Electronic Supplementary Material (ESI) for New Journal of Chemistry.

Supplementary Information

In Search of Bioinspired Hydrogels from Amphiphilic Peptides for sustained release of Anticancer drugs

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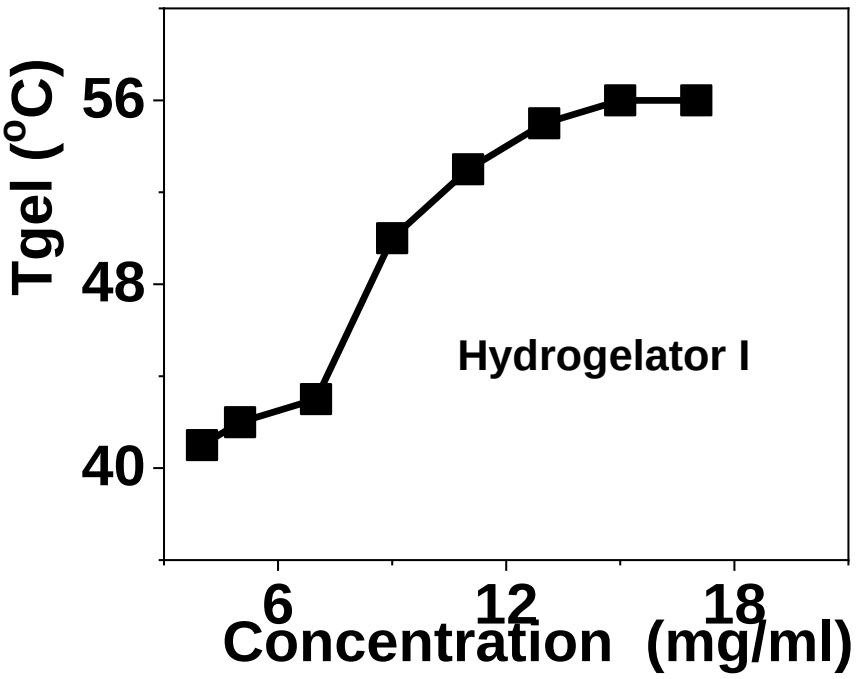

Figure S1A. Tgel graph of the Xerogel of Hydrogelator –**I**.

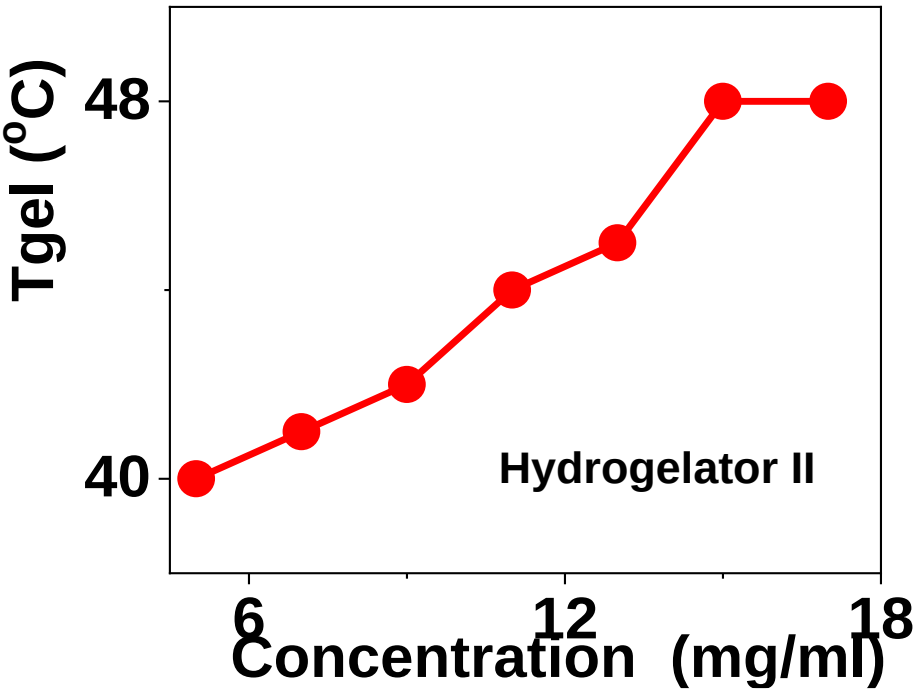

Figure S1B. Tgel graph of the Xerogel of Hydrogelator –**II**.

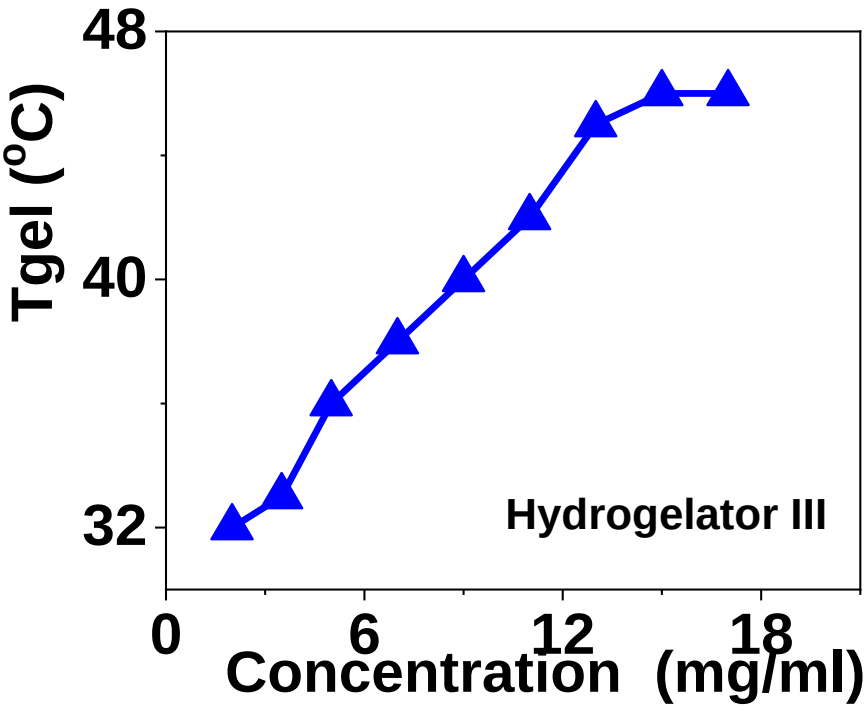

Figure S1C. Tgel graph of the Xerogel of Hydrogelator –**III**.

