

Supplementary Material

Caffeine release from magneto-responsive hydrogels controlled by external magnetic field and Calcium ions and its effect on the viability of neuronal cells

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Figure S1 - The discs of hydrogels were positioned inside the wells of a 24-well plate. 24 NdFeB magnets (0.4 T) were arranged in a 24-well plate with a north face oriented upwards. 24 NdFeB magnets (0.4 T) were arranged with the south face oriented downwards in another plate. The 24-well plate containing the hydrogels was positioned between the magnet plates.

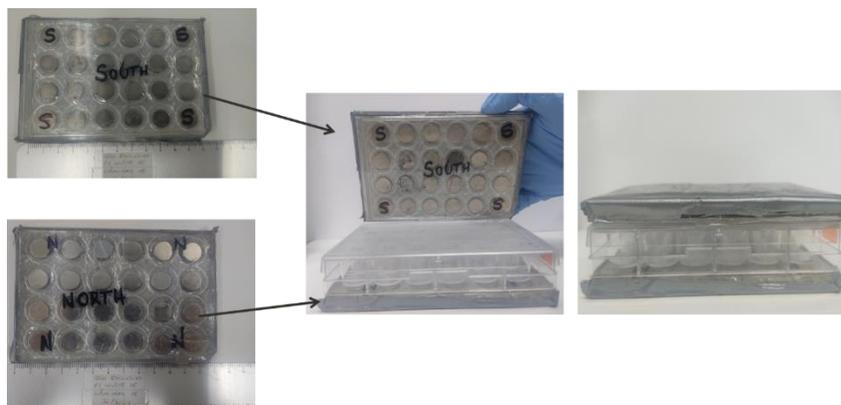


Figure S2 - (a) UV-Vis absorption spectrum of CAF at 0.02 g L⁻¹ and (b) CAF calibration curve performed at 272 nm.

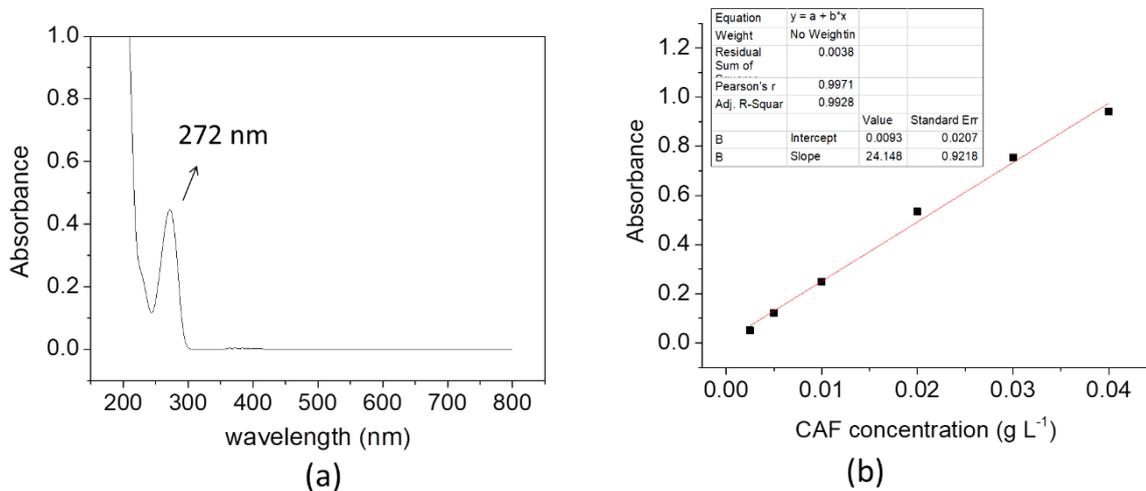


Figure S3 - The experimental setup used for the diffusion assays of Ca²⁺ ions through the hydrogels from the lower compartment to the upper compartment.

