Electronic Supplementary Material (ESI) for ChemComm.

Photodimerisation of a Coumarin-Dipeptide Gelator

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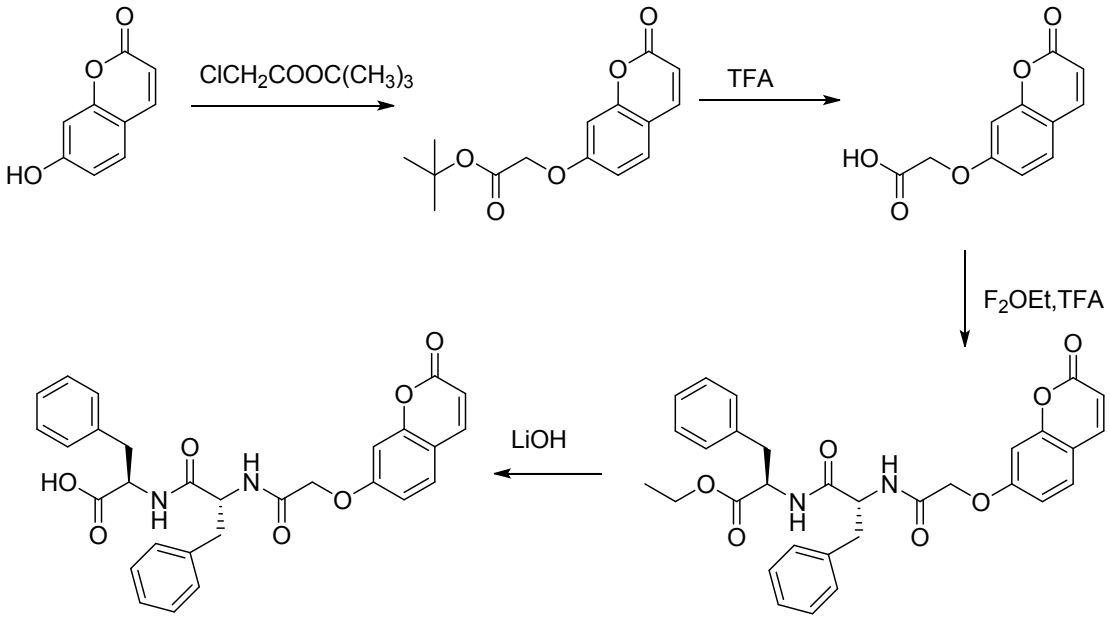
SUPPORTING INFORMATION

Synthetic Procedures

All chemicals were obtained from Sigma Aldrich and were used as received. Deionized water was used throughout.

The LMWG was prepared using the following synthetic route:



To a solution of 7-hydroxycoumarin (5.36 g, 0.033 mol) in acetone (100 mL) was added chloro-tert-butylacetate (5.21 mL, 0.036 mol) and potassium carbonate (13.8 g, 0.099 mol). The mixture was heated to 70 °C for 9 hours. After cooling, chloroform (200 mL) was added and the organic layer washed four times with water (200 mL). The organic layer was dried with magnesium sulphate and the solvent removed *in vacuo* to give a clear oil.

^{1}H NMR ($CDCl_3$) 7.63 (d, C=CH, 1H, J_{HH} = 9.5 Hz), 7.39 (d, ArH, 1H, J_{HH} = 8.6 Hz), 6.87 (dd, ArH, 1H, J_{HH} = 8.6 Hz, J_{HH} = 2.5 Hz), 6.76 (d, ArH, 1H, J_{HH} = 2.5 Hz), 6.27 (d, C=CH, 1H, J_{HH} = 9.5 Hz), 4.58 (s, OCH_2, 2H), 1.49 (s, $(CH_3)_3$, 9H) ppm. ^{13}C NMR ($CDCl_3$) 167.0, 161.1, 155.7, 143.3, 128.9, 113.7, 113.2, 112.9, 101.6, 83.1, 65.7, 41.9, 28.1 ppm. MS (ES) 299 ($[M+Na]^+$). Accurate mass calculated for $C_{15}H_{16}O_5Na$: 299.0895. Found: 299.0891.

The corresponding acid was formed by deprotecting the above product. The product was dissolved in chloroform (20 mL) and trifluoroacetic acid was added (10 mL). The solution was stirred overnight. Diethyl ether (200 mL) was added to precipitate a white solid, which was collected by filtration, washed well with ether. The solid was suspended in chloroform (100 mL), stirred for 1 hour to remove traces of the starting

material and collected by filtration. The final product was isolated as a white solid in an overall 47 % yield for both steps.

^{1}H NMR (DMSO) 7.99 (d, ArH, 1H, J_{HH} = 9.5 Hz), 7.64 (d, ArH, 1H, J_{HH} = 9.1 Hz), 6.96 (m, ArH, 2H), 6.31 (d, ArH, 1H, J_{HH} = 9.5 Hz), 4.83 (s, OCH_2, 2H) ppm. ^{13}C NMR (DMSO) 169.6, 160.8, 160.2, 155.2, 129.5, 112.8, 112.7, 112.6, 101.4, 64.8 ppm. MS (CI) 221 ($[M+H]^+$). Accurate mass calculated for $C_{11}H_8O_5$: 221.0444. Found: 221.0453.

To a solution of the coumarin (1.07 g, 0.0049 mol) in chloroform (50 mL) was added N-methylmorpholine (0.60 mL, 0.0054 mol). The solution was cooled using an ice bath. To this solution was added isobutylchloroformate (0.64 mL, 0.0049 mol), followed by a mixture of the trifluoroacetate salt of diphenylalanine ethyl ester (2.24 g, 0.0049 mol) and N-methylmorpholine (0.60 mL, 0.0054 mol)[1]. The solution was stirred overnight, before being washed with water (100 mL), dilute hydrochloric acid (0.1 M, 100 mL) and water (100 mL). The organic layer was dried using magnesium sulphate. The solvent was removed *in vacuo* to give a white solid, which was washed with methanol and dried. The final product was collected in an 87 % yield.

