

Supporting Information For

A metal ion triggered shrinkable supramolecular hydrogel and controlled release by an amphiphilic peptide dendron

Long Qin,^a Pengfei Duan,^a Fan Xie,^a Li Zhang^a and Minghua Liu^{*a}

^a Beijing National Laboratory for Molecular Science (BNLMS), CAS Key Laboratory of Colloid, Interface and Chemical Thermodynamics, Institute of Chemistry, Chinese Academy of Sciences, Beijing, 100190, People's Republic of China.

Tel: +86-10-82615803;

E-mail: liumh@iccas.ac.cn

Experimental Section

Materials: The gelator **OGAc** was synthesized according to our previous work (P. Duan, L. Qin, X. Zhu, M. Liu, *Chem. Eur. J.* 2011, **17**, 6389-6395). Vitamin B₁ were purchased from Acros organics and used without any additional purification. Milli-Q water (18.2 MΩ·cm) was used in all cases. Solvents were purified and dried according to standard methods.

Instruments: UV-vis, FT-IR and CD spectra were obtained using JASCO UV-550, FT/IR-660 plus and JASCO J-810 spectrophotometers, respectively. X-ray diffraction (XRD) was achieved on Rigaku D/Max-2500 X-ray diffractometer (Japan) with Cu/Kα radiation ($\lambda=1.5406\text{\AA}$), which was operated at 45 kV, 100 mA. Transmission electron microscopy (TEM) images were obtained on a JEM-1011 TEM at accelerating voltages of 100 kV. Cryo-TEM was performed on a JEOL JEM-2200FS TEM operating at 200 kV equipped with a Gatan cryoholder. AFM was performed by using Tapping Mode (Nanoscope IIIa, Digital Instruments) with a pyramidal Si₃N₄ tip. TLC was performed on silica gel HF254 flake and column chromatography was carried out with 230–400 mesh silica gel. ¹H NMR spectra were recorded with a Bruker ARX400 (400 MHz) spectrometer in CDCl₃ or DMSO-*d*₆ by using Me₄Si as an internal standard. MS spectra were determined with BEFLEX III for the MALDI-TOF mass spectrometer. Elemental analyses were performed on a Carlo-Erba-1106 instrument. The rheological properties of the gel were measured at 25 ± 0.05 °C with a Thermo Haake RS300 rheometer (cone and plate geometry of 35 mm in diameter with the cone gap equal to 0.105 mm). All the photographs of the gel were taken by Canon EOS60D camera.

Procedures: For cryo-TEM sample preparation before the gel shrinkage. A small droplet of the diluted solution (5-10 μL) of **OGAc**/Mg²⁺ hydrogel was placed on a holey carbon film supported on a TEM copper grid within a controlled environment vitrification system. The specimen was blotted and plunged into a liquid ethane reservoir cooled by liquid nitrogen. The vitrified samples were transferred to the cryo-holder and cryo-transfer stage cooled by liquid nitrogen. For the TEM measurement of the hydrogel after shrinkage, the expelled water was firstly remove and the remaining shrunken gel was diluted and a small amount of diluted solution (5-10 μL) was drop-cast onto a carbon-coated copper grid (unstained). Then, the sample was cryodesiccated in a freeze dryer at -40 °C for 1 day. For AFM observation, a diluted solution of hydrogel was placed onto freshly cleaved mica and dried under vacuum for 12 h. In the case of preparing samples for XRD measurements, gels were cast onto glass plates and dried under vacuum. Pellets made from the mixture of vacuum-dried shrunken hydrogels and KBr powder were used for FT-IR spectral measurements. In the process of measuring the UV and CD spectra of hydrogels, a quartz cuvette with 0.1 mm width was used.