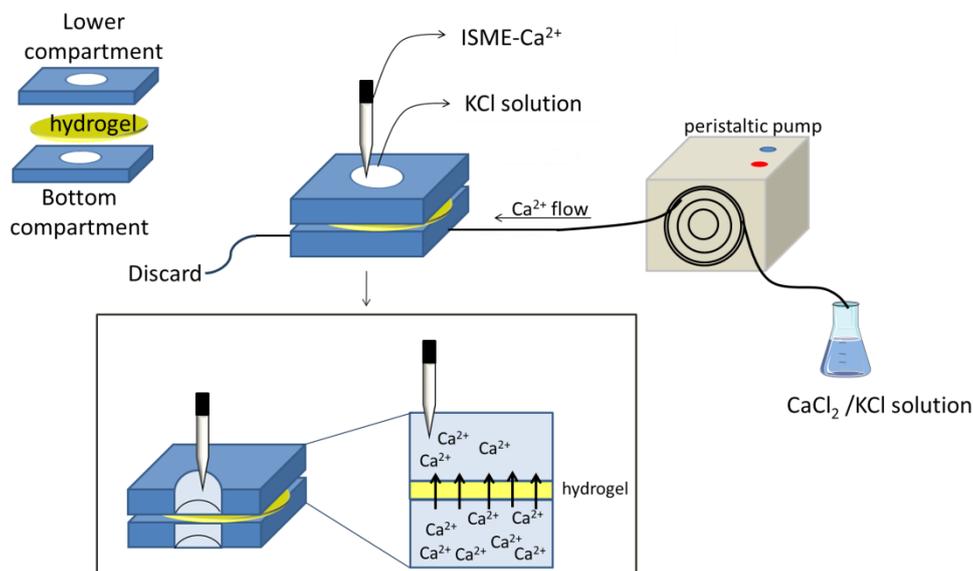


Figure S4 - X-ray diffraction pattern of Fe_3O_4 nanoparticles synthesized by co-precipitation method.

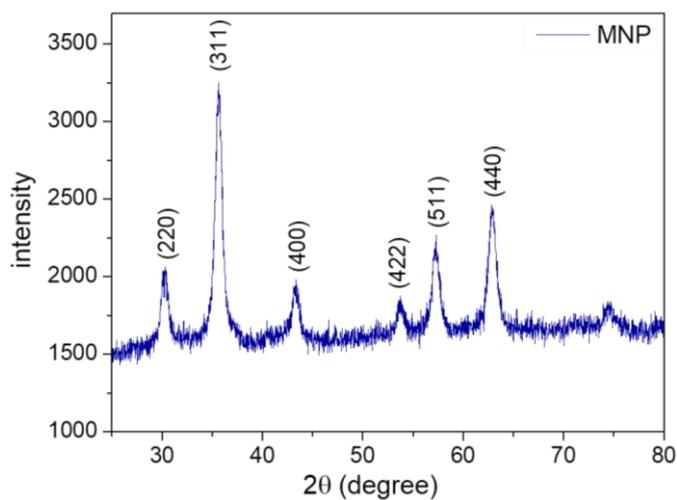


Figure S5 - (a) and (b) AFM topographic images of the MNP deposited on Si wafers by spin coating and (c) size distribution of MNP determined by AFM images ($n = 106$).

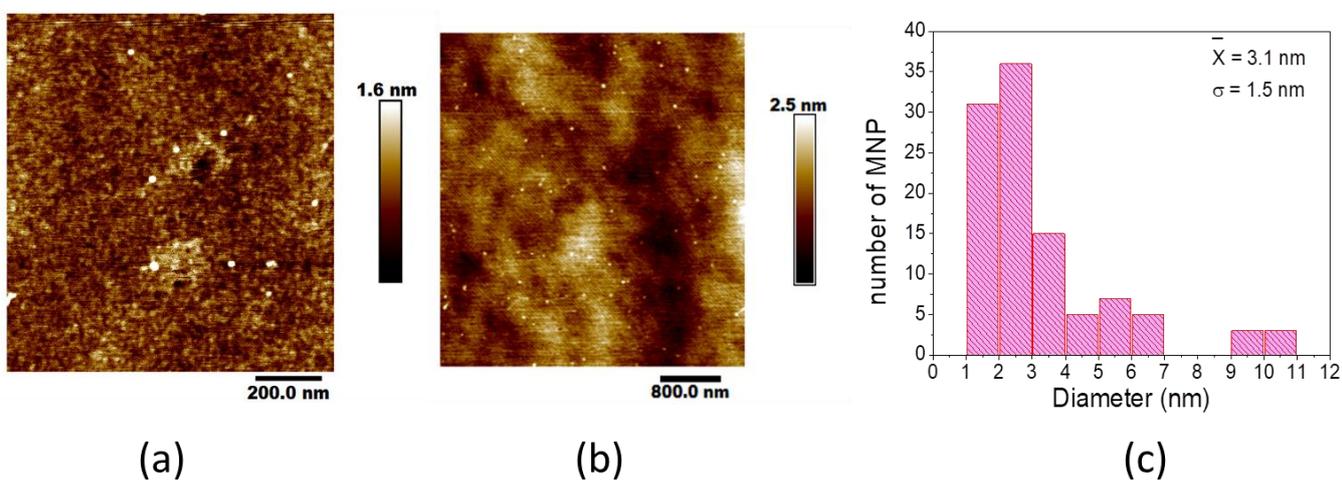


Figure S6 - SEM images of hydrogels Gel/Alg in the (A) absence and (B) presence of caffeine.