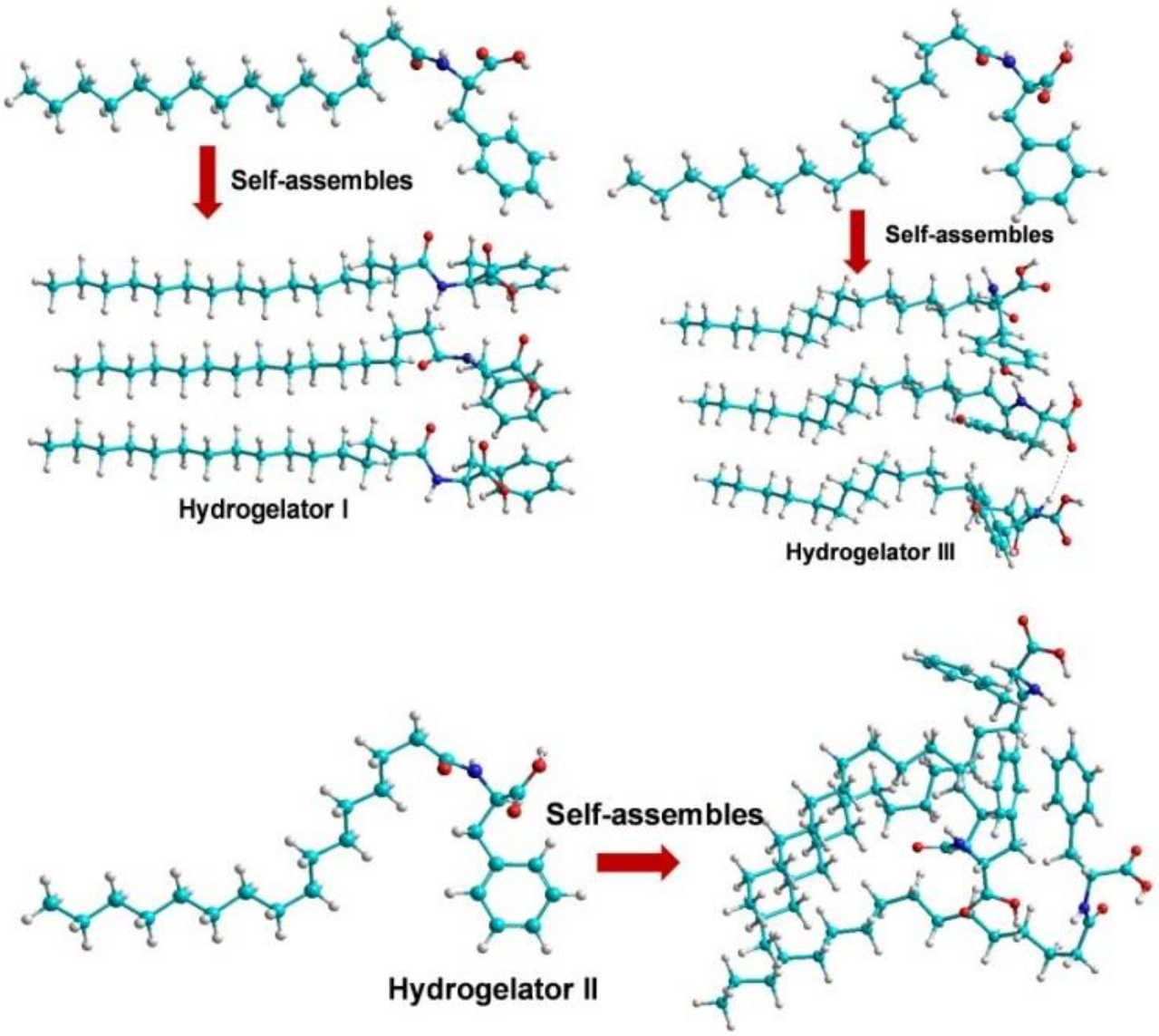

Figure S2. Energy Optimized Structure of Hydrogel **I - III**.

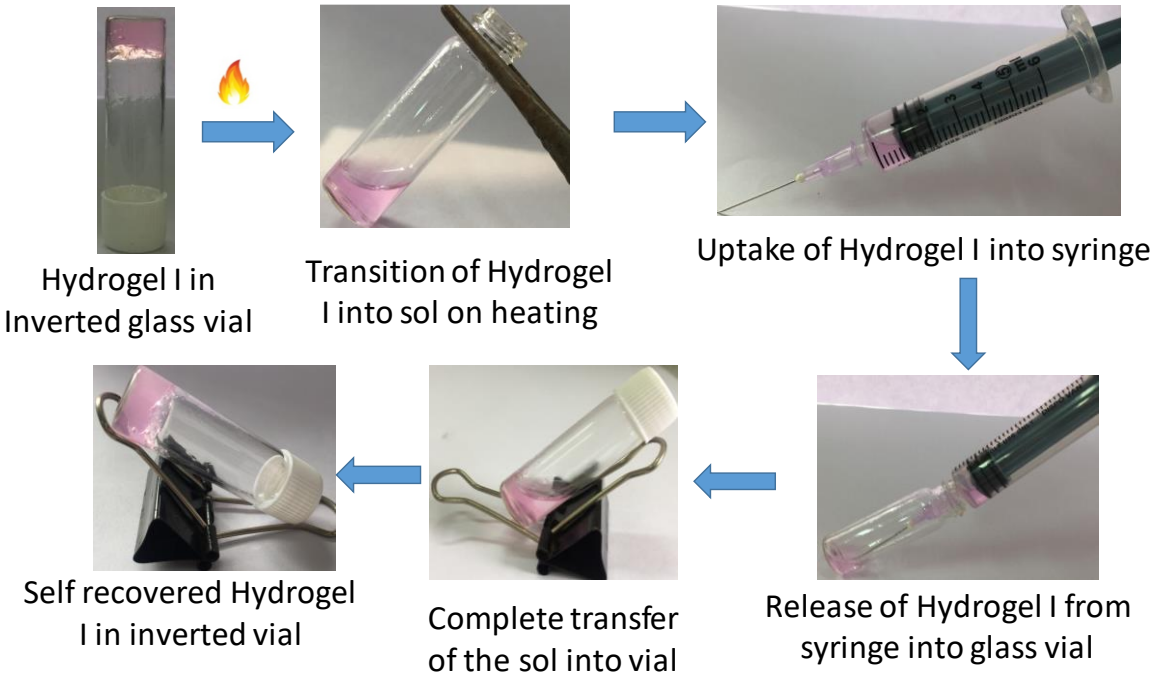

Figure S3. Demonstration of Thermoreversible and Injectable property of Hydrogelator- **I**.

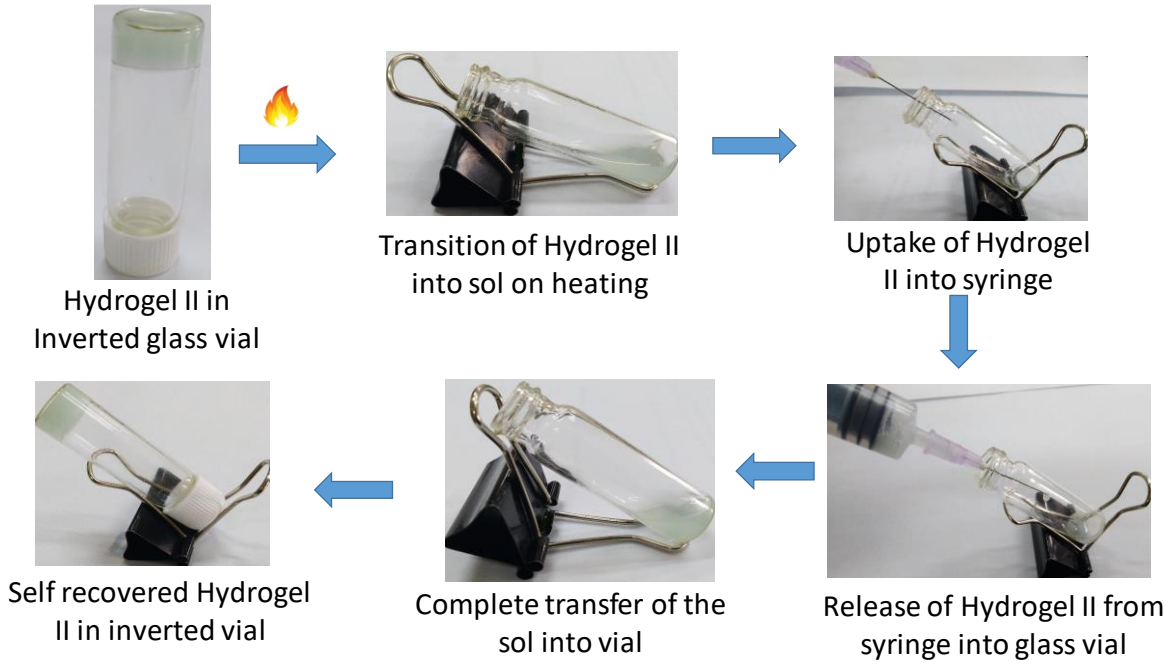

Figure S4. Demonstration of Thermoreversible and Injectable property of Hydrogelator- **II**.

