789
3.785
3.500
3.441
3.434
3.429
3.421
3.418
3.417
3.413
3.411
3.409
5.18
16.57
8.83
0.64
9.90
149.27
11.90
11.73
12.00
18.79
8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
ppm (t1)