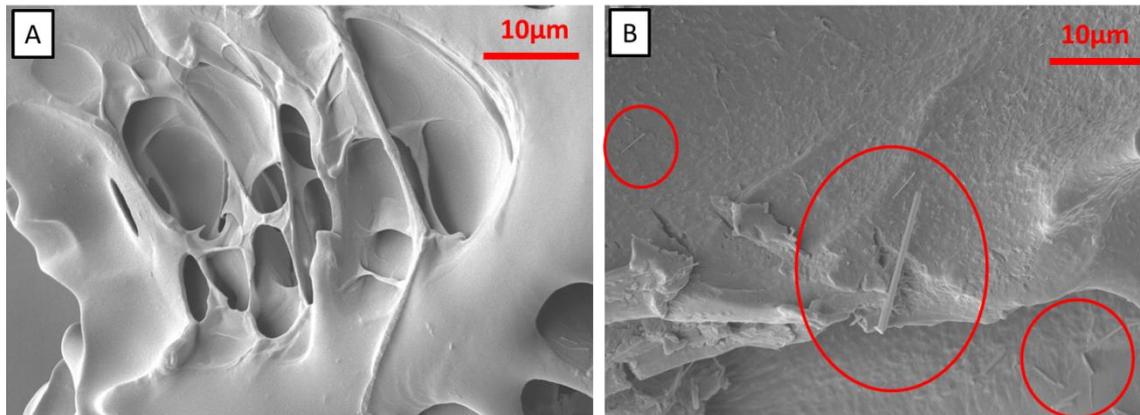


Figure S7 - Transmission FTIR spectra of Gel/Alg and Gel/Alg/MNP films with pure gelatin (Gel), and pure alginate (Alg) in the region of (a) (b) 400- 4000 cm^{-1} , and (c) (d) comparison of the FTIR spectra of the hydrogels in the presence or absence of caffeine in the region of 500-900 cm^{-1} .

