

Chiral carbon dots derived from guanosine 5'-monophosphate form supramolecular hydrogels

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1.0 General information: Disodium guanosine 5'-monophosphate, $\text{Na}_2(5'\text{-GMP})$ was purchased commercially from sigma Aldrich and used without further purification.

2.0 Preparation of G-dots and G-dot gel

Preparation of G-dot: Disodium salt of guanosine 5'-monophosphate, $\text{Na}_2(5'\text{-GMP})$ (0.07 mM, 1.5 mL Milli-Q water) was microwaved at 160 °C for 5 minutes to give a viscous mild yellow solution to form the G-dots. This condition of irradiation was important because exposure to lower temperature or lesser time led to incomplete conversion while precipitates were observed on exposure to higher temperatures for prolonged time.

Preparation of G-dot hydrogels: $\text{Na}_2(5'\text{-GMP})$ (0.14 mM, 1.5 mL Milli-Q water) solution was irradiated under microwave conditions at 160 °C for 8 minutes and then cooled down to room temperature to form an opaque hydrogel. The K^+ stabilised 5'-GMP nano-dot hydrogel was prepared by heating G-dots (prepared by irradiating 0.136 mM $\text{Na}_2(5'\text{-GMP})$ in 1.5 mL 200 mM KCl solution at 80 °C for 5 minutes and then cooling down to room temperature.

3.0 TEM imaging: TEM measurements were performed on a JEOL 1200 EX electron microscope, operated at an acceleration voltage of 120 keV. The specimens were prepared by drop casting the aqueous dispersions of G-dots or G-dot hydrogel (10 μM) onto a carbon-coated 300 mesh copper grid, followed by drying under room temperature.

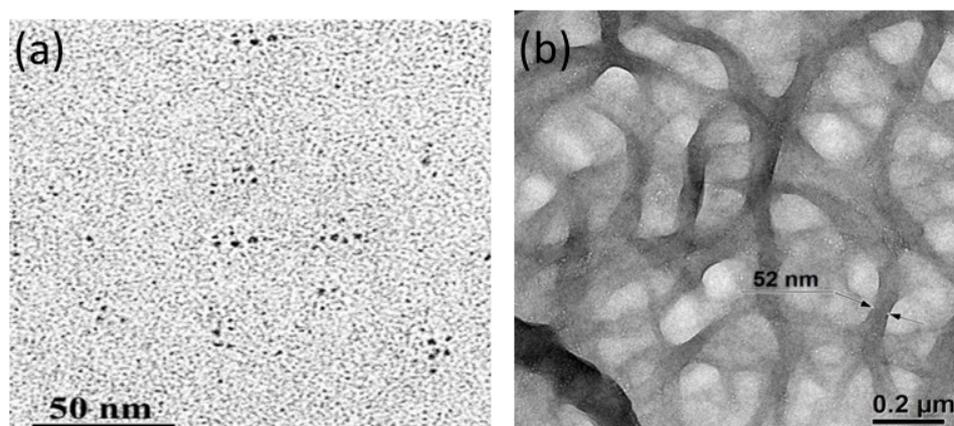


Figure S1. (a) TEM image of G-dots, (b) TEM image of G-dot gel + K^+ .

4.0 AFM Imaging: AFM measurements were performed using a tapping mode. Veeco diCP-II AFM. Disperse solutions of bare G-dots and G-dot gel were dropped onto freshly cleaved mica surface and dried in air in room temperature.

5.0 Optical property of G-dots:

(a) UV/Vis spectroscopy: The steady-state absorption spectra of G-dots were recorded with a UV-vis spectrophotometer (Cary100) at 25 °C using quartz cuvette with a 1 cm path-length.

(b) Photoluminescent spectroscopy: The steady state fluorescence spectra of G-dots were recorded with a spectrofluorimeter (Horiba Fluorolog) at 25 °C using quartz cuvette with a 1 cm path-length.

