

# The first pamidronate containing polymer and copolymer

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## Supporting Information

**Materials.** Pamidronate disodium was bought from Duqiao chemicals of Linhai (China). All the other starting materials were obtained from Aldrich or Acros. Commercially available reagents were used without further purification unless stated otherwise. Water was purified by Barnstead/Thermolyne Nanopure Diamond Life Science (UV/UF) ultrapure water system.

**General methods.** The synthesized compounds were characterized using  $^1\text{H}$  NMR (Bruker ARX 300) with  $\text{D}_2\text{O}$  as the solvent. The molecular weight of the polymers were determined by gel permeation chromatography (GPC) using 0.1 M  $\text{NaNO}_3$  as the eluent and poly(acrylic acid) as the standard. The samples were cyro-dried for scanning electron microscopy (SEM) and X-ray powder diffraction (XRD) analysis. SEM was recorded on JEOL-6300F with associated energy dispersive spectroscopy (EDS). XRD was performed on powder X-ray diffractometer\_Philips (PW1830).

### Syntheses and characterizations.

N-acryl pamidronate (**3**): N-acryloxysuccinimide (**1**) (845 mg, 5 mmol) and pamidronate disodium salt (**2**) (1.40 g, 5 mmol) were dissolved in water (pH = 8.0) together and stirred for reaction. After 24 h reaction at room temperature, the crude product was precipitated from water upon the addition of absolute ethanol. The precipitate was collected by filtration and washed with ethanol to give a 95% yield of the desired monomer **3** as a white solid.  $^1\text{H}$  NMR (300 MHz,  $\text{D}_2\text{O}$ )  $\delta$  2.11-2.25(m, 2H), 3.56-3.61(m, 2H), 5.72-5.76(m, 1H), 6.14-6.31(m, 2H) ppm.  $^{31}\text{P}$  NMR (300 MHz,  $\text{D}_2\text{O}$ )  $\delta$  17.70(s) ppm. MS: calc.  $\text{M}^+ = 333.08$ , obsvd.  $[\text{M}+\text{Na}]^+ = 356.3$ .

### Poly(N-acryl pamidronate) (**4**):

Procedure I: In a two-neck round-bottom flask equipped with a reflux condenser and a nitrogen inlet, 2 ml water solution of **3** (333 mg, 1 mmol) was introduced. After bubbling for 10 min. with nitrogen, the flask was then transferred to a water bath at 60 °C and stirred with a continuous bubbling of nitrogen. The initiator potassium persulfate (KPS) (13.5 mg, 5.0 mol% of total monomers) was added when the temperature of water solution was up to 60 °C. Thereafter, the temperature of water bath was increased to 70 °C. After 24 h polymerization, the polymer was precipitated by the addition of excess amount of ethanol and washed three times with ethanol. Then the polymer (**4-1**) was redissolved in water and further purified by dialysis using a membrane with the molecular weight cutoff of 2000 (yield: 75%).  $^1\text{H}$  NMR (300 MHz,  $\text{D}_2\text{O}$ )  $\delta$  1.63(br s, 3H), 2.19(br s, 2H), 3.52(br s, 2H) ppm.  $^{31}\text{P}$  NMR (300 MHz,  $\text{D}_2\text{O}$ )  $\delta$  17.79(s) ppm.

Procedure II: In a two-neck round-bottom flask equipped with a reflux condenser and a nitrogen inlet, 2 ml water solution of **3** (166 mg, 0.5 mmol) was introduced. After bubbling for 10 min. with nitrogen, the flask was then transferred to a water bath at 60 °C and stirred with a continuous bubbling of nitrogen. The initiator potassium persulfate (KPS) (2.7 mg, 2.0 mol% of total monomers) was added when the temperature of water solution was up to 60 °C. Thereafter, the temperature of water bath was increased to 70 °C. After 12 h polymerization, 2.7 mg of KPS was added again under  $\text{N}_2$  protection and the reaction continued for another 12 h. The polymer was precipitated by the addition of excess amount of ethanol and washed three times with ethanol. Then the polymer (**4-2**) was redissolved in water and further purified by dialysis using a membrane with the molecular weight cutoff of 2000 (yield: 80%).  $^1\text{H}$  NMR (300 MHz,  $\text{D}_2\text{O}$ )  $\delta$  1.60-1.64(br m, 3H), 2.15(br s, 2H), 3.50(br s, 2H) ppm.  $^{31}\text{P}$  NMR (300 MHz,  $\text{D}_2\text{O}$ )  $\delta$  18.00(s) ppm.

Poly(N-acryl pamidronate-co-N-isopropylacrylamide) (**6**): In a two-neck round-bottom flask equipped with a reflux condenser and a nitrogen inlet, 5.7 ml water solution of **3** (66.6 mg, 0.2 mmol) and N-isopropylacrylamide (**5**) (429.4 mg, 3.8 mmol) was introduced. After bubbling for 10 min. with nitrogen, the flask was then transferred to a water bath at 60 °C and stirred with a continuous bubbling of nitrogen. The initiator potassium persulfate (KPS) (10.8 mg, 1.0 mol% of total monomers) was added when the temperature of water solution was up to 60 °C. Thereafter, the temperature of water bath was increased to 70 °C. After 24 h polymerization, the polymer was precipitated by the addition of excess amount of methanol and washed three times with methanol (yield: 90%). <sup>1</sup>H NMR(300 MHz, D<sub>2</sub>O) δ 1.16(s, 6H), 1.60(br s, 6H), 2.14(br s, 2H), 3.56(br s, 2H), 3.92(br s, 1H) ppm. <sup>31</sup>P NMR (300 MHz, D<sub>2</sub>O) δ 17.57(s) ppm.