Formation of OGAc/metal hydrogels and shrunken gels: A typical process for the preparation of hydrogels with different metal ions is as follows: 1.34 mg **OGAc** (0.002 mmol) and the corresponding mol ratio metal salts aqueous solution (50 mmol/L, and 4 μL, 8 μL, 40 μL for **OGAc**/Metal mol ratio 10/1, 5/1, 1/1, respectively) were mixed in a sealed tube with 1 mL pure water. The mixtures were slightly heated to get a transparent solution. The sealed hot test tube was then allowed to cool down to room temperature unaffectedly and further rest for 24 h to obtain the shrunken gels, after which gelation state was recorded and the photo was taken subsequently. Gelation was confirmed by the absence of flow, as observed by the tube inversion method. The hydrogels were then used for further characterization including TEM characterization, XRD, FT-IR and CD measurement respectively.

Characterization of the hydrogel shrinkage process: The shrinking hydrogels with different divalent metal ions were prepared in a quartz cuvette with a 1 cm light path by the above gelation method and the **OGAc**/Metal mol ratio was controlled at 5/1. The shrinking process was then characterized by monitoring the transmittance change as

a function of time at 400 nm through the UV-vis spectrometer. The reversible sol-shrunk gel transition process was similarly characterized by recording the transmittance of heated sol and shrunken gel of **OGAc**/ Mg^{2+} system. **Evaluation of Shrinkage ratio and rate:** The plots of change of transmittance ratio (T_t/T_0) in **OGAc** hydrogels with different metal ions as a function of time were nonlinearly fitted by the following exponential equation with a coefficient of variation (R^2) of more than 0.98

$$Y(t) = y_0 + A_1 \exp(t/B_1)$$

where y_0 , A_1 , B_1 is the constant and the corresponding values were shown in Table S3, $Y(t)$ and t indicate the transmittance ratio and time, respectively. The shrinking ratio can be calculated by the formula: $(T_0 - T_m)/T_0$, among which T_m is the equilibrium-transmittance at adequate time, and here we use the transmittance value at 20h. Meanwhile, the shrinkage process of the hydrogels also follows first order kinetics and shrinkage rate can be calculated by the following equation.

$$r = -d(V_t/V_0)/dt = K_d(V_t - V_m)/V_0$$

where V_0 , V_t , V_m is the volume of the gel at beginning, t moment or equilibrium-shrinkage and K_d is the shrinking rate constant. As mentioned in main text, $Y(t) = T_t/T_0 \approx V_t/V_0$, so the kinetics equation can be rewritten as following:

$$r = -dY(t)/dt = K_d[Y(t) - T_m/T_0]$$

The shrinking rate was obtained through linearly plotting the $-dY(t)/dt$ with $Y(t)$ (as shown in Figure S7) and then calculating the slope of the lines. The linearly fitting parameters and variation coefficient (R^2) were listed in the Table S4. The plot was then linearly fitted, which indicated the shrinkage process can be apparently governed by the first-ordered manner. The slope K_d values were fitted and the corresponding order of shrinking rate was obtained.

Controlled release of VB₁: At first, the typical VB₁ doped with the **OGAc**/metal hydrogels was carried out as following. 2.72 mg **OGAc** was dissolved in 1 mL pure water by shaking or gentle heating, which was divided into four parts. Then 250 μL VB₁ aqueous solution (0.1 mmol/L) and 4 μL Mg^{2+} , Ca^{2+} , Zn^{2+} aqueous solution (50 mmol/L) or 4 μL pure water (as control experiment) was injected by a syringe into above solutions respectively. Next, the mixtures were heated until completely dissolved. The solutions were cooled down naturally to room temperature and rest for 5 min, after which 2.5 mL pure water was carefully spread on the top the hydrogels. The UV-vis spectra of upper solution were recorded and the release rate was calculated based on the maximum absorption of VB₁ at 262 nm subsequently.