^{1}H NMR ($CDCl_3$) 7.64 (d, ArH, 1H, J_{HH} = 9.4 Hz), 7.40 (d, ArH, 1H, J_{HH} = 8.6 Hz), 7.16 (m, ArH and NH, 10H), 6.98 (m, ArH, 3H), 6.81 (dd, ArH, 1H, J_{HH} = 8.6 Hz, J_{HH} = 2.5 Hz), 6.76 (d, ArH, 1H, J_{HH} = 2.5 Hz), 6.31(m, ArH and NH, 3H), 4.73 (m, CHNH, 2H), 4.46 (d, OCH, 1H, J_{HH} = 14.8 Hz), 4.41 (d, OCH, 1H, J_{HH} = 14.8 Hz), 4.12 (m, CH_2, 2H), 3.08 (m, CH_2Ph, 4H), 1.24 (t, CH_3, 3H, J_{HH} = 7.1 Hz) ppm. ^{13}C NMR ($CDCl_3$) 170.8, 169.8, 167.0 160.7, 159.8, 155.7, 140.1, 135.9, 135.6, 129.3, 129.2, 129.2, 128.8,128.7, 128.6, 128.5, 127.1, 128.1, 114.2, 113.8, 112.3, 102.2, 67.3, 61.6, 53.8, 53.4, 37.9, 37.8, 14.1 ppm. MS (ES) 565 $(M+Na]^+$). Accurate mass calculated for $C_{31}H_{30}N_2O_7Na$: 565.1951. Found: 565.1939.

The above product was dissolved in THF (20 mL). After dissolution, water was added (10 mL), followed by lithium hydroxide (0.25 g). Periodically, small samples were removed and added to excess water. When no precipitate was observed (typically after around 20 minutes), water was added (200 mL). The solution was filtered and then hydrochloric acid (1 M) was added until the pH of the solution was around 4. The solid product was collected by filtration, washed well with water and dried under vacuum to give the pure product in a 75 % yield.

^{1}H NMR (DMSO) 8.41 (d, NH, 1H, J_{HH} = 7.8 Hz), 8.18 (d, NH, 1H, J_{HH} = 8.6 Hz), 7.98 (d, ArH, 1H, J_{HH} = 9.5 Hz),7.60 (d, ArH, 1H, J_{HH} = 8.4 Hz), 7.16 (m, ArH, 11H), 6.83 (m, ArH, 2H), 6.31 (d, ArH, 1H, J_{HH} = 9.5 Hz), 4.63 (m, CHNH, 1H), 4.55 (s, OCH_2, 2H), 4.46 (m, CHNH, 1H), 3.09 (dd, CHPh, 1H, J_{HH} = 13.9 Hz, J_{HH} = 5.1 Hz), 3.02 (dd, CHPh, 1H, J_{HH} = 13.9 Hz, J_{HH} = 5.1 Hz), 2.93 (dd, CHPh, 1H, J_{HH} = 8.7 Hz, J_{HH} = 8.7 Hz), 2.81 (dd, CHPh, 1H, J_{HH} = 9.9 Hz, J_{HH} = 9.8 Hz) ppm. ^{13}C NMR (DMSO) 172.8, 170.7, 166.6, 160.7, 160.2, 155.1, 144.2, 137.5, 137.4, 129.4, 129.2, 129.1, 128.1, 127.9, 126.4, 126.2, 112.8, 112.6, 101.5, 66.8, 53.6, 53.3, 37.4,

36.7 ppm. MS (ES) 537 ([M+Na]$^+$). Accurate mass calculated for $C_{29}H_{26}N_2O_7Na$: 537.1638. Found: 537.1635.

Instruments and Procedures

Mass Spectroscopy

Measurements were carried out using a Micromass LCT Mass Spectrometer in positive mode at 40 V in methanol. Samples were run by the University of Liverpool mass spectrometry service.

Nuclear Magnetic Resonance Spectroscopy

NMR spectra were recorded using a Bruker DPX-400 spectrometer operating at 400 MHz for ^{1}H NMR and 100 MHz for ^{13}C, in deuterated DMSO.

Hydrogel Formation

Gels formed using a pH switch were prepared at a concentration of 5 mg/mL of gelator unless otherwise stated. Glucono-δ-lactone (GdL) was used to lower the pH. The gelator was added to 2 mL of water with an equimolar amount of sodium hydroxide (0.1 M, aqueous). The solution was stirred until all the gelator was dissolved. This solution was then transferred to a vial containing 8 mg/mL of GdL and shaken gently. This was then left to stand to allow gelation to occur within a few hours. Rheological properties of these gels were measured around 16 hours after the GdL was added.

For gels formed electrochemically the gelator was added to 20 mL of water at a concentration of 5 mg/mL. After the gelator was fully dissolved using an equimolar amount of 0.1 M sodium hydroxide solution the pH was adjusted to pH 9 using 0.1 M aqueous hydrochloric acid. A background electrolyte of 0.1 M sodium chloride (100 μL per 10 mL solution) was then added to the gelator solution and then 7.2 mg/mL of hydroquinone. The solution was stirred until the hydroquinone was fully dissolved. Gels were formed on an indium doped tin oxide (ITO) covered glass slide working electrode cut to 2.5 cm by 5 cm. The counter and reference electrode used were Dropsens printed electrodes. The working, counter and reference electrodes were placed in the gelator solution and a current of 1 μA was applied for 800 seconds using a Dropsens potentiostat. This current reduced the pH at the glass slide by oxidising the hydroquinone to quinone. The glass slide with the now formed gel could be removed from solution. The same solution was then used to form the gels for all the rheological measurements.

Rheological Measurements

Dynamic rheological measurements were performed using an Anton Paar Physica MCR101 rheometer. Measurements were carried out using a parallel plate system with a gap of 1 mm. A 25 mm plate was used for both strain and frequency sweeps. Gels were formed electrochemically on ITO as described above. These gels were carefully removed from the glass and onto the plate so that measurements could be performed. For gels formed by GdL a vane and cup measuring system was used with a gap of 1 mm. All experiments were performed at 25 °C.

Strain sweep: Strain scans were performed from 0.1 – 1000% strain at a frequency of 10 rad/s. Gel breakdown was quoted as the strain at which the storage modulus (G′) deviates from linearity.

Frequency sweep: Frequency scans were performed at 1 rad/s to 100 rad/s under a strain of 0.5 %. The shear modulus (storage modulus (G′) and loss modulus (G″)) are read at 10 rad/s. These measurements were done within the viscoelastic region were G′ and G″ are independent of strain amplitude.

UV-Vis Absorption Measurements

Solution UV-Vis absorption data were measured using a Thermo Scientific Nanodrop 2000/2000c spectrophotometer. The spectrophotometer was used in cuvette mode where samples were prepared in PMMA plastic cuvettes with a pathlength of 1.0 cm. Aqueous samples were prepared at high pH using equimolar amounts of 0.1 M aqueous NaOH solution to gelator and made up to 2 mL with distilled water. The solution was then diluted with basic water until the absorbance was visible in the spectrum. The cuvette top was then covered before being irradiated with 365 nm LED and the absorption measured again.