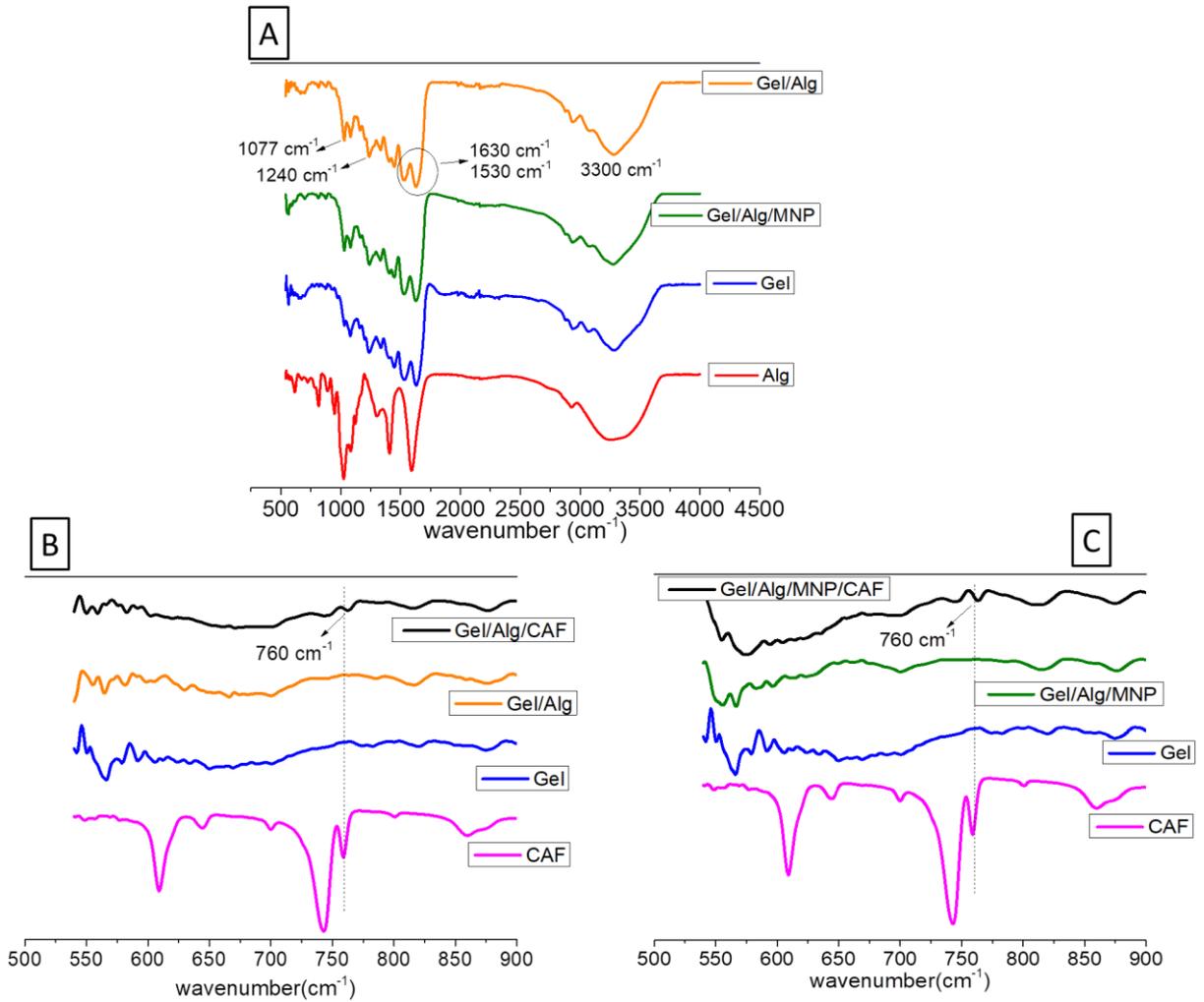


Figure S8 - Typical stress-strain curves determined for dried Gel/Alg, Gel/Alg/CAF, Gel/Alg/MNP and Gel/Alg/MNP/CAF hydrogels. The Young modulus (E) values were calculated as the slope of the initial region (dash lines).

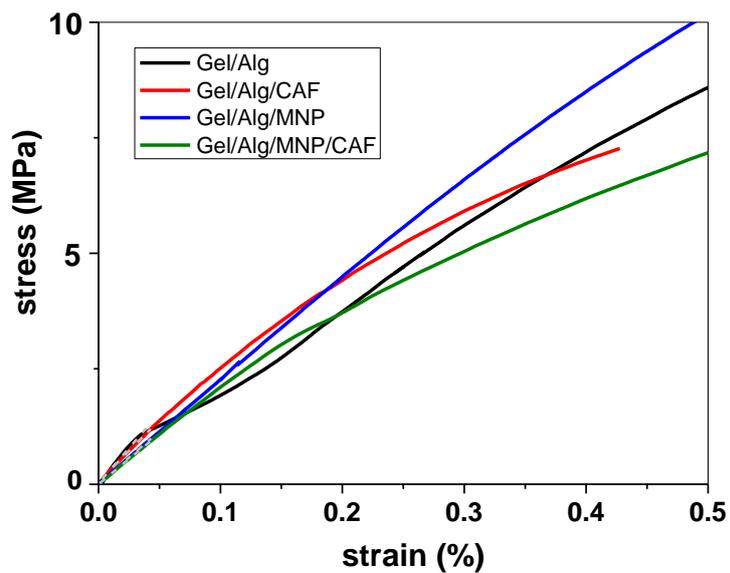


Figure S9 - Thermogravimetric curves and the corresponding 1st derivatives determined for (a) Gel/Alg, (b) Gel/Alg/CAF, (c) Gel/Alg/MNP and (d) Gel/Alg/MNP/CAF.