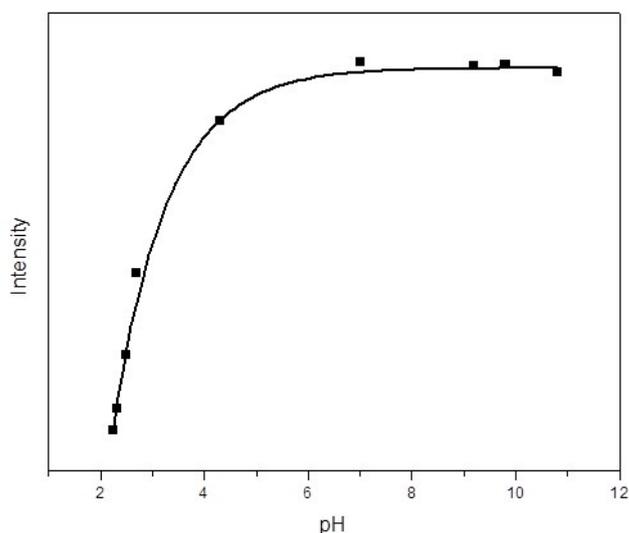


Figure S2. Fluorescence intensity of G-dots at different pH.

For quantum yield (QY) calculation quinine sulfate in 0.1 M H₂SO₄ (literature quantum yield 0.54 at 360 nm) and coumarin 6 in ethanol (literature quantum yield 0.78 at 420 nm) were chosen as standards. Absolute QY are calculated using this standard reference sample and by using the following equation:

$$QY = \frac{Q_R \frac{I_R A \eta^2}{I_R A \eta_R^2}}{I_R A \eta^2}$$

Where Q is the quantum yield, I and I_R is the measured integrated emission intensity of GMP dot and references respectively, n is the refractive index, and A is the optical density. In order to minimize re-absorption effects, absorbencies in the 1 cm fluorescence cuvette were kept under 0.05 optical density value.

The time resolved fluorescence decay experiments of G-dots was carried out using a time-correlated single-photon counting (TCSPC) spectrometer (5000, IBH). One diode lasers (377 nm) was used as the excitation sources, and an MCP photomultiplier was used as the detector. The instrument response function (IRF) of the instrument was detected by placing milk powder as a scatterer, and the FWHM of the IRF was 100ps. IBH DAS 6 (Version 2.2) decay analysis software was used for the fitting of all the decay curves by using nonlinear least-squares iteration procedure. χ^2 values and residuals plots were used as parameters to check the goodness of the fit (Table S1).

Table S1. PL decay constant of G-dots for different monitoring wavelength ($\lambda_{ex}= 377$ nm).

Excitation wavelength (nm)	Monitoring wavelength (nm)	τ_1 (ns)	B ₁	τ_2 (ns)	B ₂	Average Life time (ns)	χ^2
377	410	2.28	9.99	8.94	90.01	8.27	1.13
	450	2.44	11.25	8.89	88.75	8.16	1.11
	500	2.00	12.65	8.52	87.35	7.69	1.17
	540	1.75	13.33	8.14	86.67	7.28	1.30
	580	1.16	12.39	7.48	87.61	6.69	1.09

6.0 X-ray powder diffraction (PXRD): Powder X-ray diffraction of 5'-GMP and G-dots powders were performed in a X'Pert PRO X-ray Powder Diffractometer (PANalytical, Netherlands made) from an angle range of 0° to 100°. The spectra obtained showed a broad peak at $2\theta = 27.4^\circ$ ($d = 0.33$ nm), which is characteristic of the stacking of G-quartet structures.