UV-Vis absorption spectra of electrochemically formed gels were recorded by removing the gel from glass slides and dissolved in DMSO and diluted until a spectrum could be recorded. For pieces of the same gel, samples were diluted by the same amount each time.

UV-Vis absorption measurements of gelled samples were prepared at a concentration of 1.25 mg/mL in a 1 mm pathlength quartz cuvette. Measurements were recorded using a Shimadzu UV-2550 UV−Vis spectrophotometer running the UV Probe software, version 2.34 with a slit width of 5 nm. Again the cuvette top was covered before being irradiated.

Fluorescence Measurements

Fluorescence was carried out on the gels formed by GdL and solutions at a concentration of 1.25 mg/mL using 1 mm pathlength PMMA plastic cuvettes with a

pathlength of 1 cm. Measurements were recorded using a Perkin Elmer LS 55 Fluorescence Spectrometer with 5 nm slit widths and at an excitation of 340 nm.

Irradiating Samples

Gels were kept on the ITO glass electrode and placed inside a plastic petri dish with a wet paper towel in to keep the air saturated with water and to prevent the gel drying out. The lid of the petri dish had a hole cut out to allow the LED to be able to irradiate the sample. A 365 nm LED (LedEngin Inc, LZ1-10U600) light source powered by a TTi QL564P power supply operating at 1.0 W was used to irradiate gel samples. The sample was moved around to ensure whole sample was exposed to the UV light. Samples were exposed for different set time times before their rheological properties were measured. Samples for UV-vis were prepared in cuvettes and again left for set times before measurements were recorded.

SEM Imaging

SEM images were obtained using a Hitachi S-4800 FE-SEM at 3 keV. Gel was deposited onto glass cover slips which were fixed onto aluminium SEM stubs with carbon tabs and left to dry for 24 hours. The samples were gold coated for 3 minutes at 15 mA prior to imaging using a sputter coater (EMITECH K550X). Fibre widths were measured from the SEM images. The diameter of at least 90 fibres were measured using the ImageJ line tool (version 1.49) and a frequency distribution obtained.

Temperature Measurements

Temperature of the gels was recorded before and after irradiation with 365 nm LED using a Precision Gold N85FR infrared thermometer with dual laser targeting. The temperature of the gels before irradiation was typically around 18 °C and after irradiation for 2 hours the temperature of the gels rose to 20 °C.

FTIR Spectroscopy

IR spectra were collected on a Bruker Tensor 27 FTIR spectrometer at a resolution of 2 cm^{-1} with spectral averaging over 64 scans. Measurements were collected using the ATR accessory. Both GdL and electrochemically formed samples were measured wet and measured before and after irradiation with UV light.

Figures

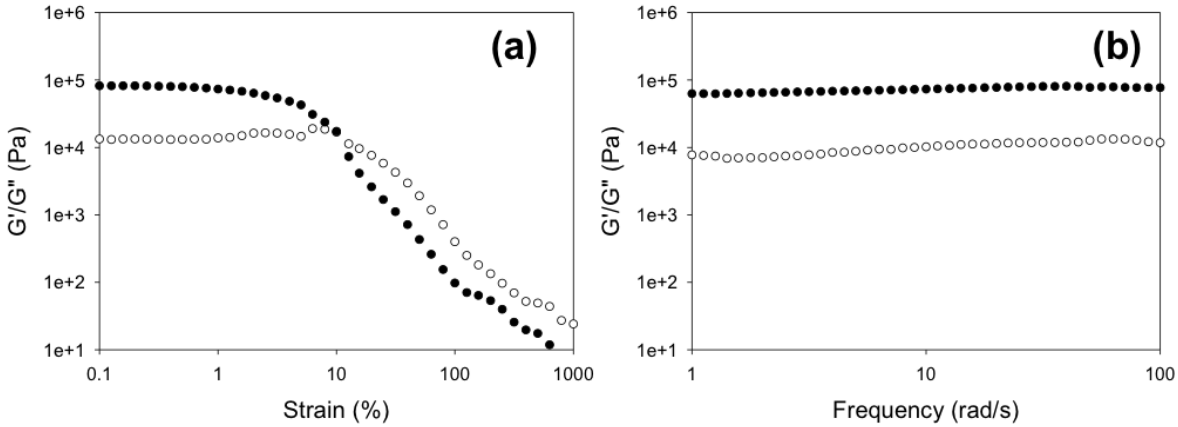


Figure S1. Rheology data of GdL grown gels (a) strain sweep performed at 10 rad/s and (b) frequency sweeps performed at 0.5% strain. Full shapes are G′ and open shapes are G″.

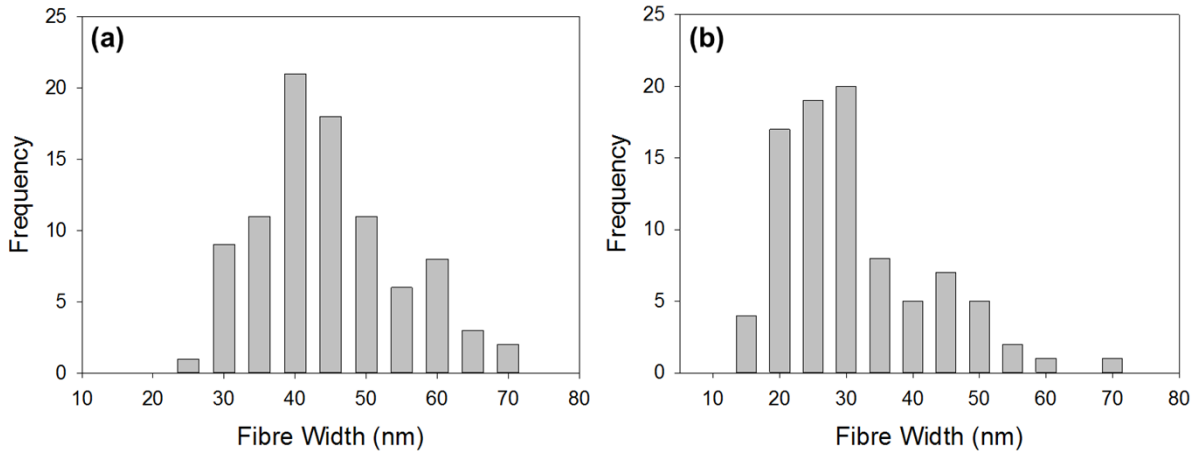


Figure S2. Graphs showing distribution fibres widths from SEM images of gels formed by GdL (a) before irradiation and (b) after irradiation.