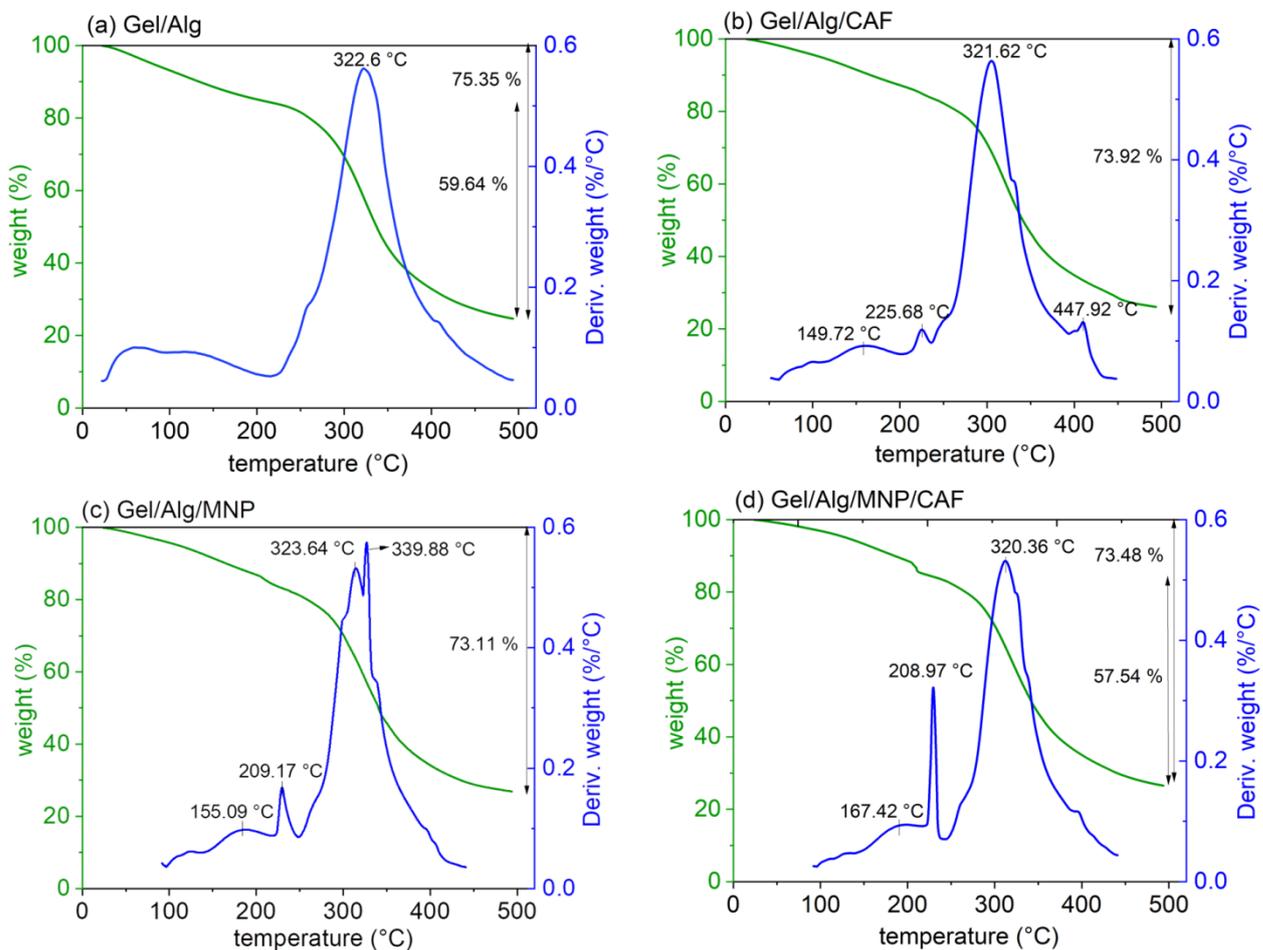


Figure S10 - (a) OCP measurements recorded in 0.1 mol L⁻¹ KCl solution using the ISME-Ca²⁺ sensor in the 0.1 μmol L⁻¹ to 0.5 mol L⁻¹ CaCl₂ concentration range before (black line) and after (red line) Ca²⁺ ions diffusion experiments using the 2-compartment cell. Inset: Calibration plots (E vs pCa²⁺). (b) OCP measurements recorded using the ISME-Ca²⁺ sensor in 0.1 mol L⁻¹ KCl solution during the addition of 1 mmol L⁻¹ CAF (red line) and 1 mmol L⁻¹ Ca²⁺ + 1 mmol L⁻¹ CAF (blue line) solutions, as indicated by the arrows.

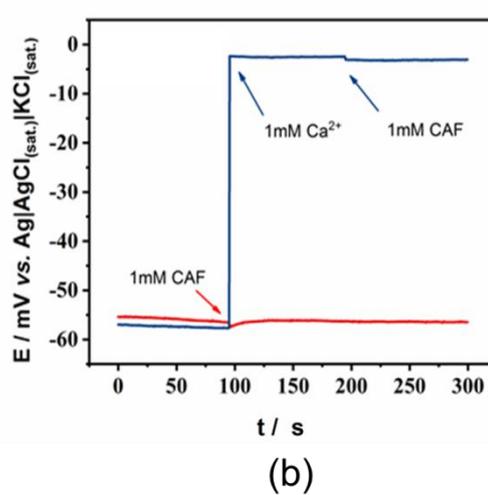
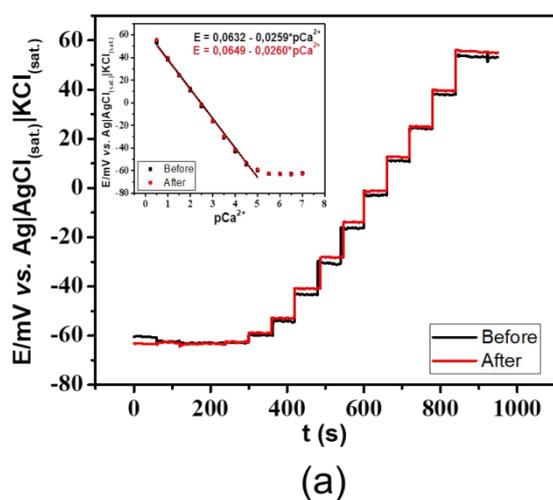


Figure S11 - Cumulative release of caffeine from the hydrogels in the absence (green and red symbols) and presence (blue) of MNP and EMF at (a) pH 5.5 and 25°C, (b) pH 5.5 and 37°C, (c) PBS buffer pH 7.4 and 37°C, (d) PBS/CaCl₂ (0.002 mol L⁻¹) and 37°C, and (e) CaCl₂ 0.5 mol L⁻¹ and 25°C. Solid lines are guides for the eyes. Solid lines were obtained by curve fitting.