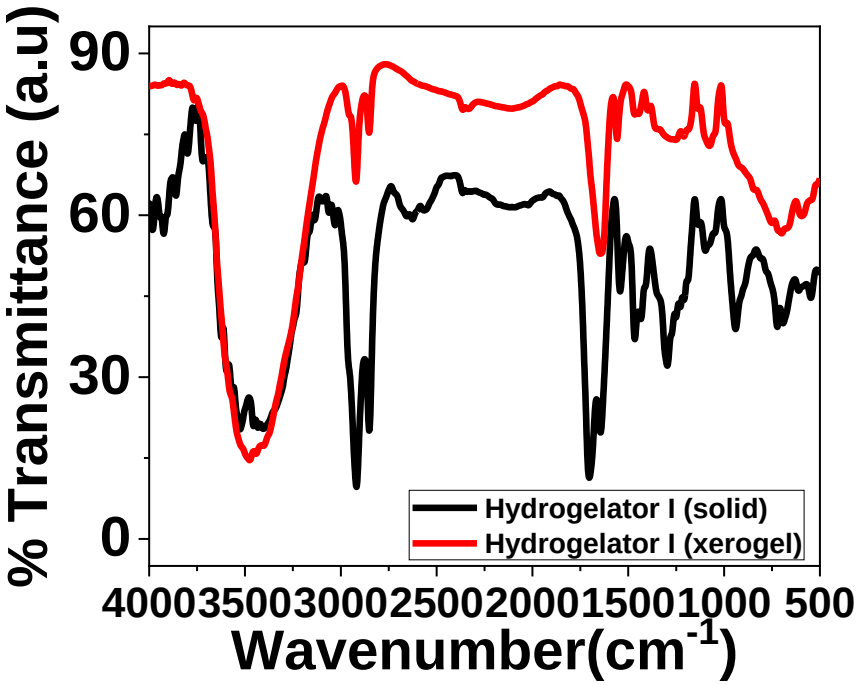

Figure S5. Overlapped FT-IR Spectra of Hydrogelator –**I**

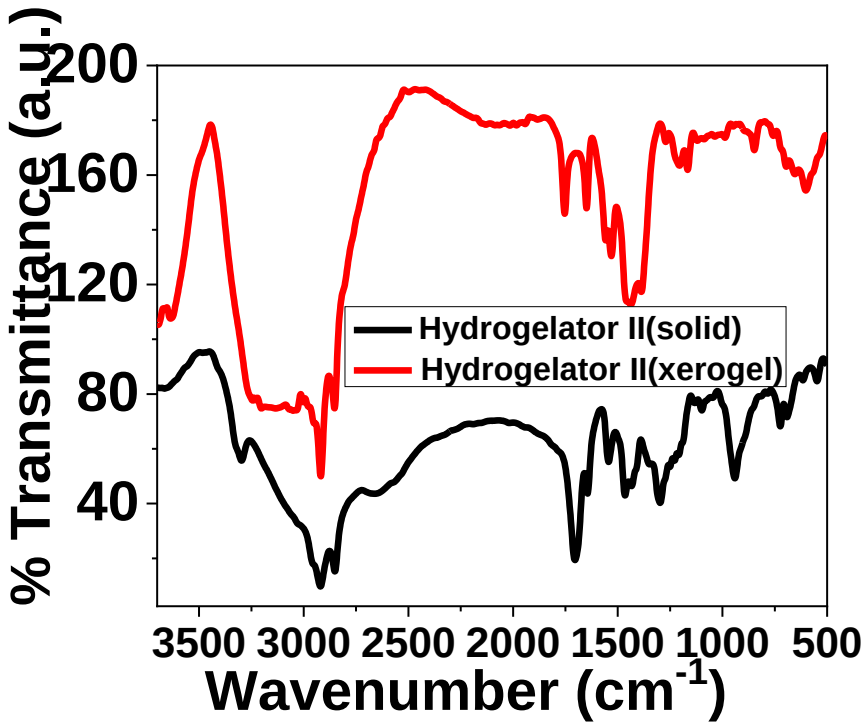

Figure S6. Overlapped FT-IR Spectra of Hydrogelator –**II**

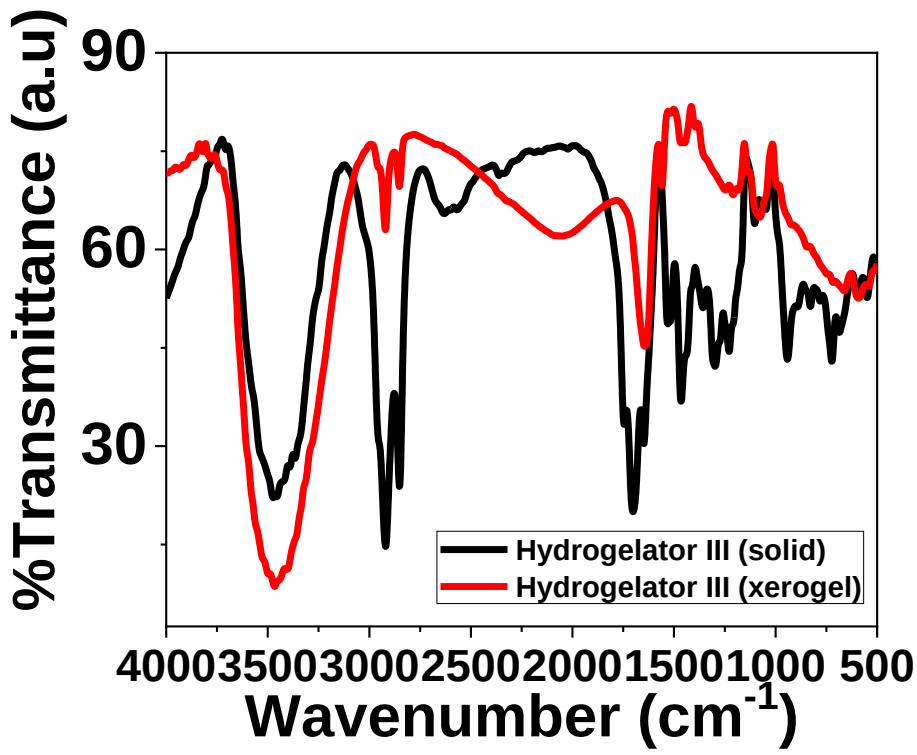

Figure S7. Overlapped FT-IR Spectra of Hydrogelator –**III**

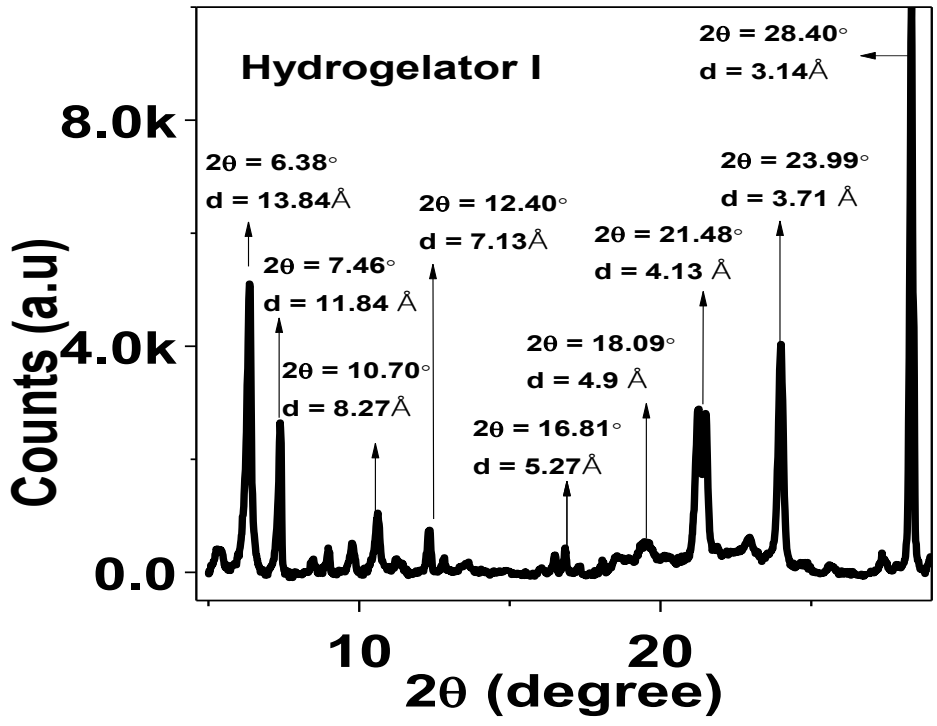

Figure S8. PXRD Spectrum For Hydrogelator I

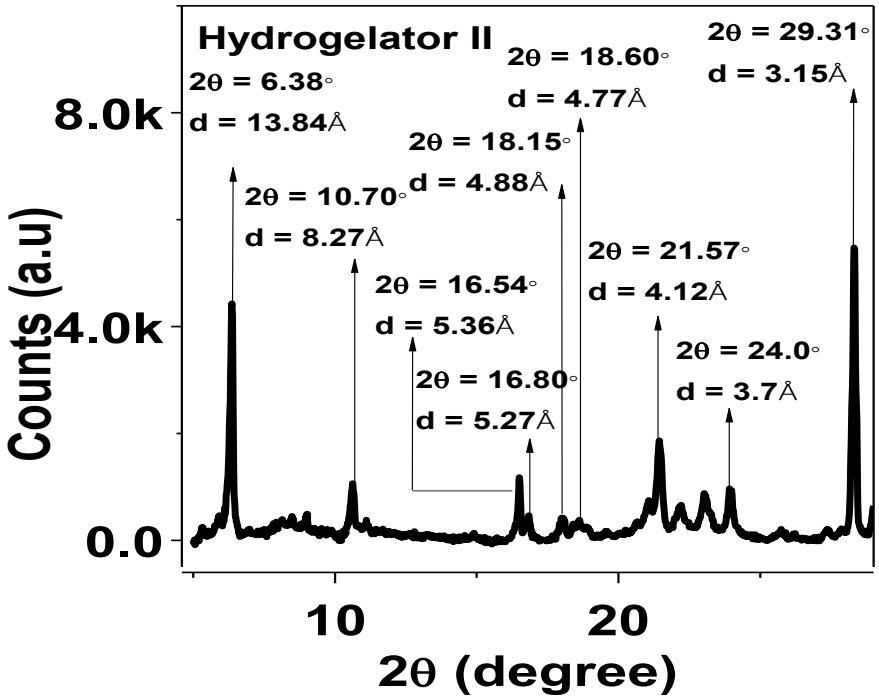

Figure S9. PXRD Spectrum For Hydrogelator II