Supporting Figures and Tables:

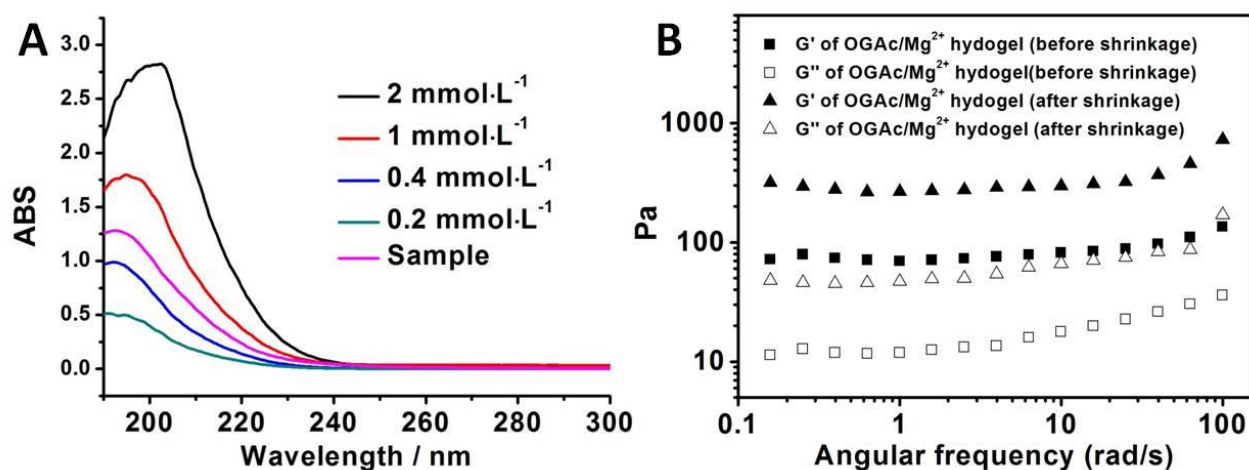


Fig. S1. (A) UV spectra of **OGAc** solution with different concentration and expelled water after **OGAc/Mg²⁺** hydrogel shrinkage (The concentration of **OGAc** in **OGAc/Mg²⁺** hydrogel before shrinkage is 2mmol L⁻¹). (B) Frequency dependence (stress = 1Pa) of the dynamic storage moduli (G') and the loss moduli (G'') of **OGAc/Mg²⁺** hydrogel before and after shrinkage.

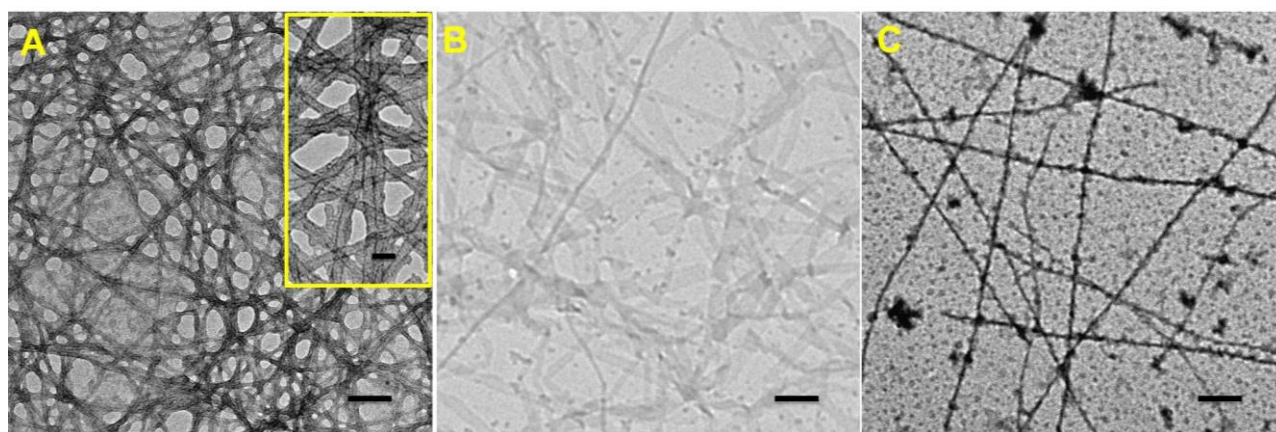


Fig. S2. (A) TEM image of **OGAc** hydrogel with magnified image inside (inside scale bar = 25 nm) (B) Cryo-TEM image of **OGAc/Mg²⁺** hydrogel before shrinkage. (C) TEM image of **OGAc/Mg²⁺** hydrogel after shrinkage. Scale bar = 100 nm.

