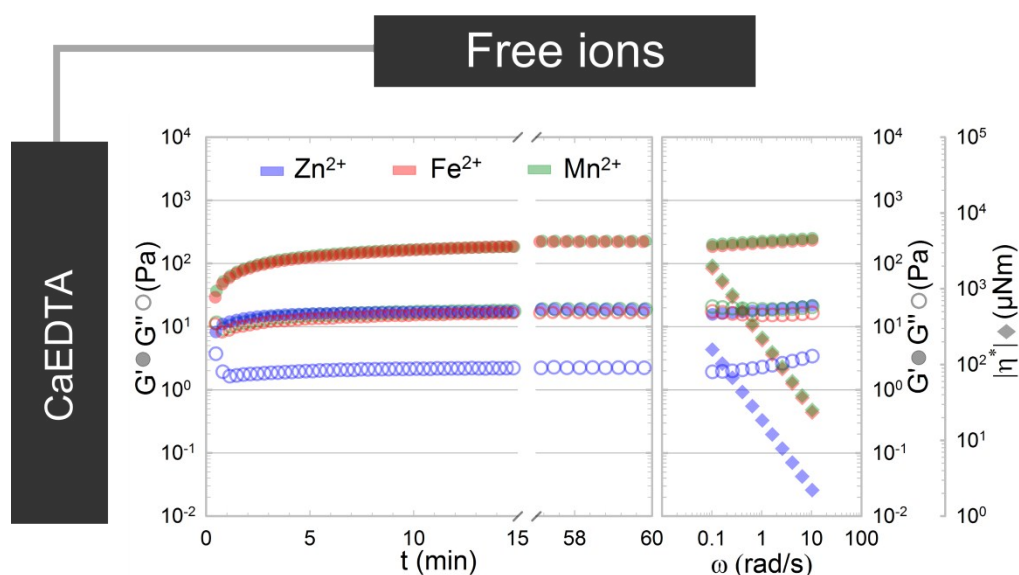


Competitive ligand exchange of crosslinking ions for ionotropic hydrogel formation

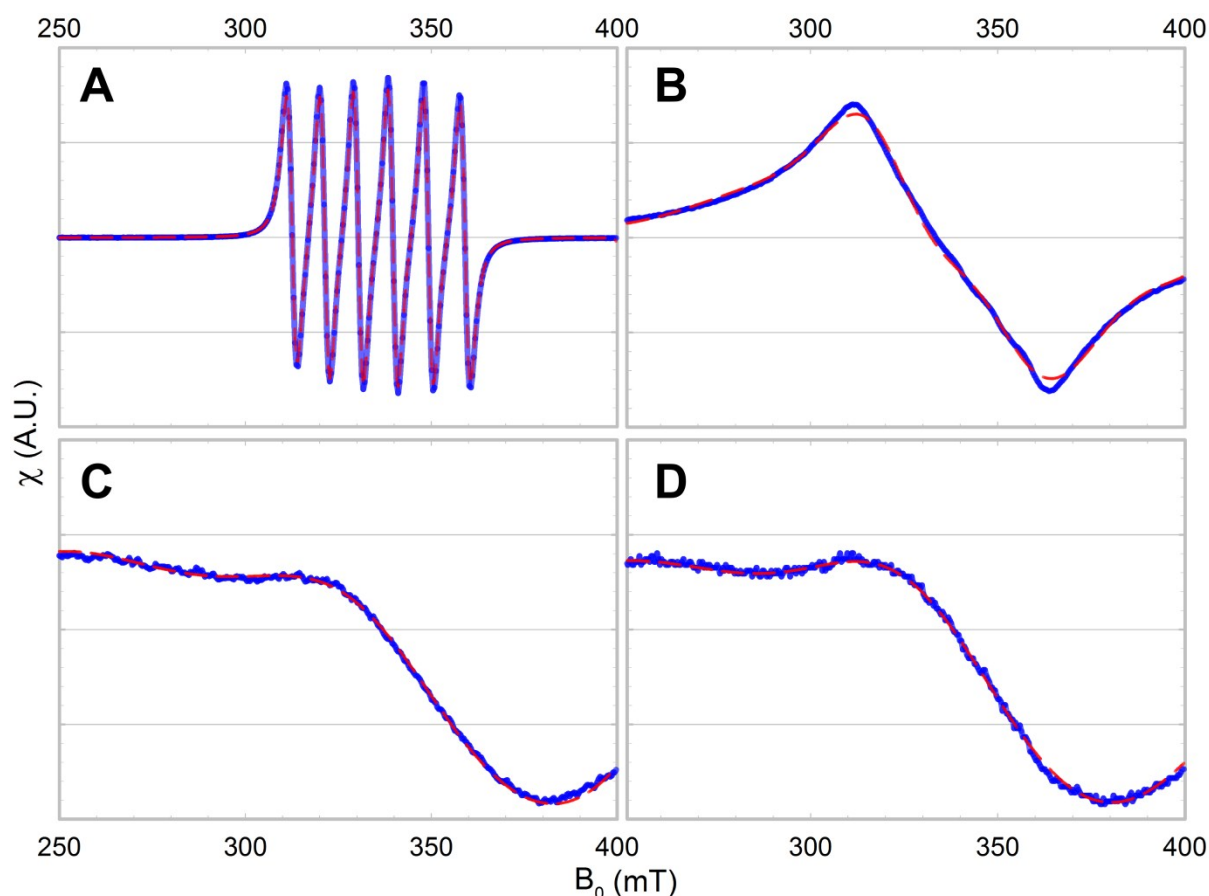
D. C. Bassett, A. G. Håti, T. B. Melø, B. T. Stokke and P. Sikorski