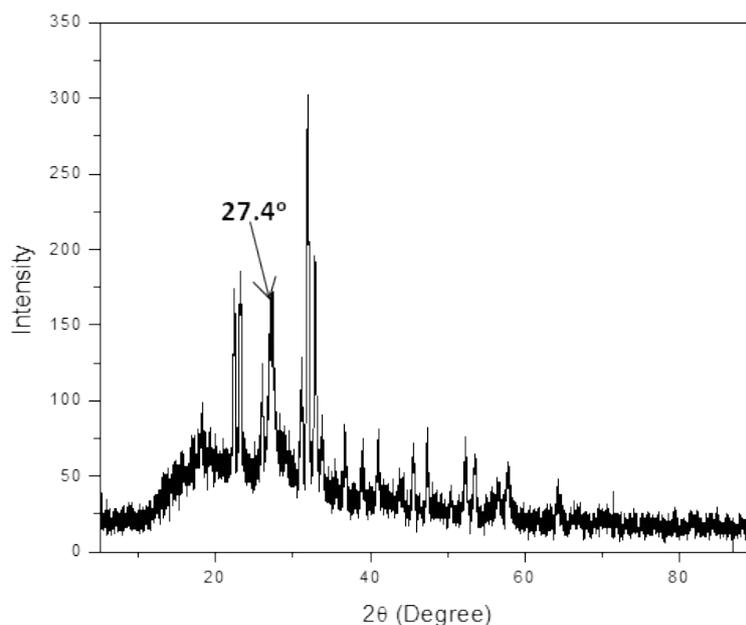


Figure S3. PXRD profile of G-dots.

7.0 Fourier Transformed Infra-red spectroscopy (FT-IR): Fourier transform infrared spectroscopy (FT-IR) spectra of the purified samples were recorded on a Nicolet 730 FT-IR spectrometer with the liquid samples.

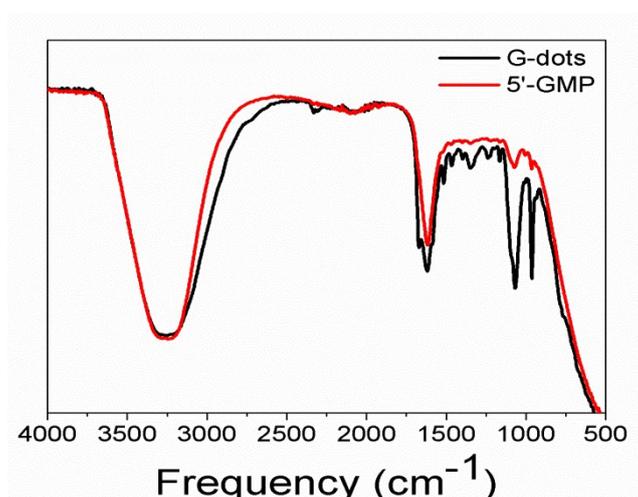


Figure S4. IR spectra of 5'-GMP and G-dots.

8.0 Surface charge analysis: A more extensive measurement of the surface charge of G-dots was analysed by zeta potential measurement of G-dots by using Zetasizer Nanosystem (Malvern Instruments Ltd.). 1 mL of G-dots solution in water was taken for zeta potential measurements. The displayed graph has been taken by three consecutive measurements with excellent repeatability. The average values of the zeta potential of G-dots were -19.4 mV.

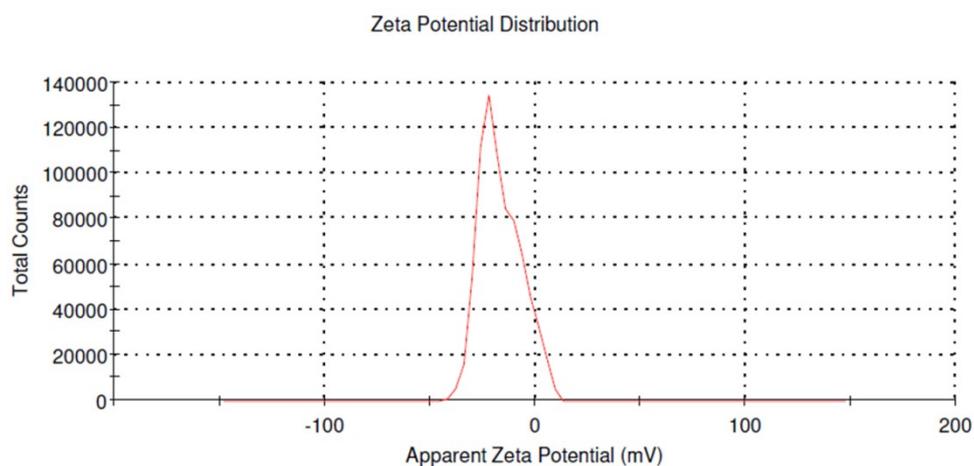


Figure S5. Zeta potential distribution of G-dots.