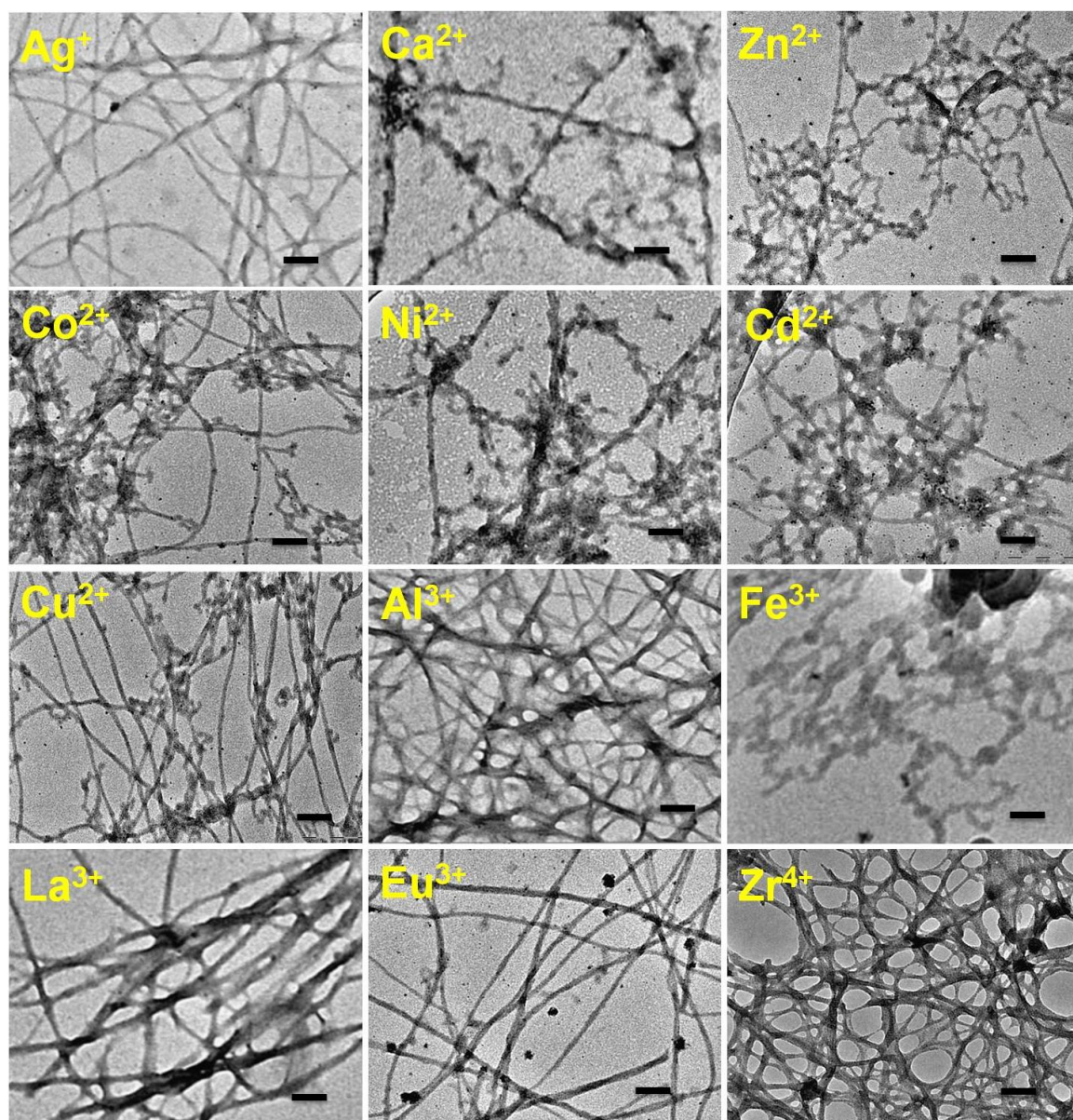


Fig. S3. TEM images of OGAc hydrogel with different metal ions. Scale bar = 100 nm.