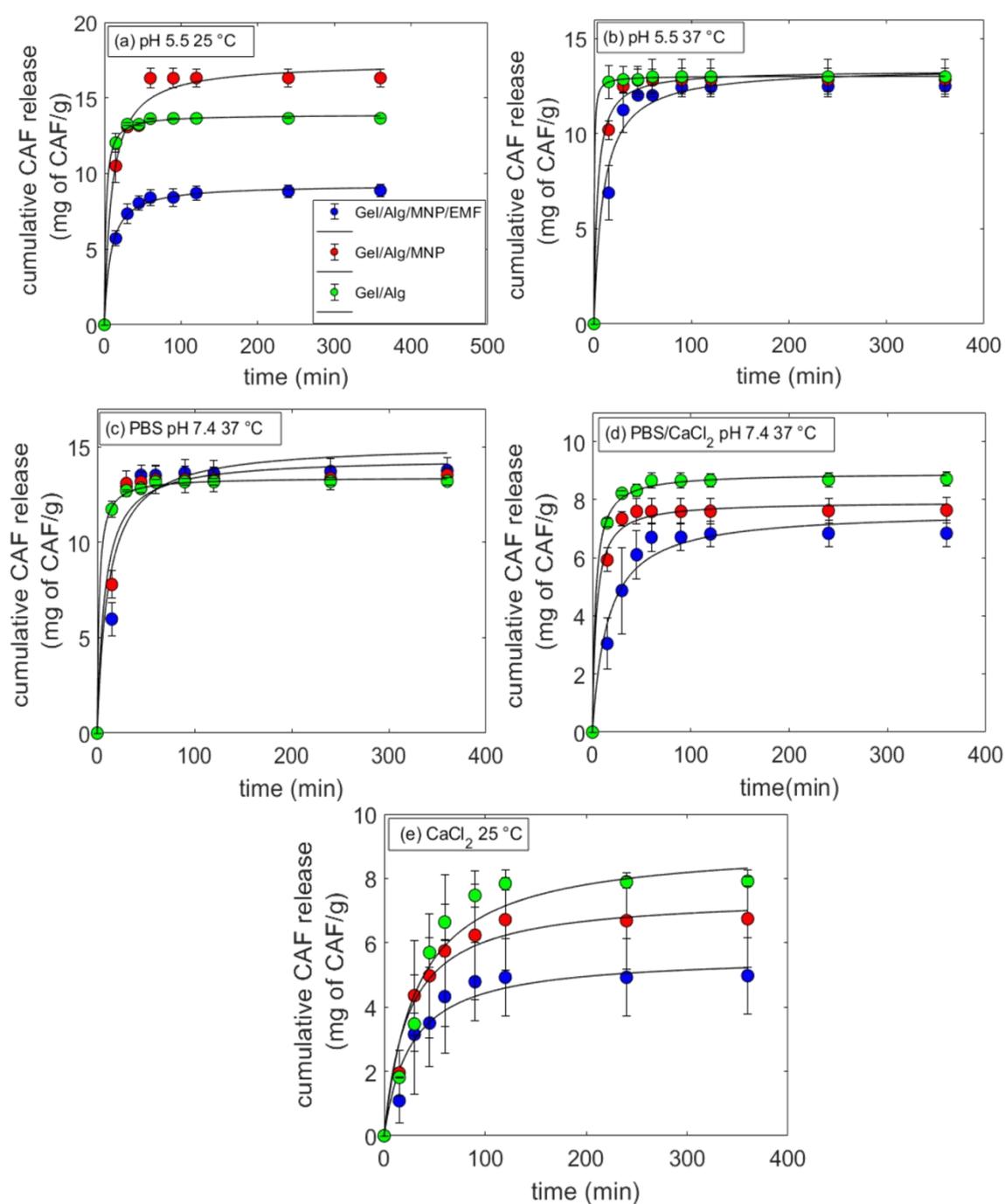


Figure S12 - Cumulative CAF release as a function of square root of time ($t^{0.5}$) from hydrogels in the absence and presence of EMF at (a) pH 5.5 and 25 °C, (b) pH 5.5 and 37 °C, (c) PBS buffer pH 7.4 and 37 °C, (d) PBS/CaCl₂ (0.002 mol L⁻¹) and 37 °C, and (e) CaCl₂ 0.5 mol L⁻¹ and 25 °C. The solid lines correspond to the fittings to the Higuchi model.