9.0 Raman Spectra: Raman spectroscopy measurements were carried out in the range 50-1000 cm^{-1} using the micro-Raman spectrometer LABRAM HR from Horiba Jobin Yvon with a 1800 grooves mm^{-1} grating. All the experiments were performed using the excitation wavelength of 488 nm of an Ar-ion laser and the spectral resolution was 2 cm^{-1} .

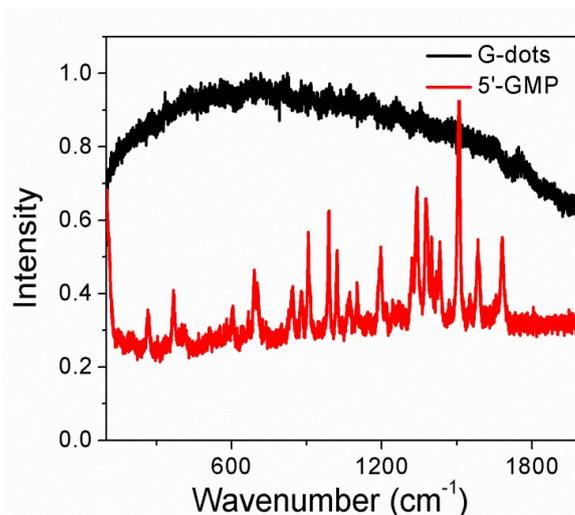


Figure S6. Raman spectra of G-dots and 5'-GMP (absence of D band and G band peak in case of G-dots).

10.0 Circular dichroism (CD) spectra: CD spectra was recorded with dilute aqueous suspension of G-dots and G-dot hydrogels at room temperature using a 1 mm quartz cuvette in a JASCO J-815 spectrophotometer. The CD spectra represented an average of three scans and were smoothed and zero corrected.

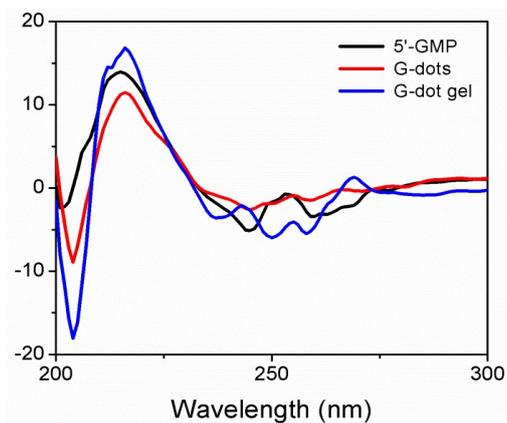


Figure S7. CD spectrum of 5'-GMP, G-dots, G-dot gel.

11.0 XPS analysis: X-ray photoelectron spectroscopy (XPS) analysis were carried out using an Omicron Nanotechnology instrument.