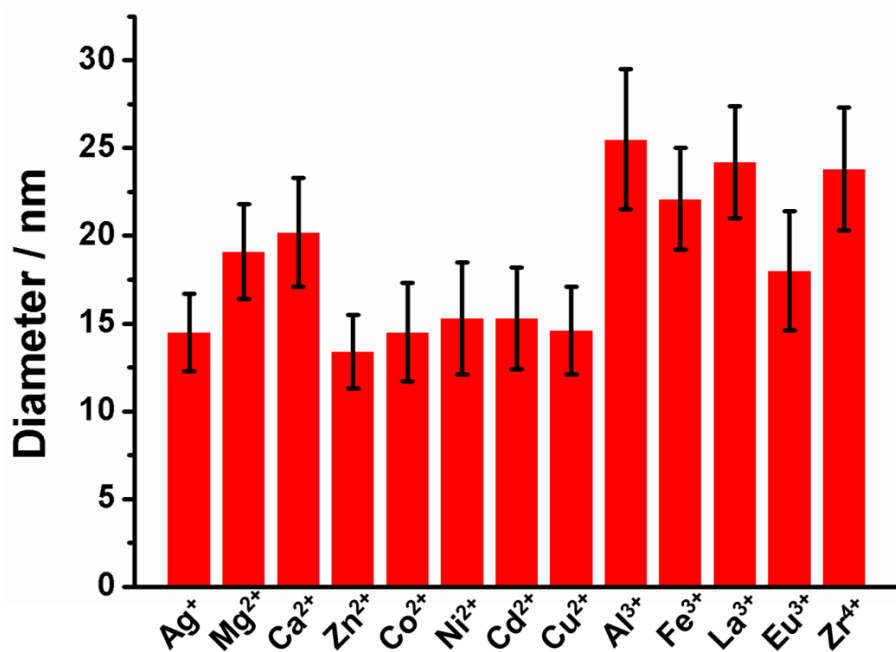


Fig. S4. Diameter of the nanofibers obtained in different OGAc/metal hydrogels

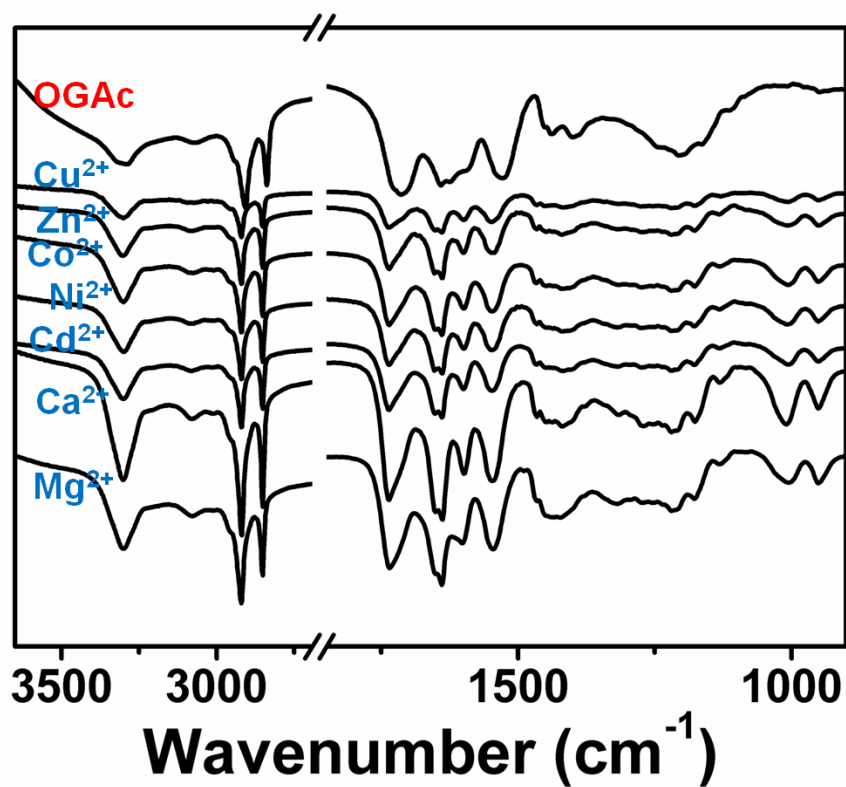


Fig. S5. FTIR of OGAc xerogel and OGAc hydrogels