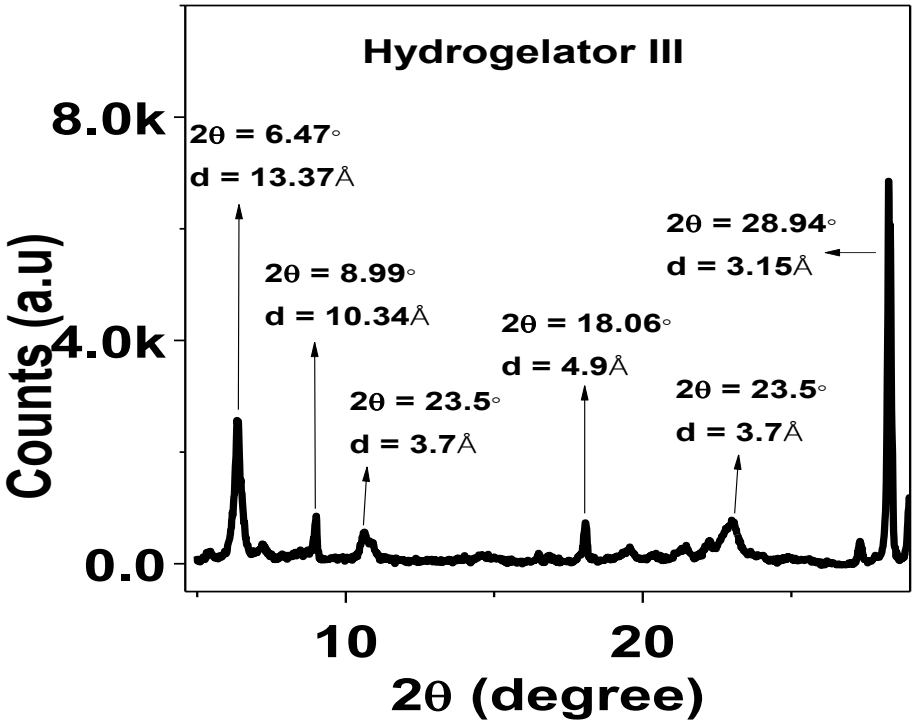

Figure S10. PXRD Spectrum For Hydrogelator III

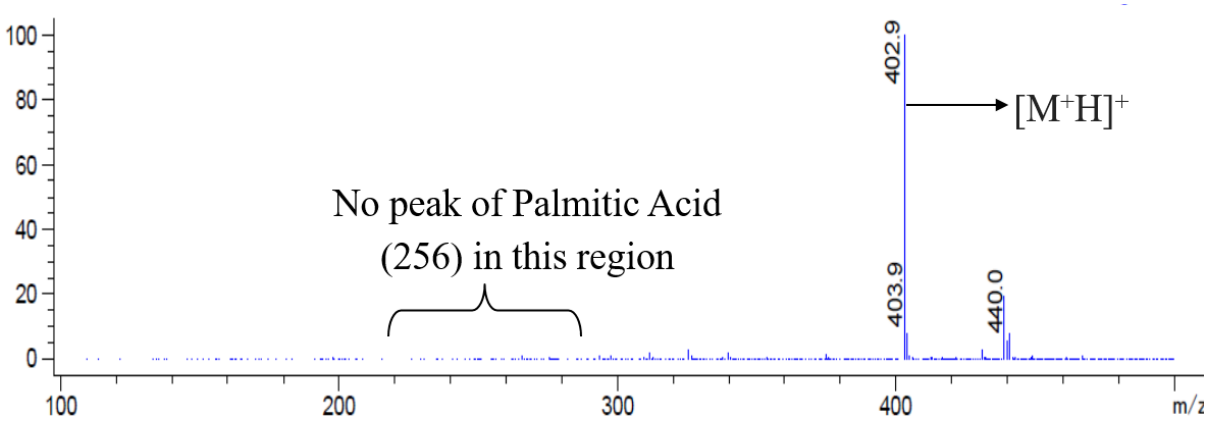

Figure S11: Mass spectrum of Hydrogelator - **I** before incubation with Proteinase K.

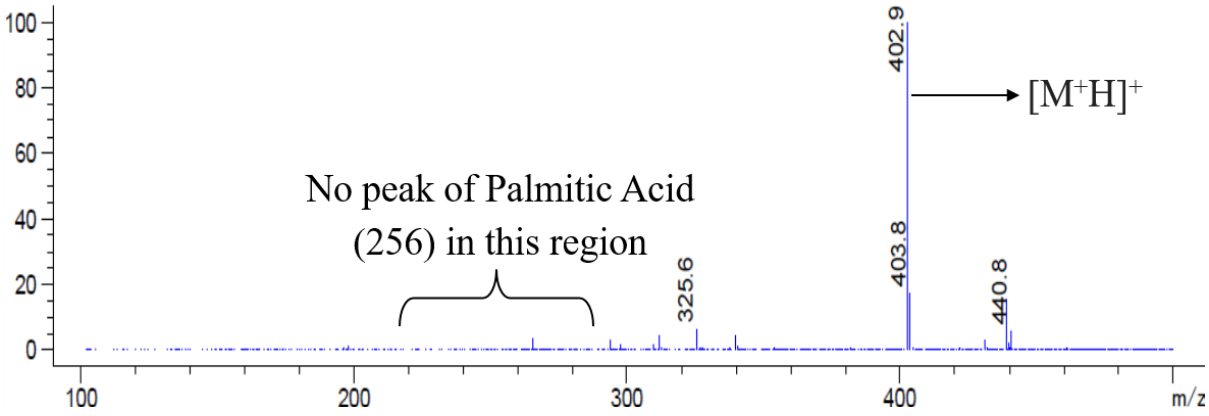

Figure S12: Mass spectrum of Hydrogelator -**I** after 24 hours incubation with proteinase K.

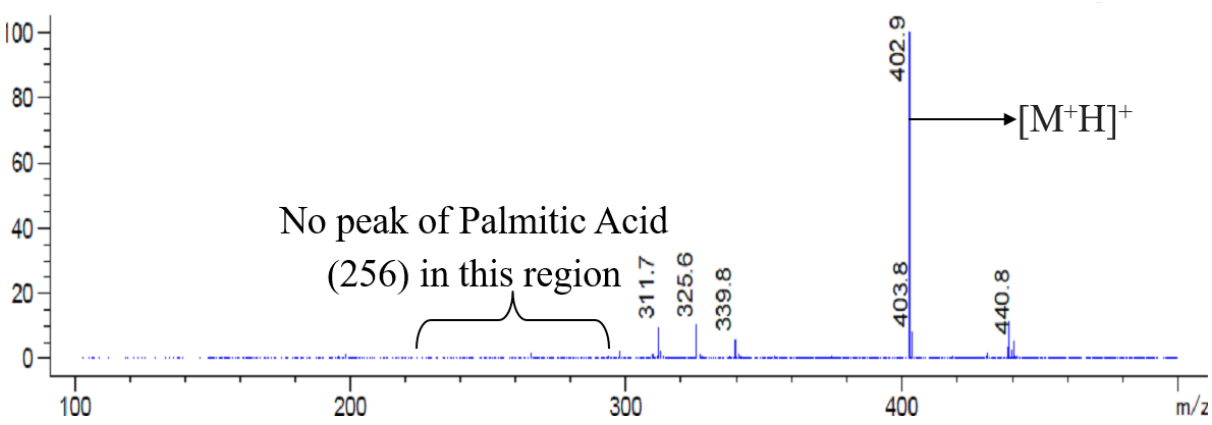

Figure S13: Mass spectrum of Hydrogelator -**I** after 48hours of incubation with proteinase K.