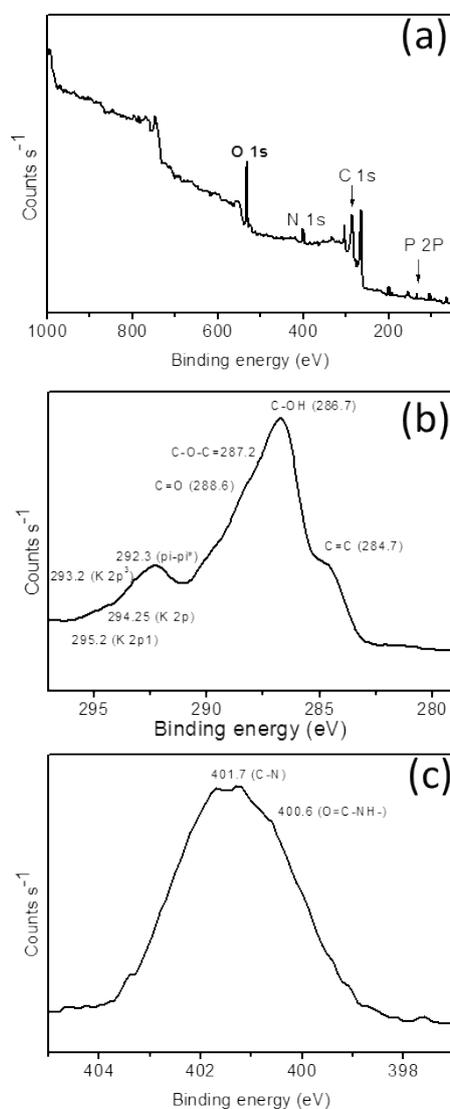


Figure S8. (a) XPS spectra of G-dots (b) XPS spectra of the G-dots (Expanded carbon region). (c) XPS spectra of the G-dots (Expanded nitrogen region)

12.0 NMR spectroscopy: ^1H NMR spectra were recorded at 500 MHz instrument at 298 K. ^{13}C NMR spectra were recorded on a 125 MHz instruments with complete proton decoupling. The NMR spectra were recorded in deuterated solvents using residual protonated solvent signals as internal standard (^1H : $\delta((\text{CD}_3)_2\text{SO}) = 2.50$ ppm, $\delta(\text{D}_2\text{O}) = 4.79$ ppm). For ^{31}P NMR spectra, 500 MHz was used and triphenylphosphine (PPh_3) was used as external standard.

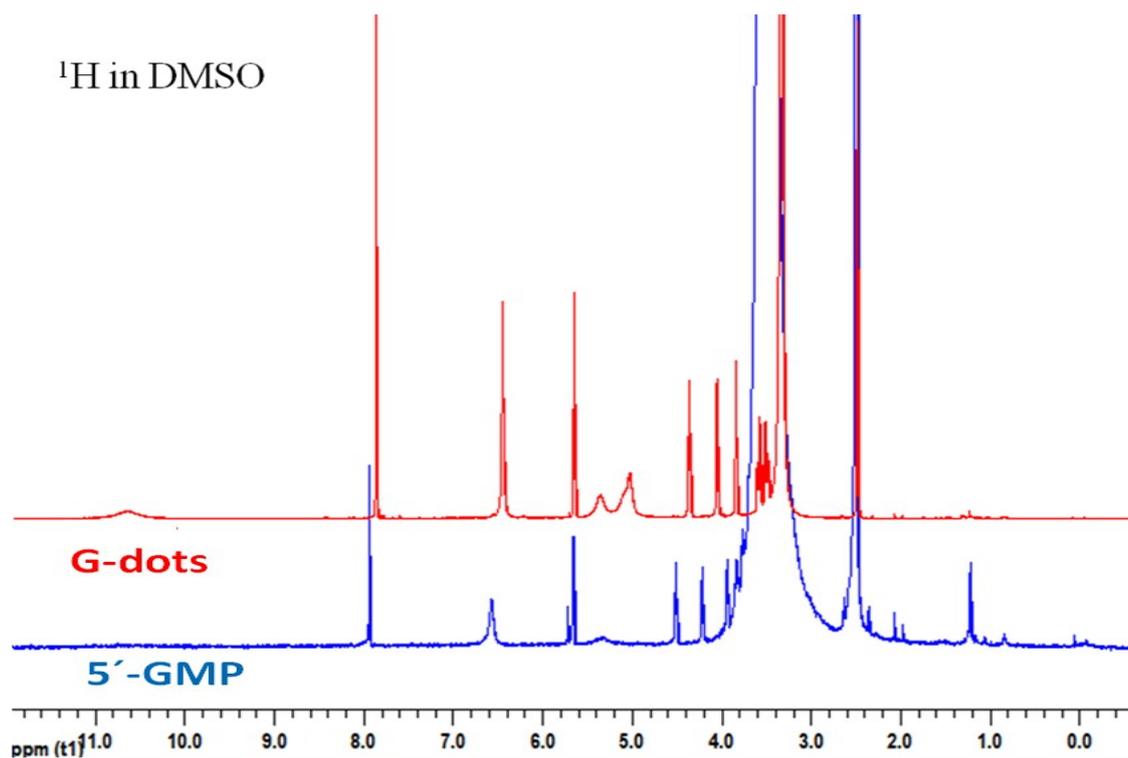


Figure S9. ^1H NMR spectra of G-dots and 5'-GMP in DMSO-d_6 .