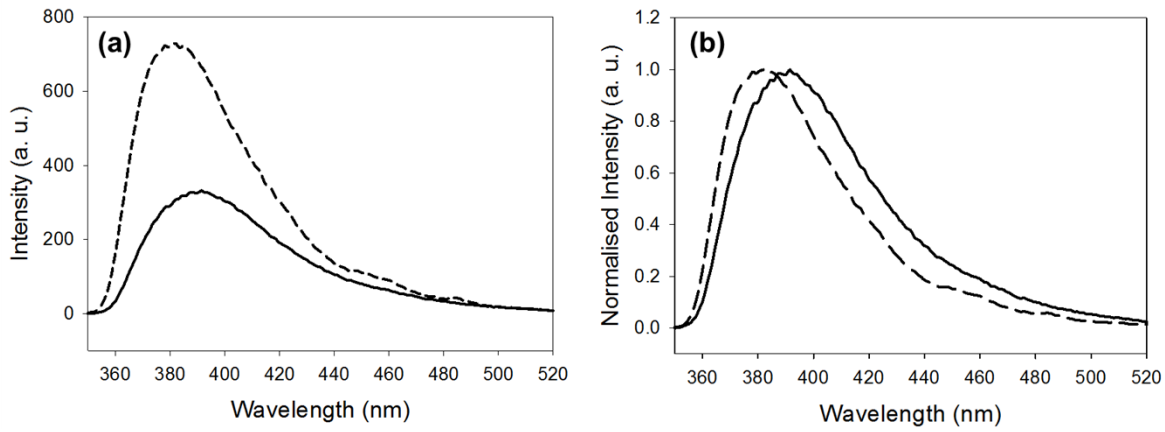


Figure S3. Fluorescence spectra of gelator in solution (solid line) and gelled using GdL (dashed line). (a) has not been normalised and (b) has been normalised.

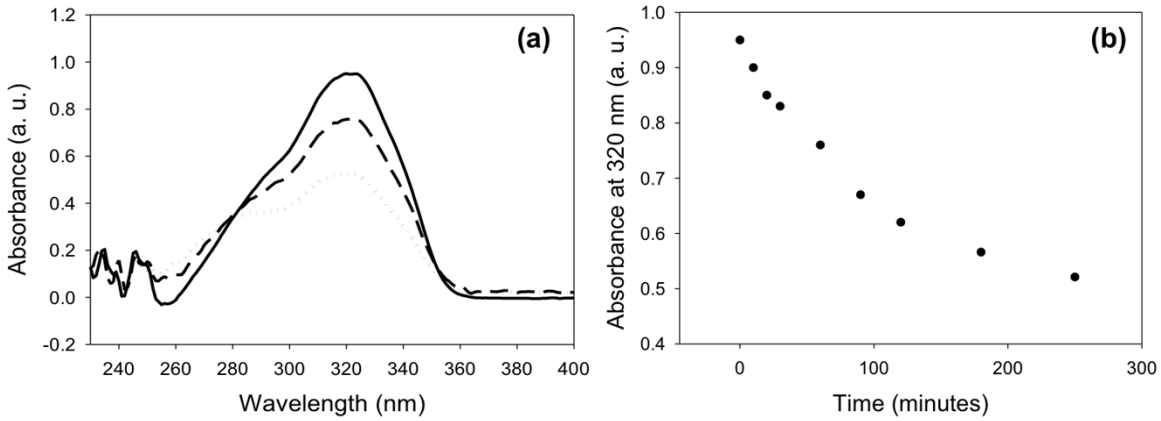


Figure S4. (a) UV-vis absorption spectra of solution (solid line) and after 15 minutes (dashed line) and 120 minutes (dotted line) of irradiation with 365 nm LED. (b) Graph showing decrease of absorption of solution at 320 nm after irradiation with 365 nm LED.

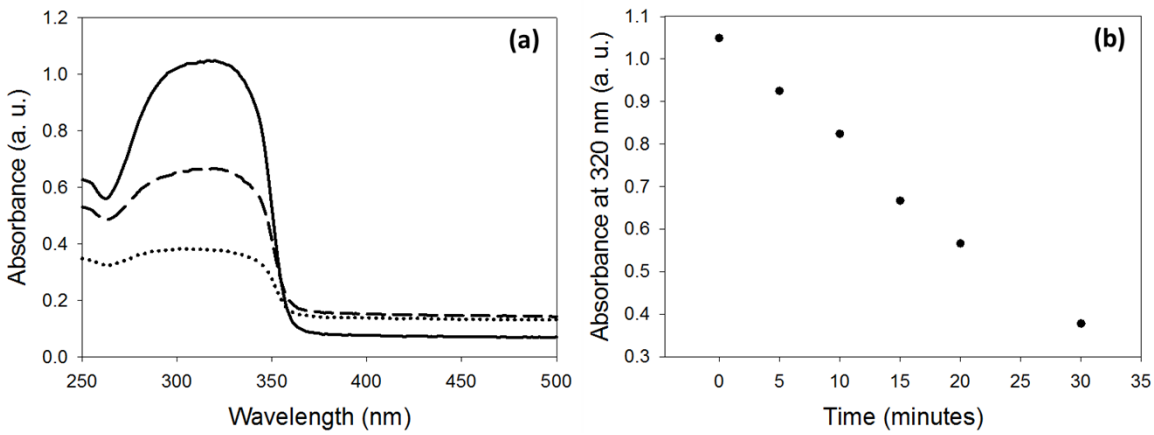


Figure S5 (a) UV-vis absorption spectra of gel (solid line) and after 15 minutes (dashed line) and 30 minutes (dotted line) of irradiation with 365 nm LED. (b) Graph showing decrease of absorption of gel at 320 nm after irradiation with 365 nm LED.