with different divalent metal ions

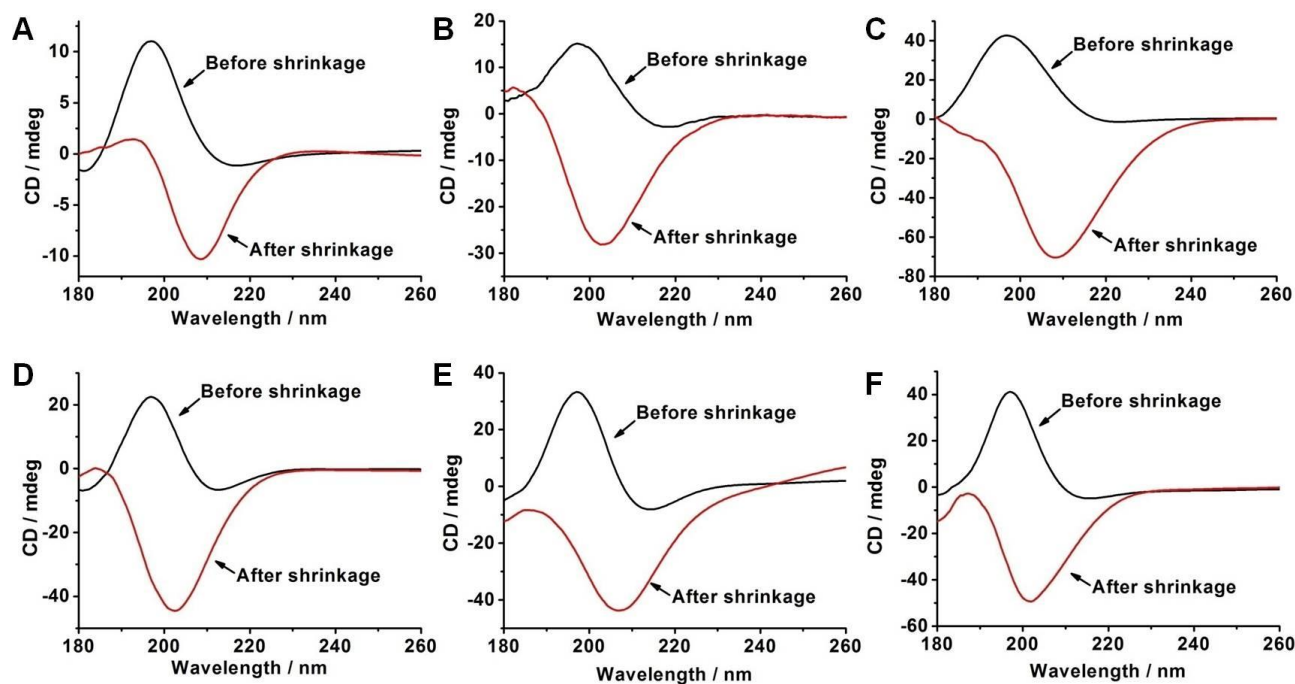


Fig. S6. CD spectra of OGAc hydrogel with (A) Ca²⁺, (B) Zn²⁺, (C) Ni²⁺, (D) Cd²⁺, (E) Co²⁺, (F) Cu²⁺ before and after shrinkage.