13.0 MALDI TOF spectra: MALDI-TOF mass spectrometry were recorded using Bruker Daltonics flex Analyser in positive reflectron mode, using dithranol (DT) as the matrix. Sample (5'-GMP or G-dots) and matrix were dissolved in water at concentrations of 1, 20 mg mL⁻¹ respectively. Sample preparation involved depositing 2 μ L of the sample (5'-GMP or G-dots) and matrix mixture on the wells of a 384-well ground-steel plate and allowing the spots to dry.

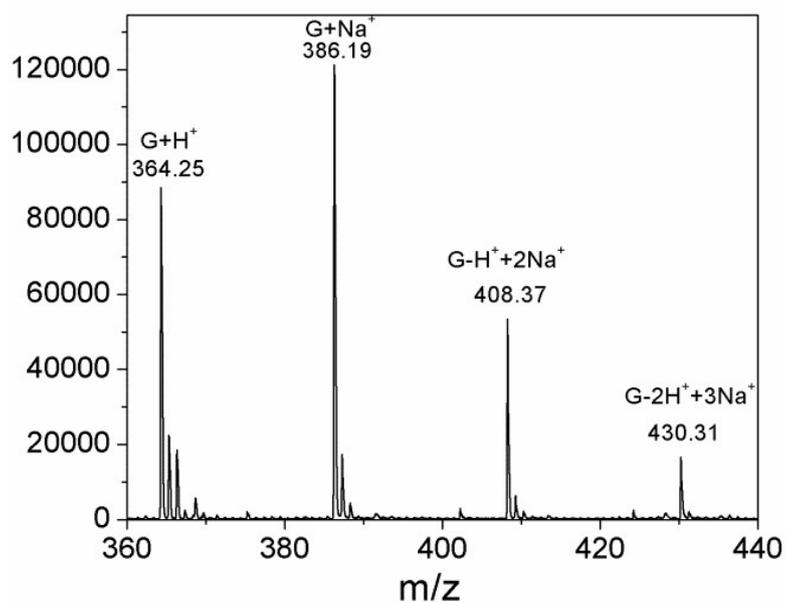


Figure S10. MALDI TOF spectra of 5'-GMP.