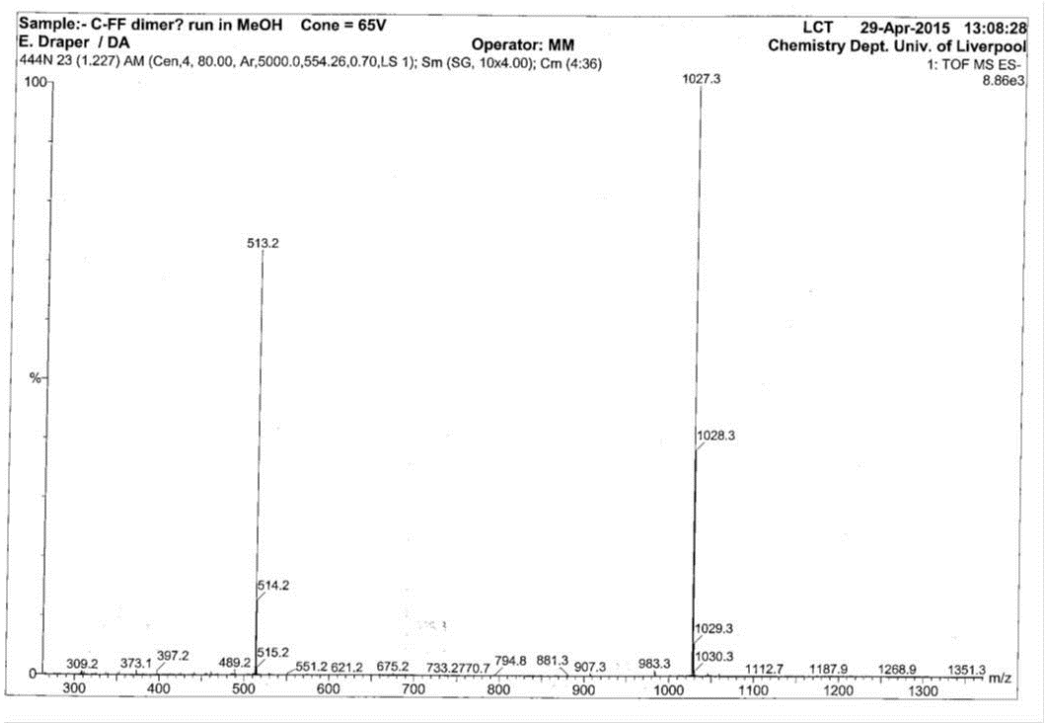


Figure S6. Mass spectrum of air dried gel formed by GdL after 4 hours of irradiation with 365 nm LED showing $[M^{-}H]^{-}$ peaks for coumarin gelator and the coumarin dimer.

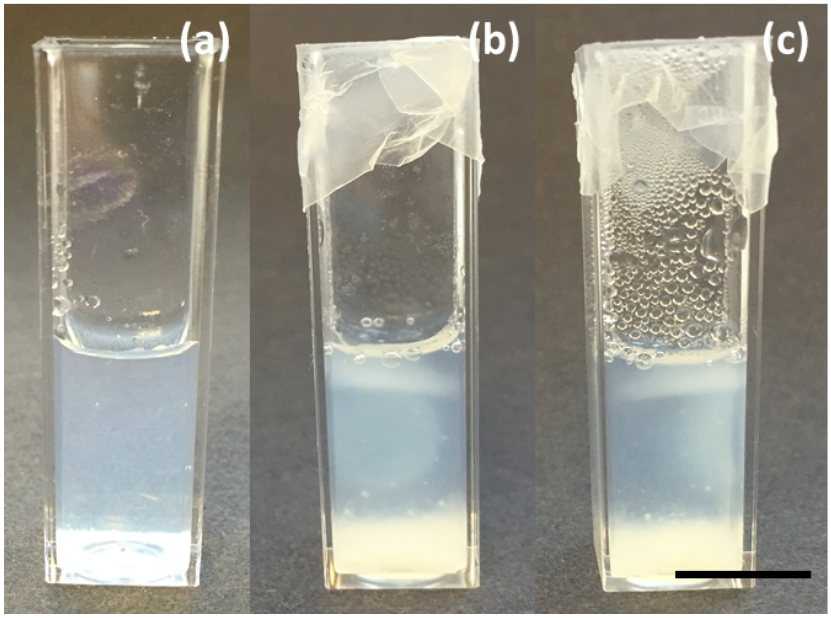


Figure S7. Photograph showing gel (a) before irradiation (b) after 2 hours of irradiation viewed from the front (c) and viewed from the side showing irradiation of gel is not homogeneous and is more pronounced at the front of the gel. Scale bar represents 1 cm.

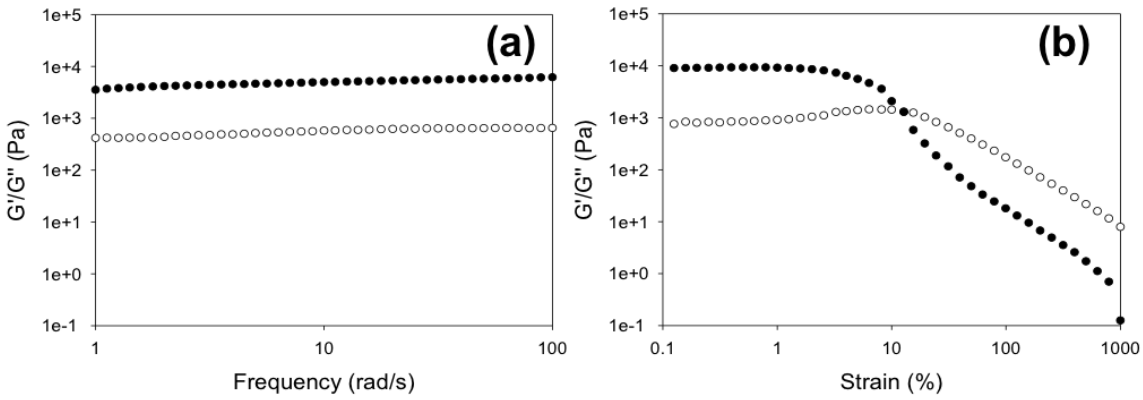


Figure S8. Rheology data of electrochemically grown gels (a) strain sweep performed at 10 rad/s and (b) frequency sweeps performed at 0.5% strain. Full shapes are G′ and open shapes are G″.

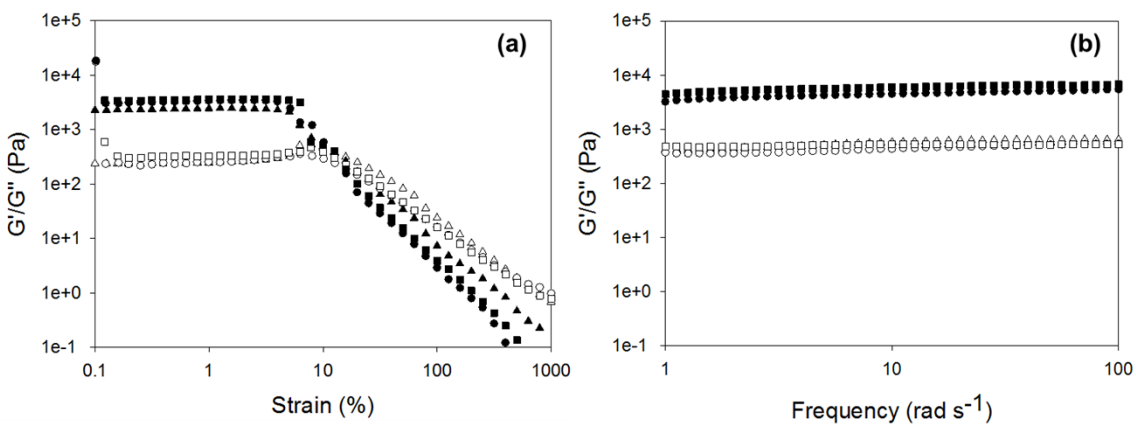


Figure S9. Rheology data showing the reproducibility of electrochemically grown gels (a) strain sweep performed at 10 rad/s and (b) frequency sweeps performed at 0.5% strain. Full shapes are G′ and open shapes are G″.