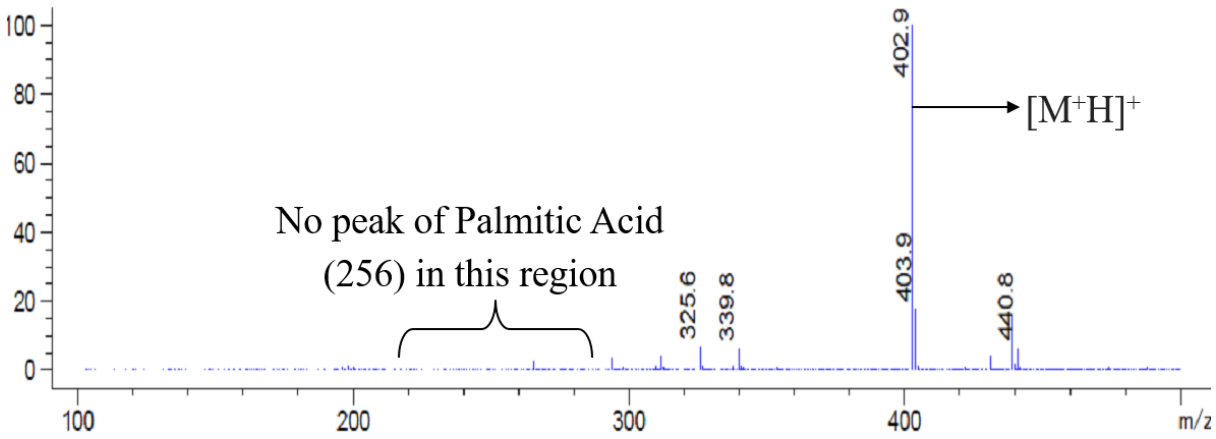

Figure S14: Mass spectrum of Hydrogelator -**II** before incubation with proteinase K.

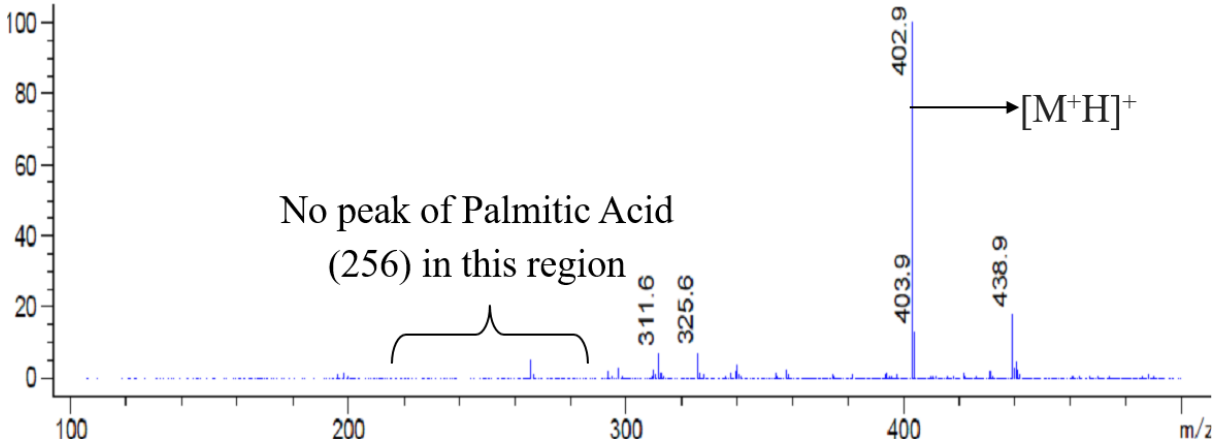

Figure S15: Mass spectrum of Hydrogelator -**II** after 24 hours incubation with proteinase K.

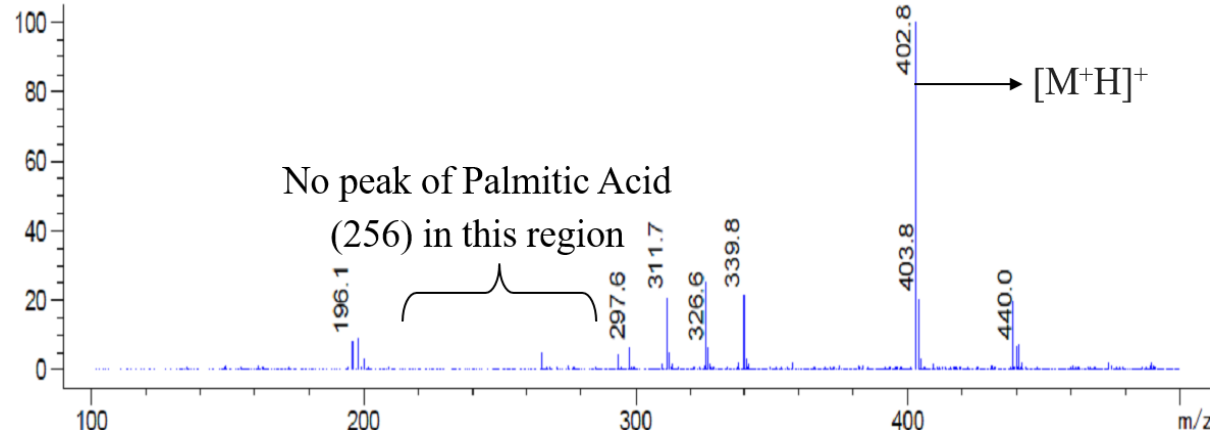

Figure S16: Mass spectrum of Hydrogelator -**II** after 48 hours of incubation with proteinase K.

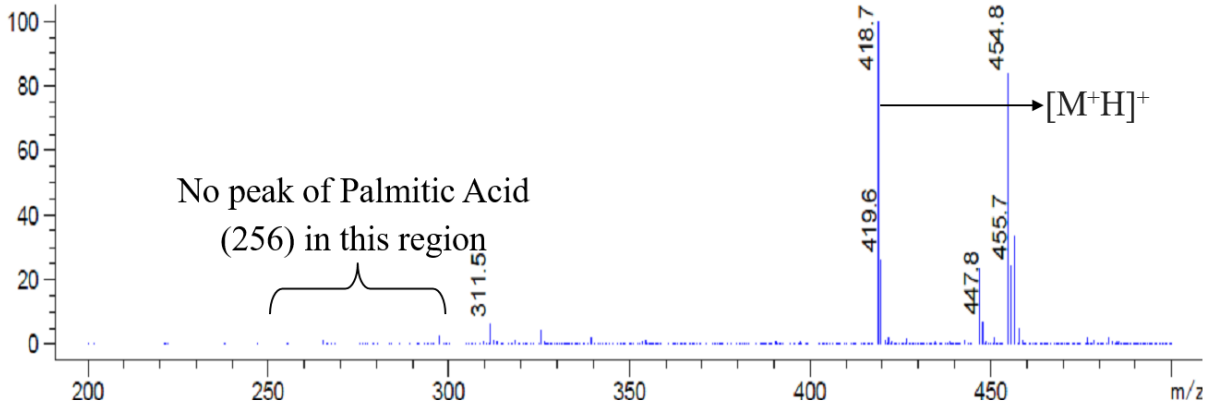

Figure S17: Mass spectrum of Hydrogelator -**III** before incubation with proteinase K.