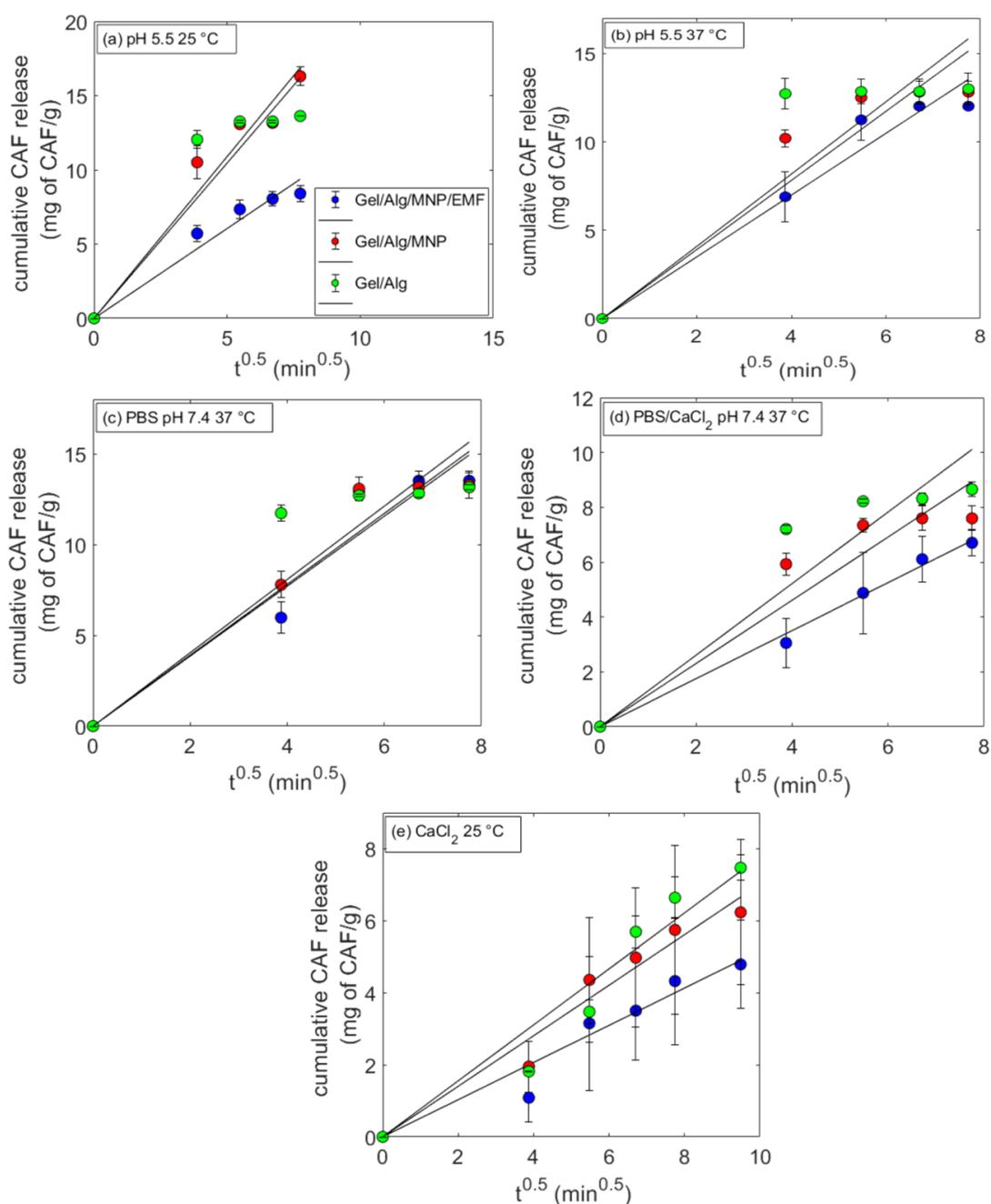


Figure S13 - Statistical analysis for k_{KP} and n values obtained from the Korsmeyer-Peppas fitting curves (**Figure S11**). All data were analyzed by ANOVA (One-way analysis of variance) to assess differences among Gel/Alg, Gel/Alg/MNP and Gel/Alg/MNP/EMF. Results are expressed mean \pm SD and their p values (* $p < 0.05$, ** $p < 0.005$ and *** $p < 0.0005$).