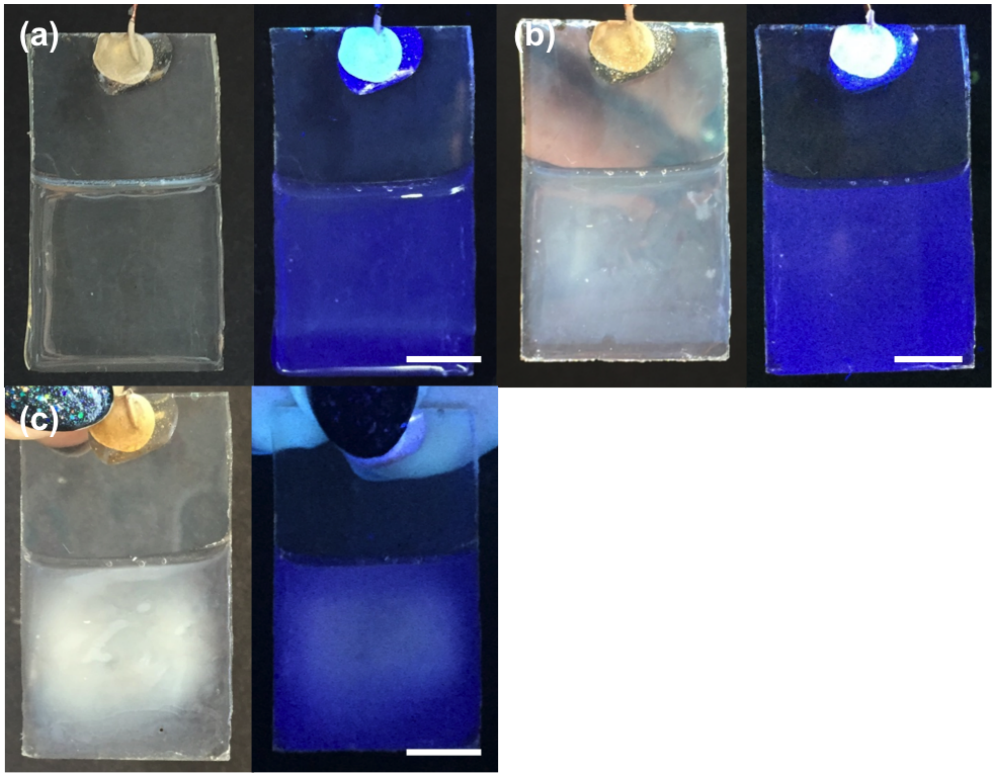


Figure S10. Photographs of electrochemically grown gels (a) before irradiation (b) after 15 minutes irradiation with 365 nm LED and (c) after 1 hour irradiation. Images on the left are in natural light and right under UV light. Scale bars represent 1 cm.

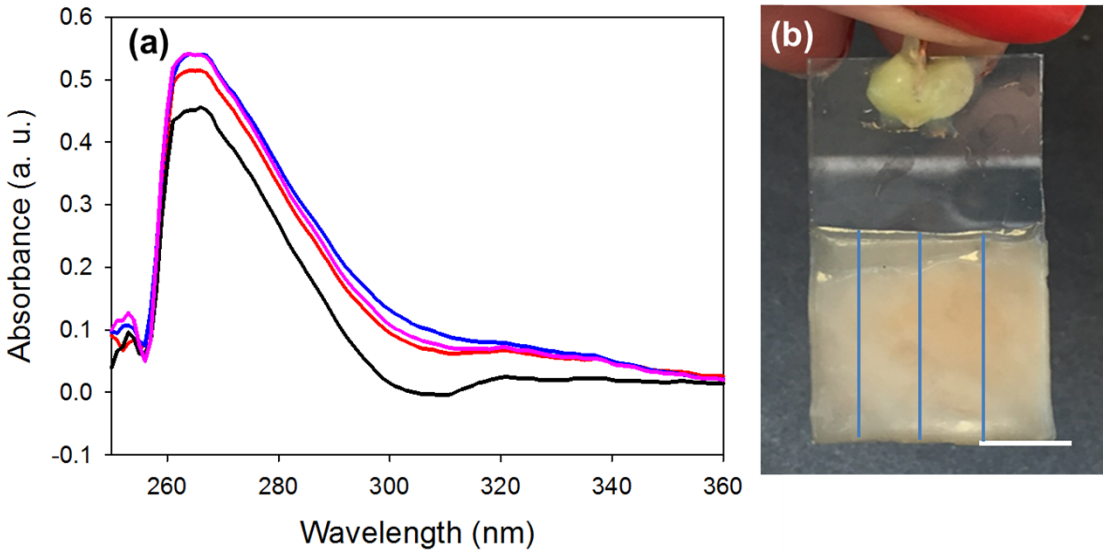


Figure S11. UV-vis spectra of different parts of the gel dissolved in DMSO. (b) Photograph of electrochemically formed gel after irradiation for 1 hour. The blue lines show how the gel was divided for the UV-vis measurements. The scale bar represents 1 cm.