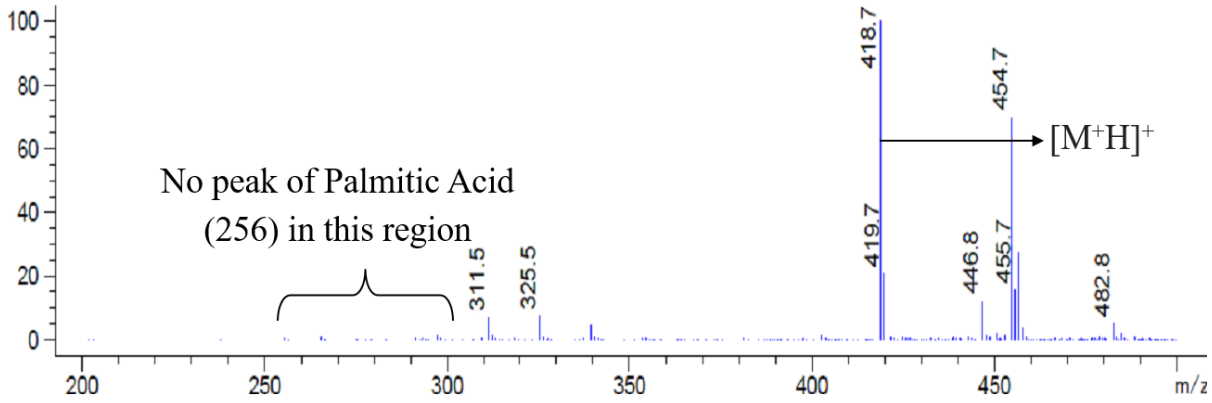

Figure S18: Mass spectrum of Hydrogelator -**III** after 24hours incubation with proteinase K.

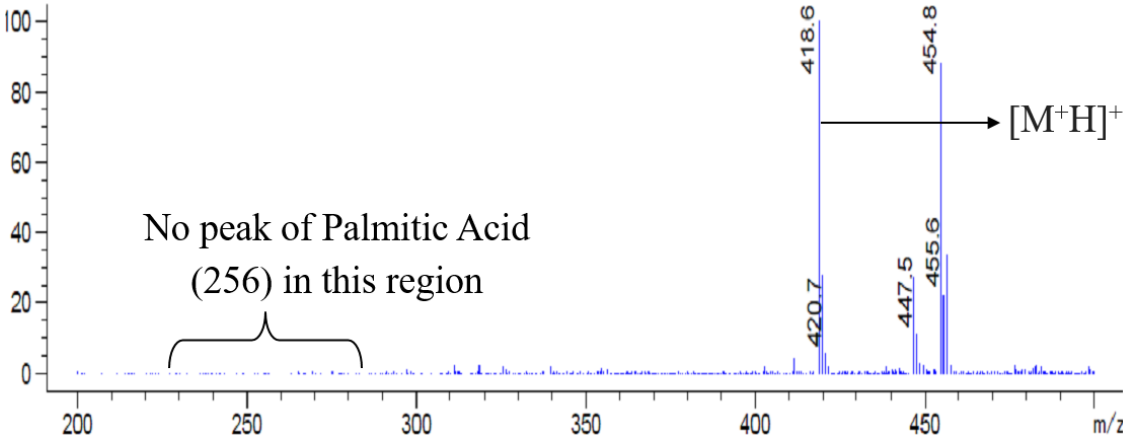

Figure S19: Mass spectrum of Hydrogelator -**III** after 48hours of incubation with proteinase K.

Experimental Procedure

DSC analysis

To gain insight into the thermodynamic parameters, the gels obtained from Hydrogelator **I - III** were subjected to DSC analysis in triplicates using a calorimeter DSC, from Perkin Elmer. For this experiments each cycle was programmed from -30 to 70 °C for heating and vice-versa for cooling at a rate of 5°C/min. For the concentration dependant measurements the same parameters were used.

Rheology

Rheological measurements were carried out on a Rheoplus MCR302 (Anton paar) rotational rheometer with parallel plate geometry and obtained data were processed with start rheometer software. For the oscillatory shear measurements, parallel top plate with a 25 mm diameter and 1.0 mm gap distance were used. Gels (6 mg/ml) for rheological experiments were prepared on the bottom plate of the rheometer. For temperature dependant experiments, the heating was carried out from room temperature(25°C) to 70 °C, with heating rates of 2°C/min and angular frequency of 1 rad/sec.

Field Emission scanning electron microscopic study (FESEM)

Morphology of the xerogel obtained from auxin-amino acid conjugate was investigated using FESEM microscope (JEOL JSM - 6700F) and were gold coated.

MTT Assay

The cells were seeded at a density of approximately 5×10 3 cells/well in a 96-well flat-bottom micro plate and maintained at 37 0 C in 95% humidity and 5% CO2 for overnight. Different concentration (600, 300, 150, 75, 37.5, 18.75 µm) of samples were treated.. The cells were incubated for another 48 hours. The cells in well were washed twice with phosphate buffer solution, and 20 µL of the MTT staining solution (5mg/ml in phosphate buffer solution) was added to each well and plate was incubated at 37 0 C.After 4h, 100 µL of di- methyl sulfoxide (DMSO)

was added to each well to dissolve the formazan crystals, and absorbance was recorded with a 570 nm using micro plate reader (1, 2).
Formula :
Surviving cells (%) = Mean OD of test compound /Mean OD of Negative control ×100
Inhibiting cells (%) =100- Surviving cells
Using graph Pad Prism Version5.1, we calculate the IC 50 of compounds

Nanoparticle characterization

Nanoparticle size diameter and surface charge were measured using Malvern Zeta-sizer, with 4mW 633 He-Ne Laser, (DTS version 4.10, Malvern, U. K.) with appropriate viscosity and refractive index settings. The temperature was maintained at 25^{o}C during the measurement.

In vitro drug release of drugs from HNPs

The in vitro release profile of the model drugs 5FU and doxorubicin from the drug loaded HNPs were performed using dialysis membrane previously soaked for 24 hrs in the dissolution membrane and stretched around at one end of the tube. The drug loaded formulations were carried out using pretreated membrane which were immersed into 30 mL of phosphate buffer solution of pH 7.4 for both the drugs at room temperature and magnetically stirred at 50 rpm. At selected time intervals aliquots were withdrawn from release medium and replaced with same amount of phosphate buffer (1 ml). The samples were analyzed thrice using UV-spectrophotometer at 267nm for 5FU and 495nm for doxorubicin. The percentage of cumulative drug release was plotted against time to obtain the release curves.

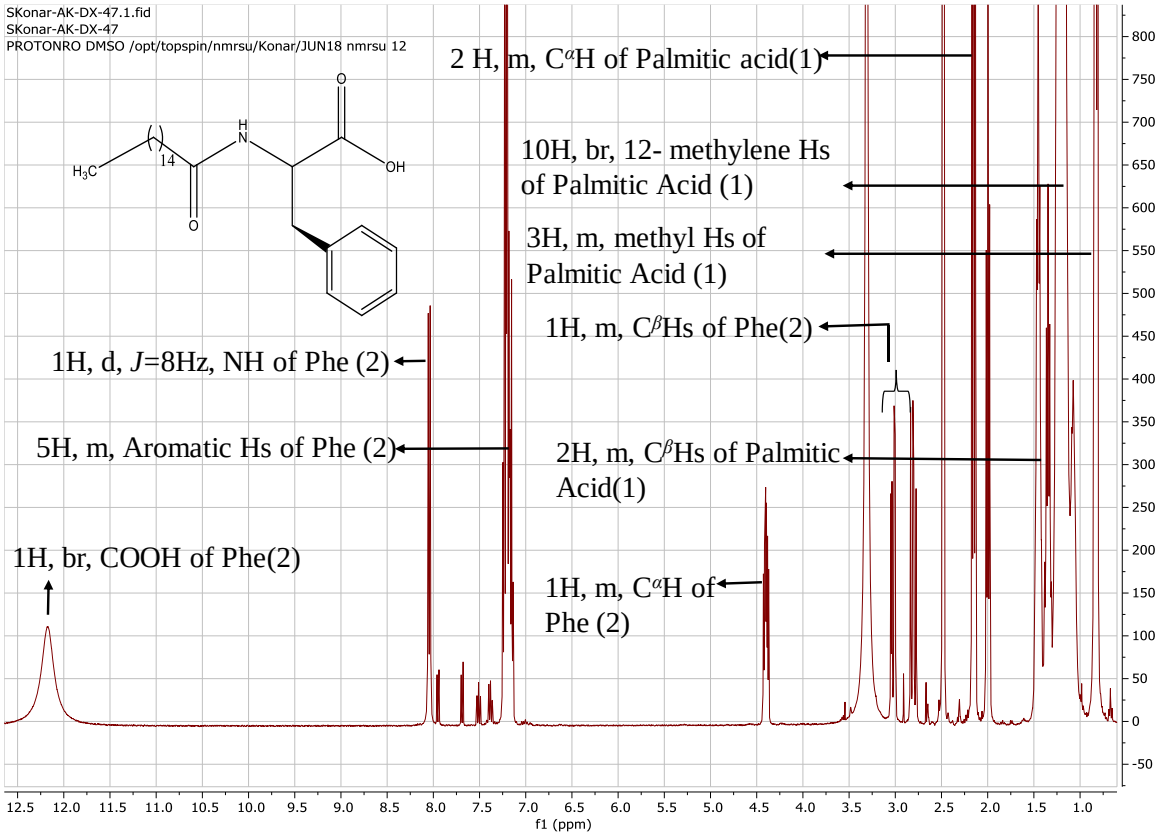

Figure S20A.^{1}H NMR Spectrum of Hydrogelator-**I**.