Biophysics and Medical Technology, Department of Physics, Norwegian University of Science and Technology, Trondheim, NO-7491, Norway

Electronic Supplementary Information



ESI Figure 1: Rheological characterization of the temporal change in mechanical properties and frequency dependence of the rheological properties for 1% alginate hydrogels formed via CLEX using EDTA as the CIC and unchelated EI, (Zn^{2+} , Mn^{2+} and Fe^{2+}) at concentrations of 9, 12 and 12mM respectively.



ESI Figure 2: Electron paramagnetic resonance (EPR) characterisation of **A:** MnCl_2 , **B:** Mn-EDDA at a molar ratio of 1:20, **C:** Mn-EDTA at a molar ratio of 1:1 and **D:** CLEX formed alginate hydrogel using Ca-EDTA and Mn-EDDA at 48 hours reaction time. Blue line shows experimental data and red line shows data fitted using EasySpin.

Supplementary discussion of EPR results

It was initially found that some Mn^{2+} ions were not fully chelated (unchelated Mn^{2+} spectra shown in **ESI Fig 2A**) when mixed at a stoichiometric ratio with EDDA (main article **Fig 2C**). In presence of excess EDDA (**ESI Fig 2B** and main article **Fig 2F**), the contribution of unchelated Mn^{2+} to the EPR spectrum vanished (see Reed *et al.*¹). Since gelation in a mixture of alginate and 1:1 Mn-EDDA was not observed, and since there is little correlation with the control spectrum of Mn-alginate (c.f. main article **Fig 2 E**), we speculate that the amount of free Mn^{2+} is below the critical concentration of Mn^{2+} -induced alginate gelation (~ 12 mM for the alginate in question) in a solution with an otherwise relatively high concentration of Mn^{2+} (30 mM). The majority of the Mn^{2+} are therefore likely chelated by the EDDA and the clear presence of free Mn^{2+} is due to a much stronger signal from Mn^{2+} when only water is present as a weakly bound ligand.

ESI Fig 2C and **D** show the similarity in EPR spectra recorded for Mn-EDTA and CLEX formed hydrogel using Ca-EDTA and Mn-EDDA at 48 hours reaction time respectively. This is clear evidence that the final ligand associated with Mn^{2+} was EDTA for this reaction.

ESI Table 1: Calculated EPR tensor and time correlation values of the experimental conditions shown in Supplementary figure 2. Calculated EPR values confirm the similarity of Mn^{2+} ligand found in completed CLEX reaction to Mn-EDTA control samples

Sample	$g_{\parallel}$	$g_{\perp}$	$A_{\parallel}$	$A_{\perp}$	$\log(\tau_{\text{corr}})$
MnCl_2	2.02	1.97	990	-1198	-12.43
MnEDDA	1.95	2.11	-330	713	-9.67
MnEDTA	1.80	2.61	-237	-213	-8.72
CLEX 48 hours	1.71	2.73	-252	-54	-9.74

ESI video 1: Gelation of 1% alginate using 50mM CaCl_2 . Food colourant has been added to the CaCl_2 solution.

ESI video 2: Gelation of 1% alginate using CLEX (Ca-EDTA and ZnEDDA at pH 7.0). Food colourant has been added to the CaEDTA solution

ESI video 3: Gelation of 1% alginate using CLEX (Ca-EDTA and ZnEDDA at pH 7.8). Food colourant has been added to the CaEDTA solution.

ESI references

1. G. H. Reed, et al., *J. Chem. Phys.*, 1971, **55**, 3311-3316.