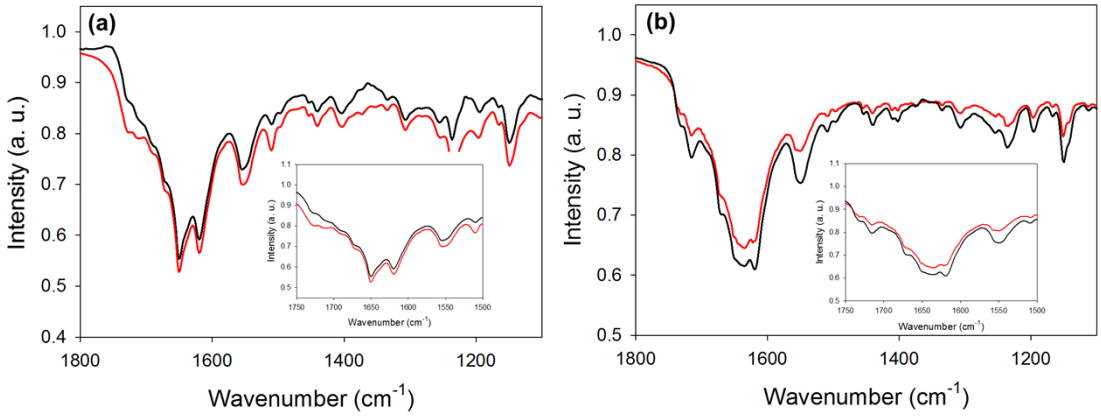


Figure S12. FTIR spectra of gels before irradiation (black line) and after irradiation (red line) (a) is gels formed by GdL and (b) is electrochemically formed gels. Inset is the peaks in the amide I region between 1500-1750 cm^{-1}.

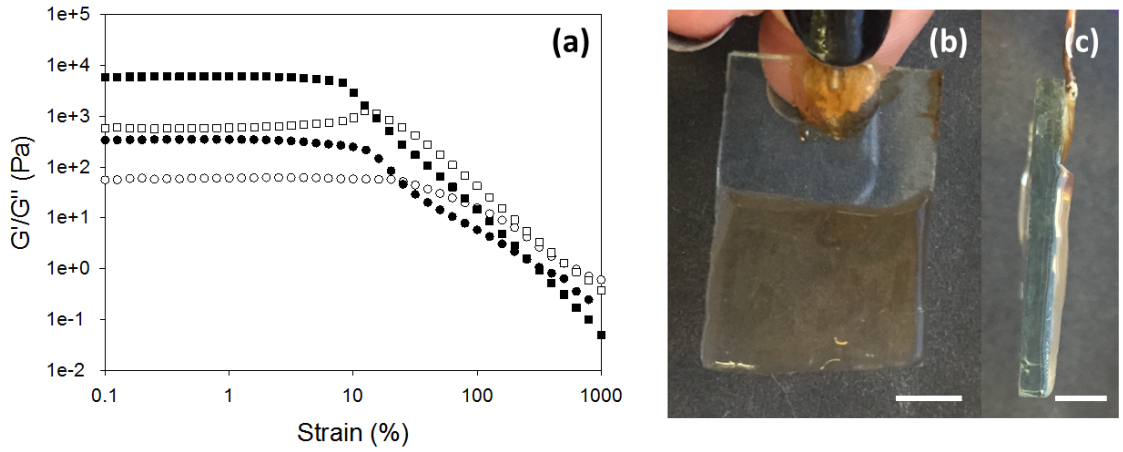


Figure S13. (a) Rheology strain sweeps of sample in hydrated chamber after two hours (circles) and sample irradiated with 365 nm LED for two hours (squares). Tests performed at 10 rad/s. Full shapes are G′ and open shapes are G″. (b) Photograph from the front of gel after two hours in chamber showing no increase in turbidity. (c) Photograph of same gel as viewed from the side showing the gel has not dried out. Scale bar represents 1 cm.

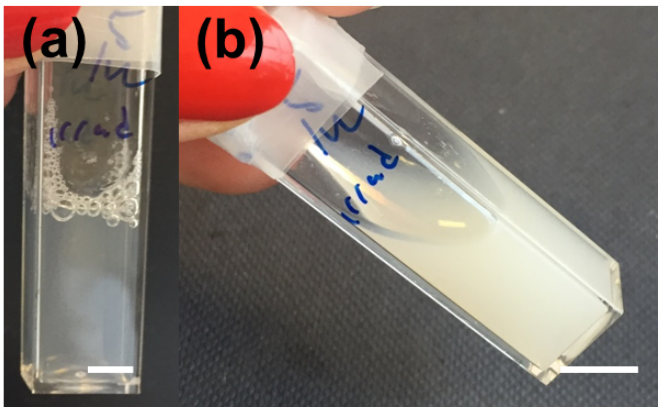


Figure S14. Photograph of a solution after irradiation (a) at high pH and (b) at low pH, showing no gel is formed due to the presence of the dimer. Scale bars represent 0.5 cm.

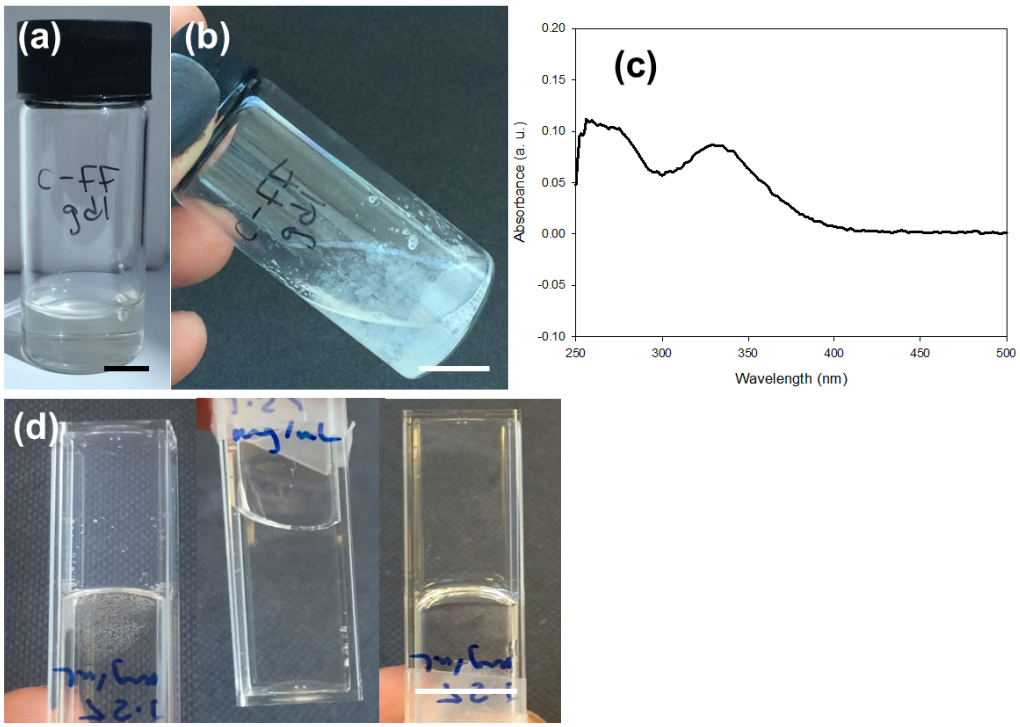


Figure S15. (a) Photograph showing an irradiated gel of gelator formed by GdL re-dissolved at high pH to form a transparent solution. (b) Photograph of when the pH is lowered again using GdL and no gel is formed. (c) UV-vis of solution in (a) showing that the dimer is present. (d) Control showing when a gel is not irradiated and the dimer is present that gelation is reversible with change in pH giving a transparent gel. Scale bars in all photographs represent 1 cm.