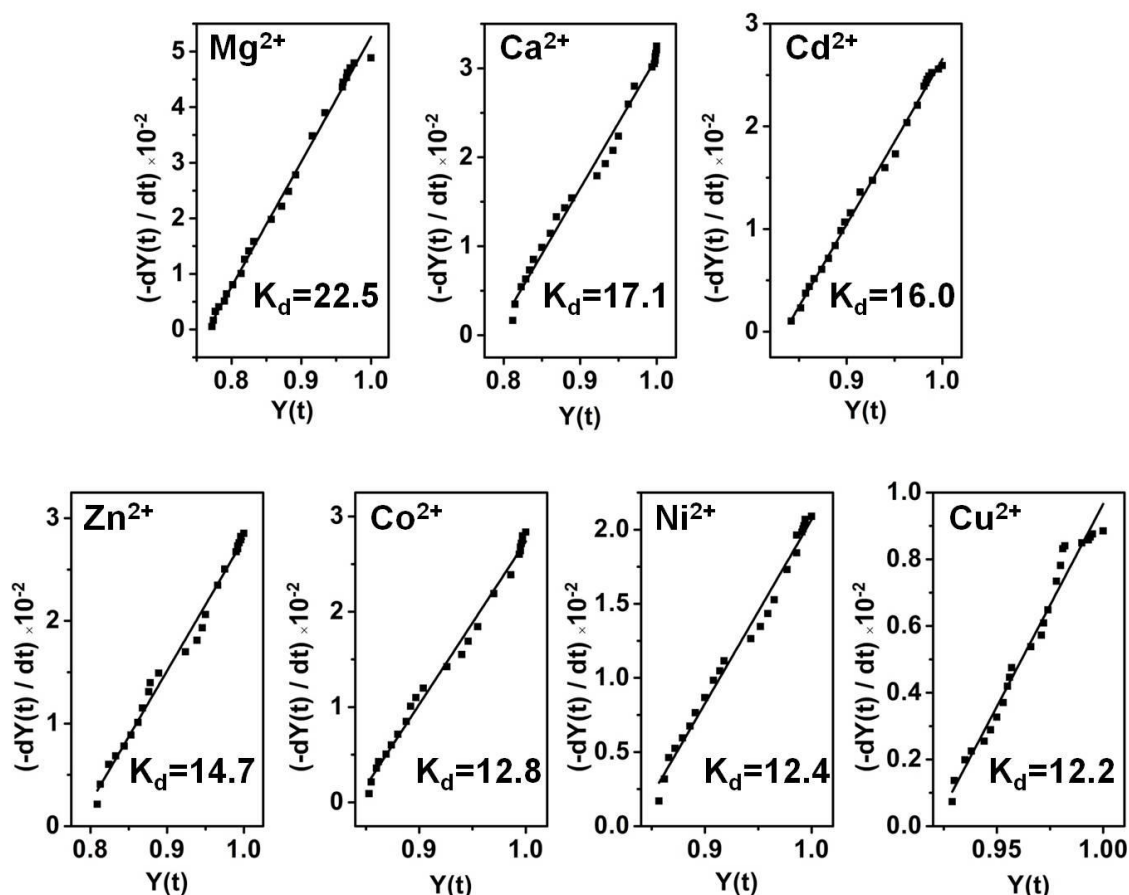


Fig. S7. Fitting linear relationship between $-dY(t)/dt$ and $Y(t)$, and corresponding shrinking rate (K_d , slope of the lines) analysis of different OGAc/metal system.