**Table S1.** The molecular weight of the homopolymer (**4**) and the copolymer (**6**).

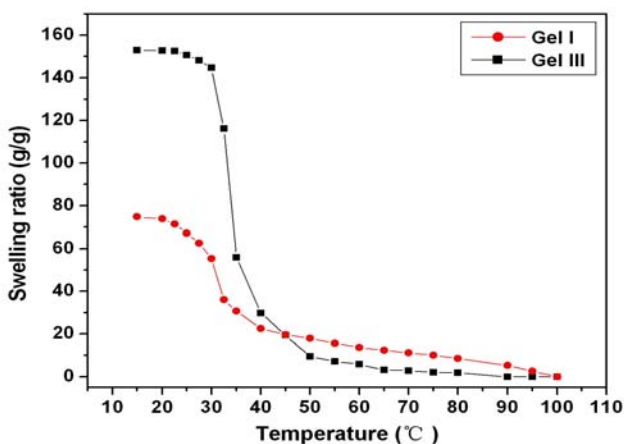
Polymer	Yield (%)	M <sub>n</sub>	M <sub>w</sub>	M <sub>w</sub> /M <sub>n</sub>
<b>4-1</b>	75	4388	5068	1.15
<b>4-2</b>	80	8147	25273	3.10
<b>6</b>	90	14971	46035	3.07

#### Temperature dependent swelling behavior of hydrogels.

The swelling ratio of crosslinked hydrogels was calculated via Eq. (1):

$$S_r = (W_w - W_d) / W_d \quad (1)$$

Where  $S_r$  is equilibrium swelling ratio,  $W_w$  and  $W_d$  are the sample weights in wet and in dry states, respectively. The temperature was raised and maintained for 8 hours to attain equilibrium for the sample. The swelling ratio of gels with different crosslinker is shown in Fig. S1. Using DHEBA as a crosslinker can dramatically increase the swelling ratios, but the phase transition temperature shows little shift because DHEBA is more hydrophilic than BIS. The swelling ratio starts to decrease from 30 °C and finally reaches to zero at 90 °C. The effect of molar ratio of **3** and **5** on swelling ratio is shown in Fig. S2. When the amount of **3** increases, the swelling ratio increases rapidly with a simultaneous enhancement of phase transition temperature due to the hydrophilicity of **3**.



**Fig.S1.** Swelling ratio dependence on temperature for Gel I and Gel III.

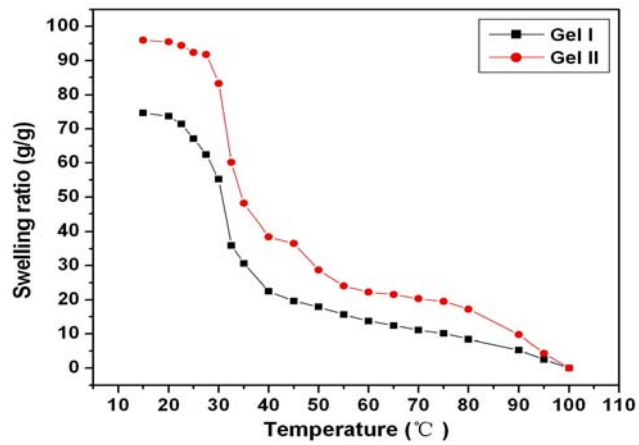


Fig.S2. Swelling ratio dependence on temperature for Gel I and Gel II.

**Energy dispersive spectroscopy (EDS) analysis:**

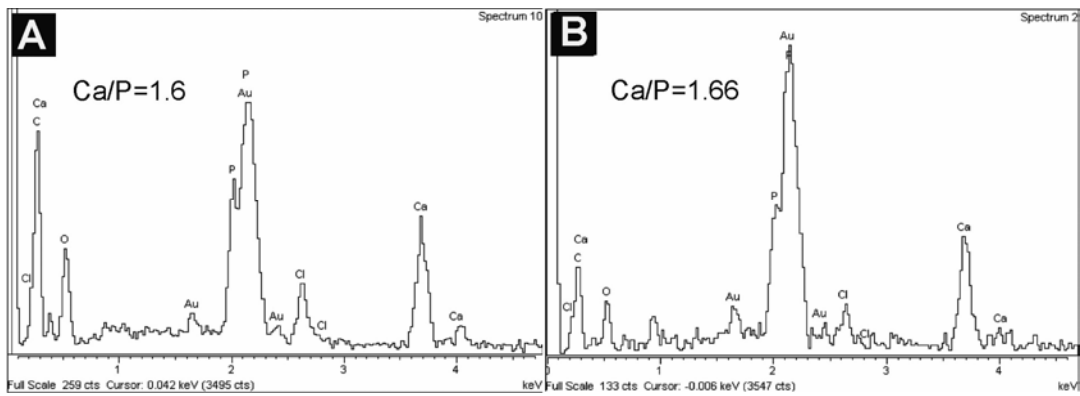


Fig. S3. SEM-associated EDS area analysis of the surface of Gel I-HA composite via urea-mediated process (A) and ammonium hydroxide-mediated process (B).