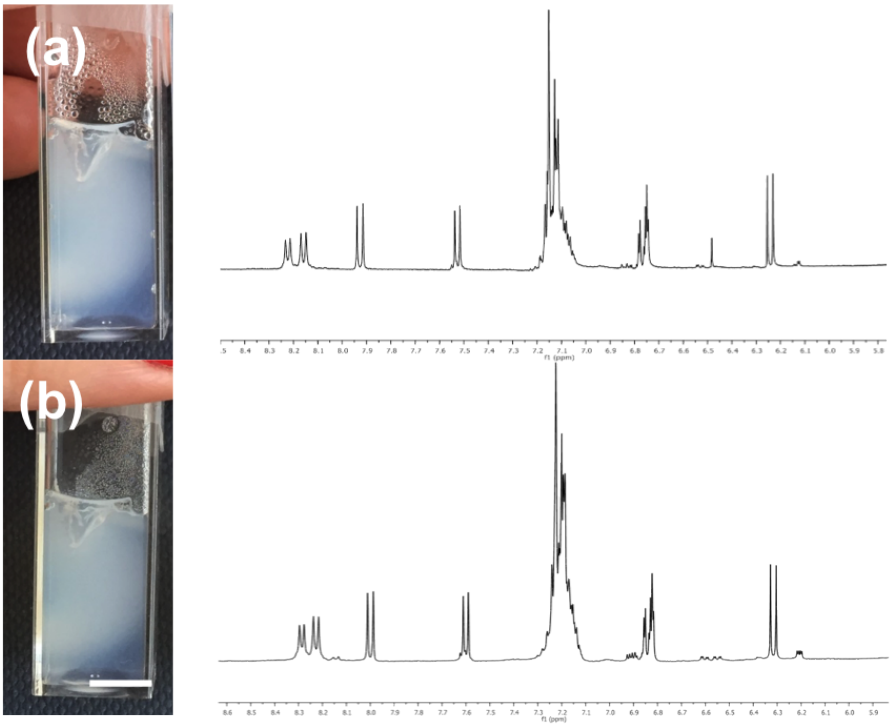


Figure S16. (a) Photograph showing a gel formed using GdL irradiated with 365 nm for 2 hours and the corresponding NMR spectrum of the dried gels re-dissolved in deuterated DMSO. (b) Photograph of (a) after being irradiated with 254 nm for 5 hours and again the corresponding NMR spectrum. Scale bar represents 0.5 cm.

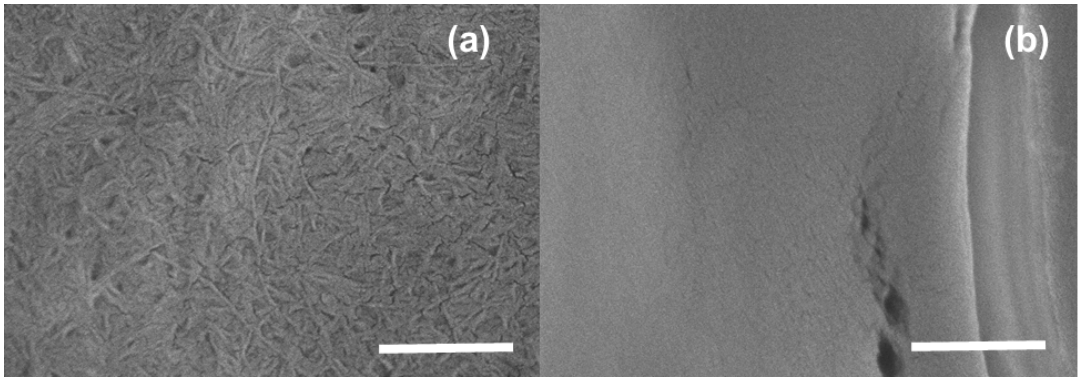


Figure S17. SEM of gels formed electrochemically before (a) and after irradiation with a 365 nm LED for an hour (b). Scale bar represents 1 μm.

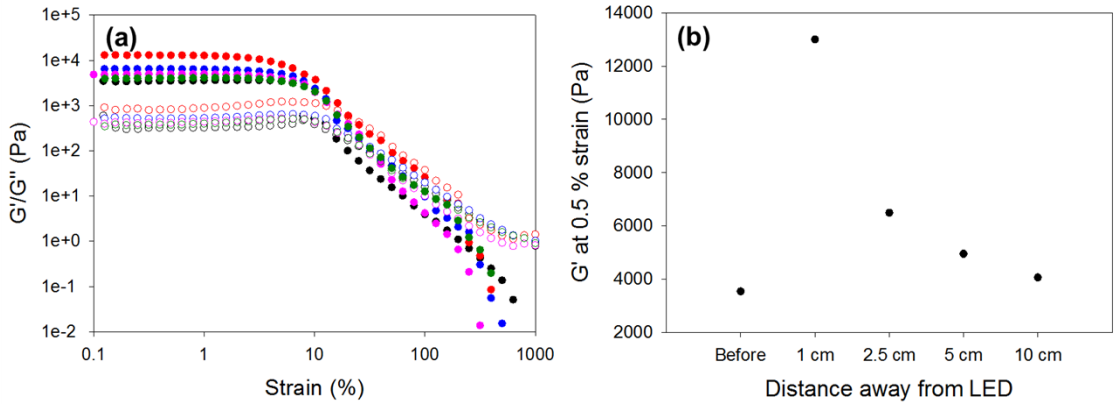


Figure S18. (a) Rheology data collected when placing the 365 nm LED at different distances away from the gel and irradiating for 15 minutes. Full circles are G′ and open circles are G″. Black data is before irradiation; red data is 1 cm away from the sample. Blue data is 2.5 cm away, pink is 5 cm away and green is 10 cm away. Strain sweeps were preformed at 10 rad/s. (b) Graph showing G′ at 5 % strain against distance away from the sample.

References

1. D. J. Adams and I. Young, *J. Polym. Sci. A Polym. Chem.*, 2008, 46, 6082-6090.