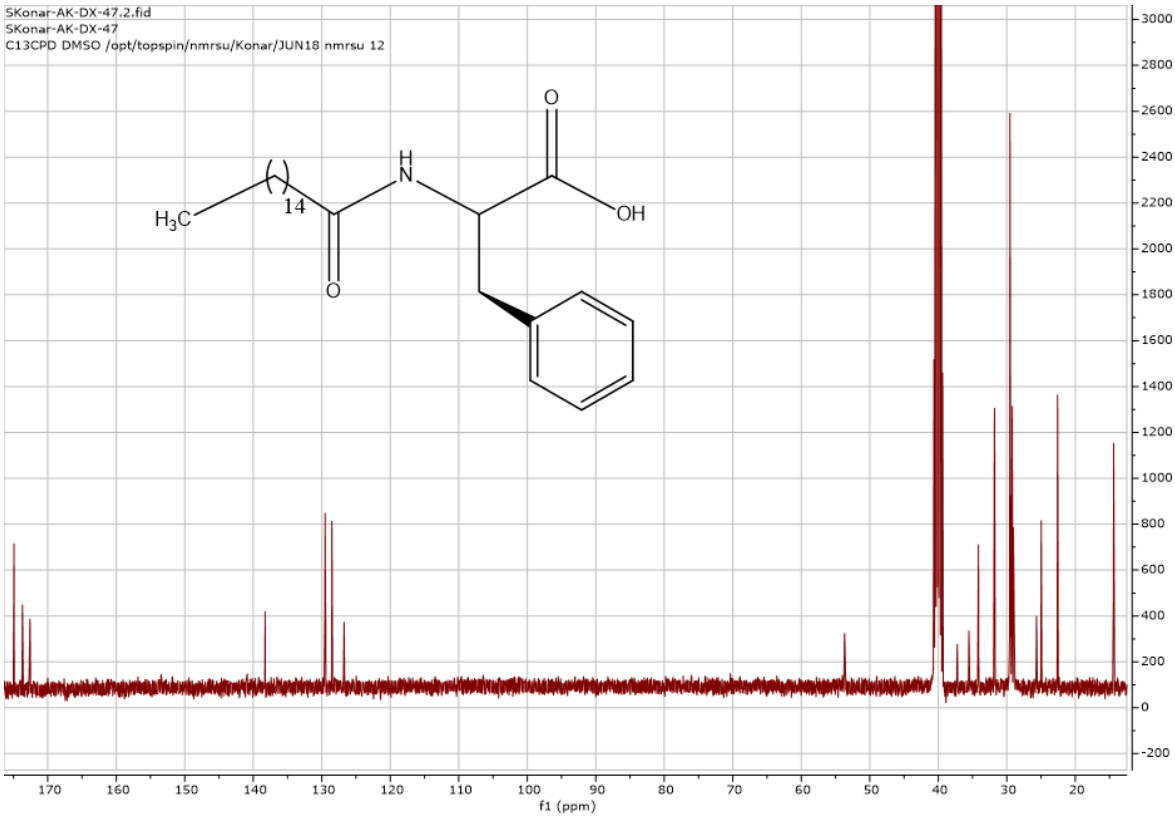

Figure S20B.^{13}C NMR Spectrum of Hydrogelator-**I**.

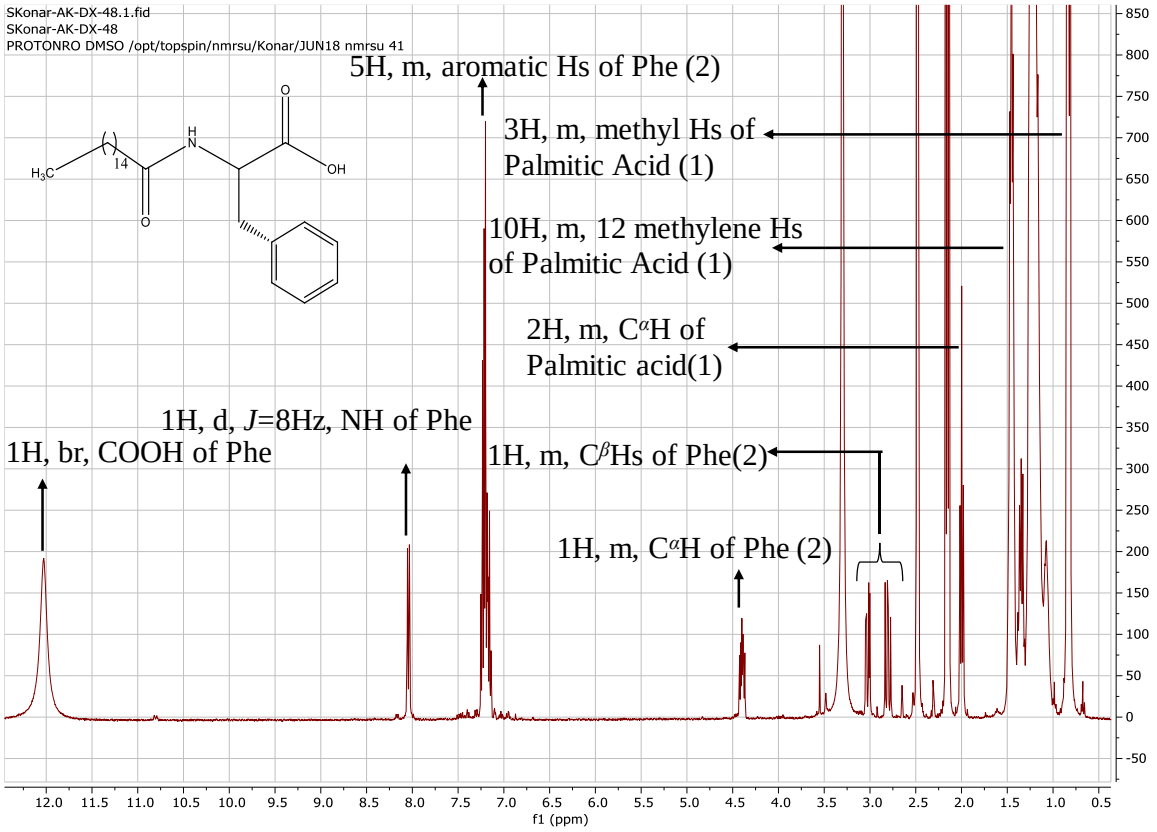

Figure S21A.^{1}H NMR Spectrum of Hydrogelator-**II**.