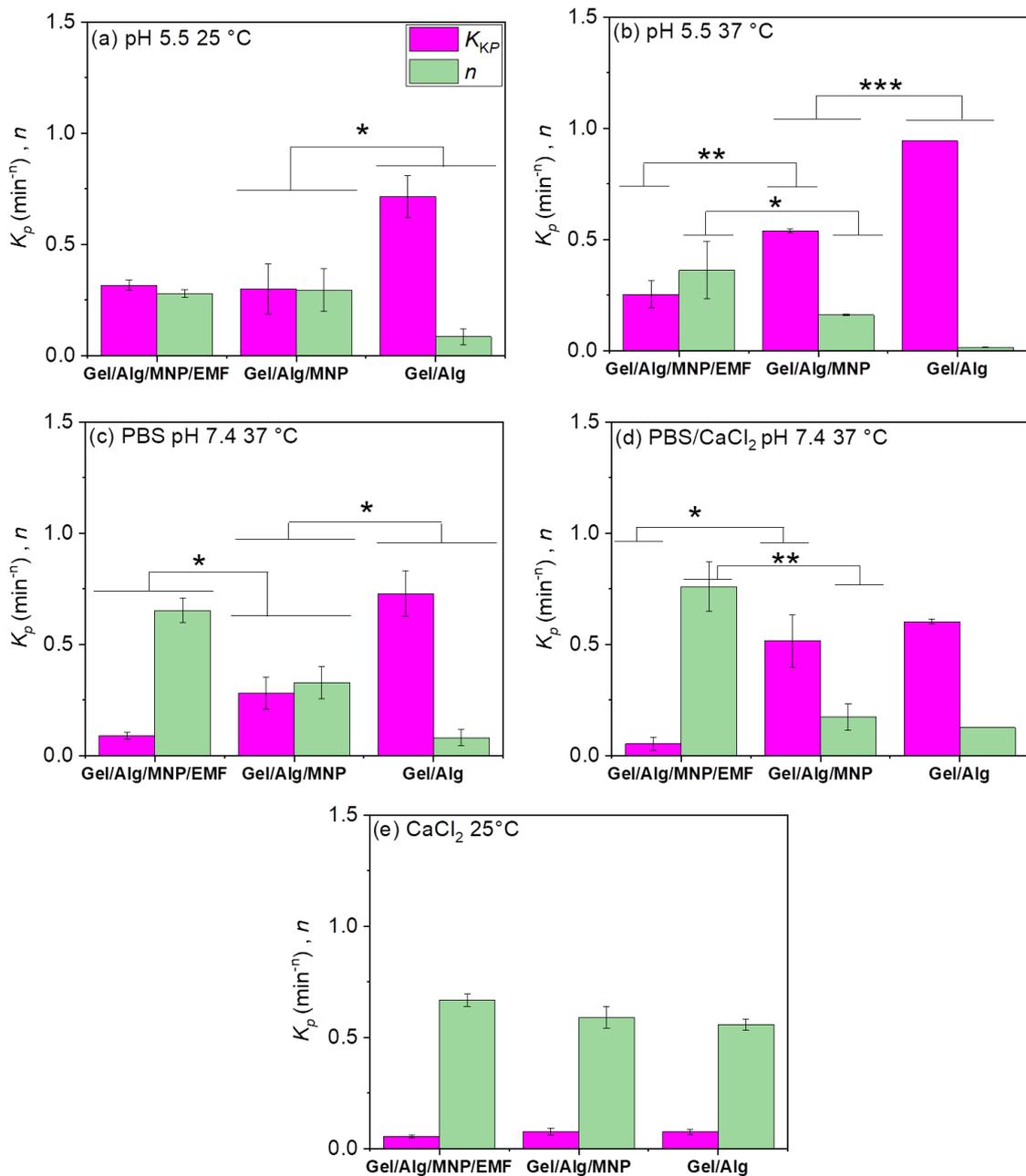


Figure S14 – Swelling degree values for Gel/Alg and Gel/Alg/MNP obtained in Milli-Q water (pH 5.5) and 25 °C, CaCl₂ (0.5 mol L⁻¹) and 25 °C, Milli-Q water and 37 °C, PBS (pH 7.4) and 37 °C.

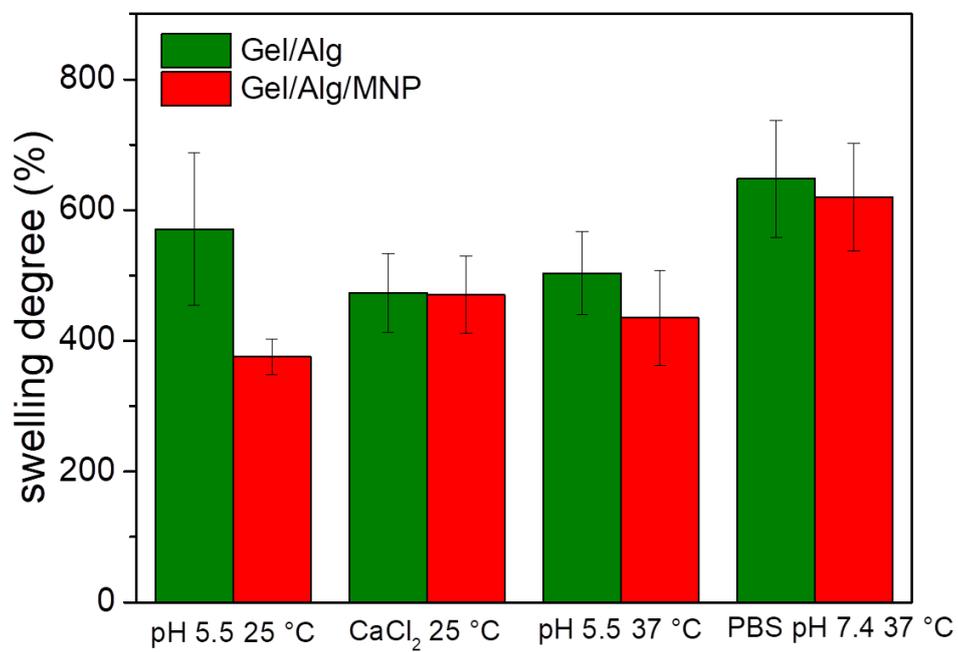


Figure S15 – Optical microscopy images of SH-SY5Y cells treated with CAF in the concentration range from 0.2 mM to 100 mM for 24 h. The scale bar corresponds to 400 μm .