Table S1. Gelation property of **OGAc** hydrogels with various metal ions

Sample ^a	Phase ^b	CGC (wt %) ^c	Sample ^a	Phase ^b	CGC (wt %) ^c
OGAc	TG	0.3	OGAc/Ni²⁺	TG	0.06
^d OGAc/Ag⁺	TG	0.03	OGAc/Cu²⁺	TG	0.05
OGAc/Mg²⁺	TG	0.06	OGAc/Al³⁺	WG	0.07
OGAc/Zn²⁺	TG	0.06	OGAc/Fe³⁺	WG	0.08
OGAc/Co²⁺	TG	0.06	OGAc/La³⁺	WG	0.08
OGAc/Cd²⁺	TG	0.06	OGAc/Eu³⁺	WG	0.08
OGAc/Ca²⁺	TG	0.06	OGAc/Zr⁴⁺	WG	0.08

^a**OGAc/Metal** mol ratio is 5/1. ^bTG = transparent gel; WG = white gel, the phase investigation in **OGAc/M²⁺** system was obtained before shrinkage. ^cCritical gelation concentration (CGC) was measured at room temperature. ^d**OGAc/Ag⁺** mol ratio is 1/1.

Table S2. Summary of XRD peaks and *d*-spacing values of **OGAc** xerogels with various metal ions

	Ag ⁺	Cu ²⁺	Zn ²⁺	Co ²⁺	Ni ²⁺	Cd ²⁺	Mg ²⁺	Ca ²⁺	Al ³⁺	Fe ³⁺	La ³⁺	Eu ³⁺	Zr ⁴⁺
2θ	2.0	2.3	2.2	2.3	2.5	2.4	2.3	2.2	2.2	2.2	2.3	2.2	2.2
<i>d</i> (Å)	44	38	40	38	36	37	38	40	40	40	38	40	40

Table S3. Nonlinearly fitting parameters of transmittance ratio equation (T_t/T_0)

Sample ^a	Fitting function ^b	(R ²) ^c
OGAc/Mg²⁺	$Y(t) = 0.766 + 0.216_1 \exp(t/4.432)$	0.995
OGAc/Zn²⁺	$Y(t) = 0.789 + 0.218_1 \exp(t/6.712)$	0.988
OGAc/Co²⁺	$Y(t) = 0.783 + 0.220 \exp(t/7.709)$	0.988
OGAc/Cd²⁺	$Y(t) = 0.835 + 0.161 \exp(t/6.197)$	0.996
OGAc/Ca²⁺	$Y(t) = 0.841 + 0.164 \exp(t/5.798)$	0.991
OGAc/Ni²⁺	$Y(t) = 0.835 + 0.166 \exp(t/7.958)$	0.987
OGAc/Cu²⁺	$Y(t) = 0.922 + 0.071 \exp(t/8.030)$	0.983

^aThe content of the total gelators is 0.13 wt% in all cases and **OGAc/Metal** mol ratio is 5/1. ^bFitted by exponential decay equation. ^cFitting variation coefficient.

Table S4. Fitting parameters of function $-dY(t)/dt = K_d Y(t) - K_d T_m / T_0$ for **OGAc** shrunken hydrogels

Sample ^a	Fitting function ^b	(R ²) ^c
OGAc/Mg²⁺	$-dY(t)/dt = 22.464Y(t) - 17.201$	0.996
OGAc/Zn²⁺	$-dY(t)/dt = 14.741Y(t) - 11.618$	0.989
OGAc/Co²⁺	$-dY(t)/dt = 12.836Y(t) - 10.037$	0.989
OGAc/Cd²⁺	$-dY(t)/dt = 16.075Y(t) - 13.420$	0.996
OGAc/Ca²⁺	$-dY(t)/dt = 17.111Y(t) - 14.373$	0.992
OGAc/Ni²⁺	$-dY(t)/dt = 12.416Y(t) - 10.347$	0.987
OGAc/Cu²⁺	$-dY(t)/dt = 12.151Y(t) - 11.185$	0.983

^aThe content of the total gelators is 0.13 wt% in all cases and **OGAc/Metal** mol ratio is 5/1. ^bFitted by linear equation. ^cFitting variation coefficient.