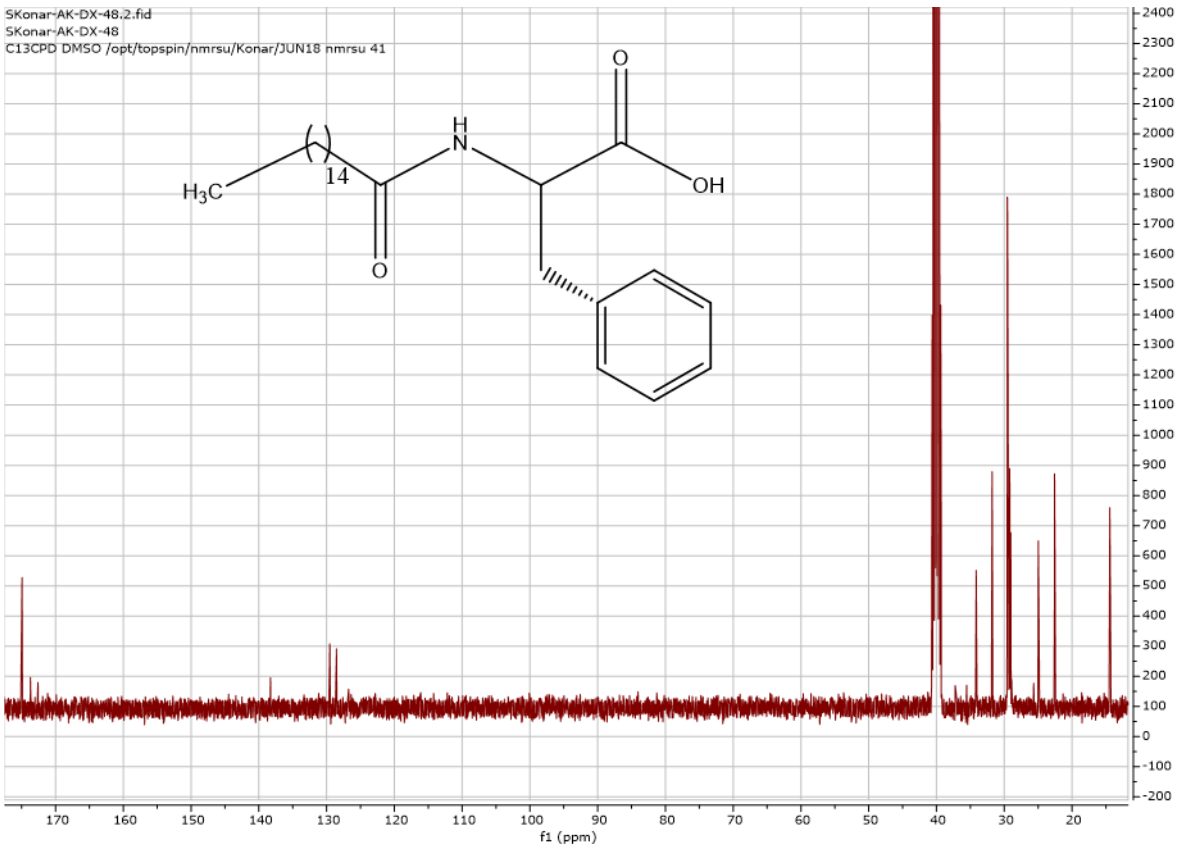

Figure S21B.^{13}C NMR Spectrum of Hydrogelator-**II**.

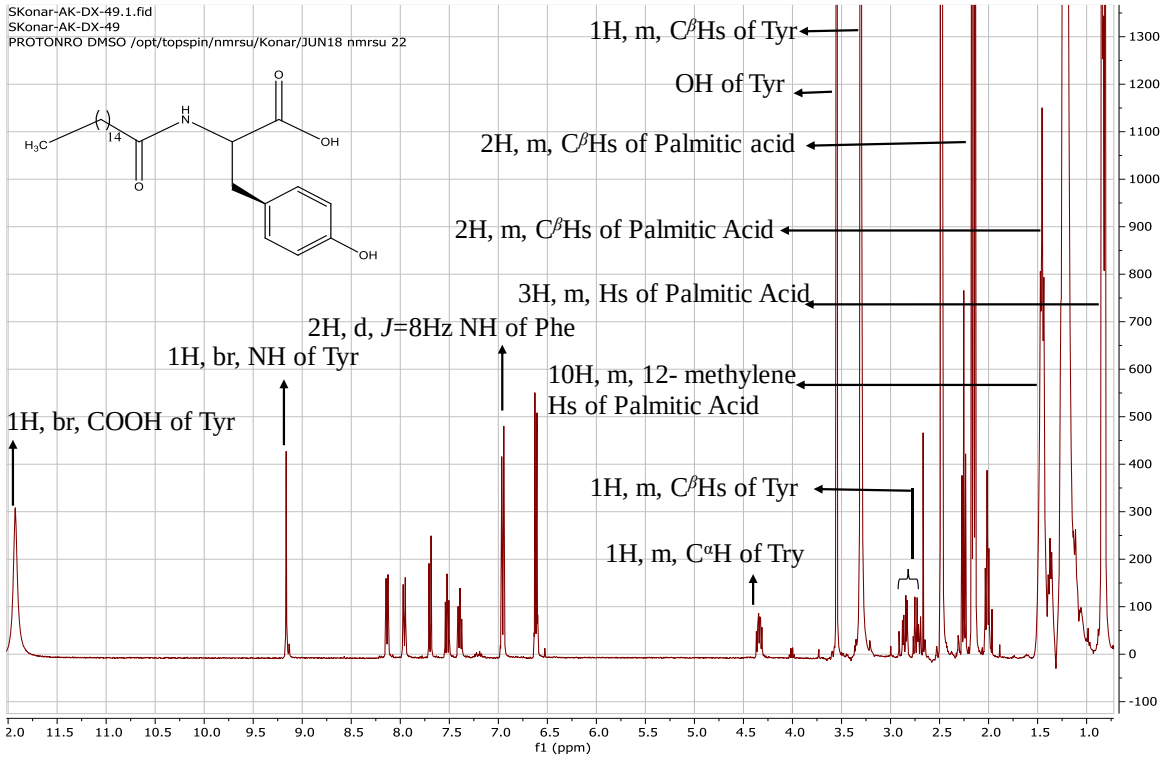

Figure S22A.[1]H NMR Spectrum of Hydrogelator-**III**.

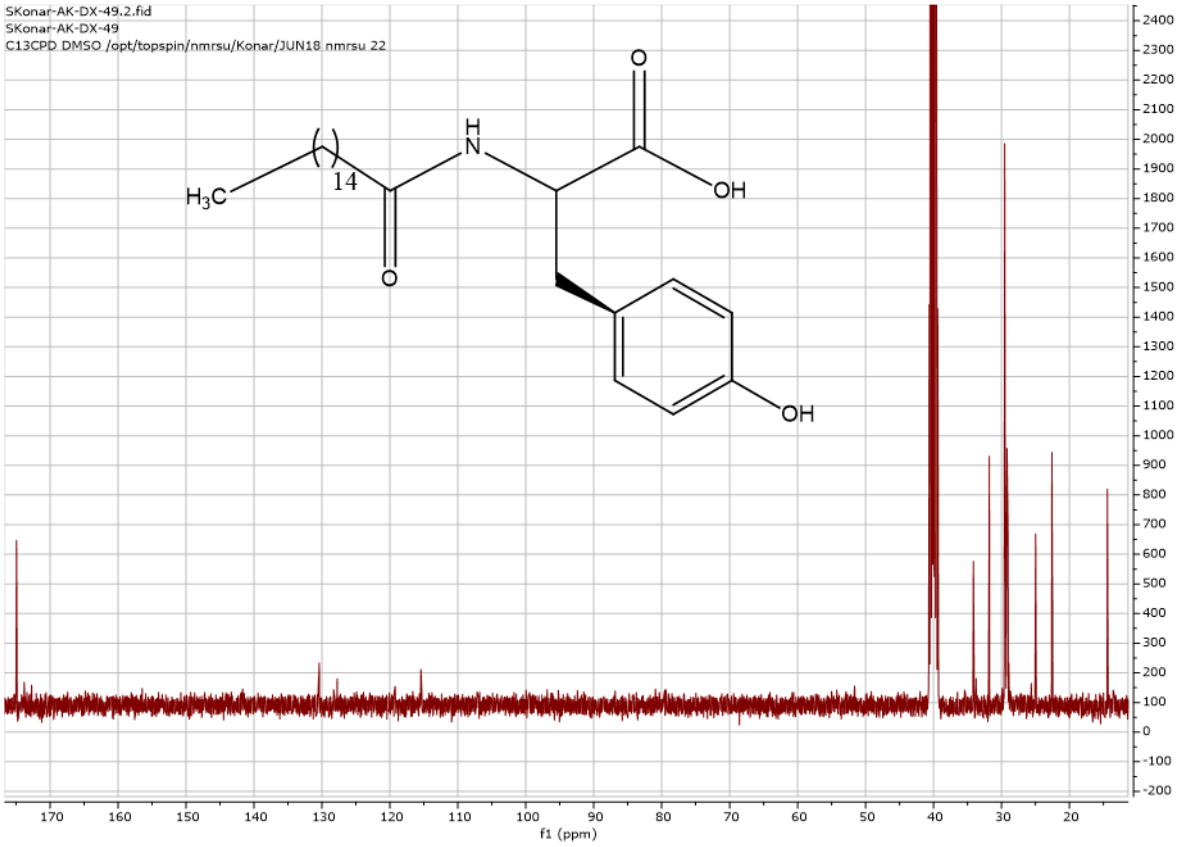

Figure S22B.[13]C NMR Spectrum of Hydrogelator-**III**.