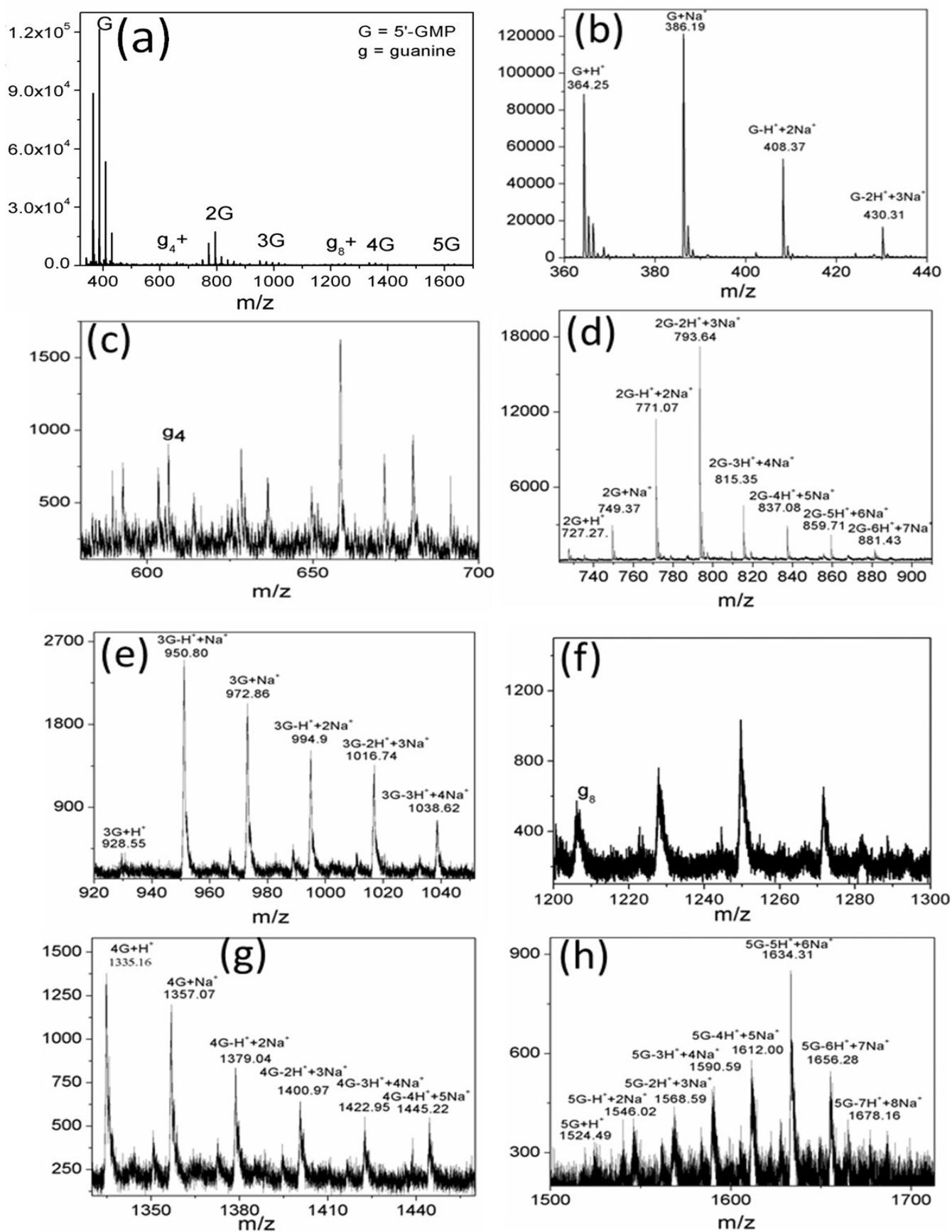


Figure S11. Expanded regions of MALDI TOF spectra of G-dots.

14.0 Differential Scanning Calorimetry (DSC): DSC was performed to determine the gel to sol transition temperature or the melting temperature of the gel. DSC was carried out with the G-dot hydrogel in the range of -10 °C to 80 °C using LVC pans in a Perkin Elmer made Diamond DSC machine at a scan rate of 2 °C min⁻¹.

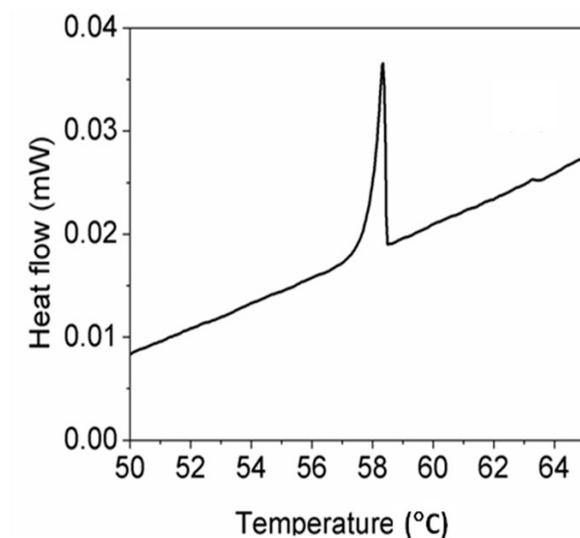


Figure S12. DSC measurement of G-dot hydrogel.

15.0 Rheological experiments: The rheological experiments of the G-dot hydrogels were performed with an advanced rheometer (AR 2000, TA Instrument, USA) using cone plate geometry on a peltier plate. The diameter of the plate was 40 mm with a cone angle of 4 degree and a plate gap of 121 μ m.