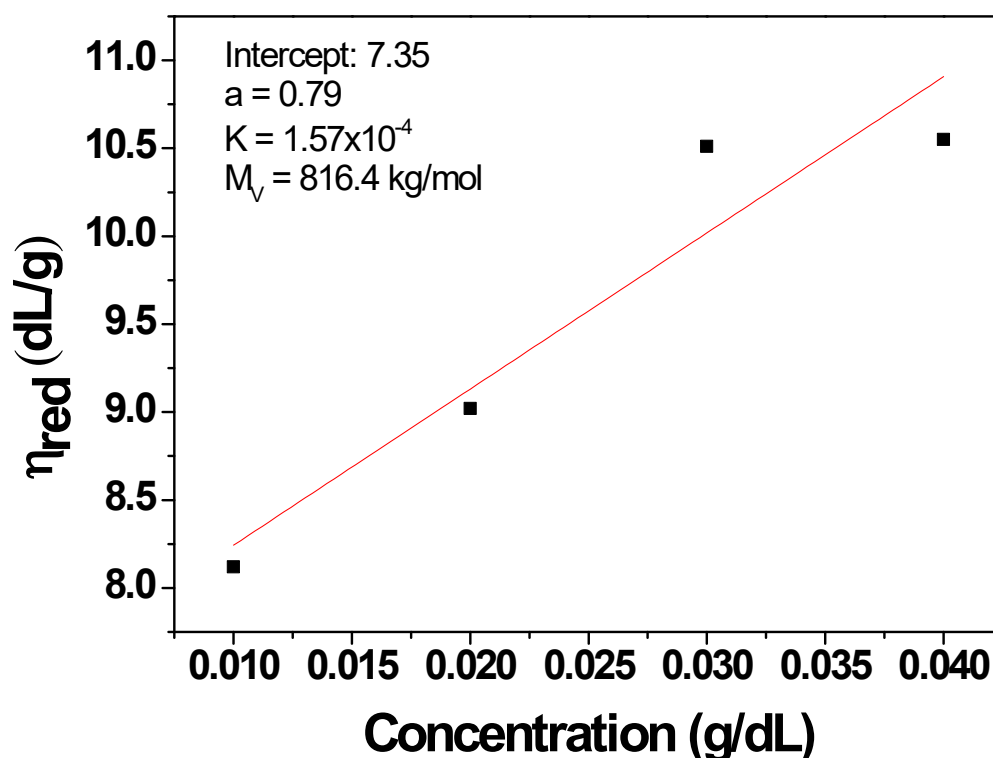
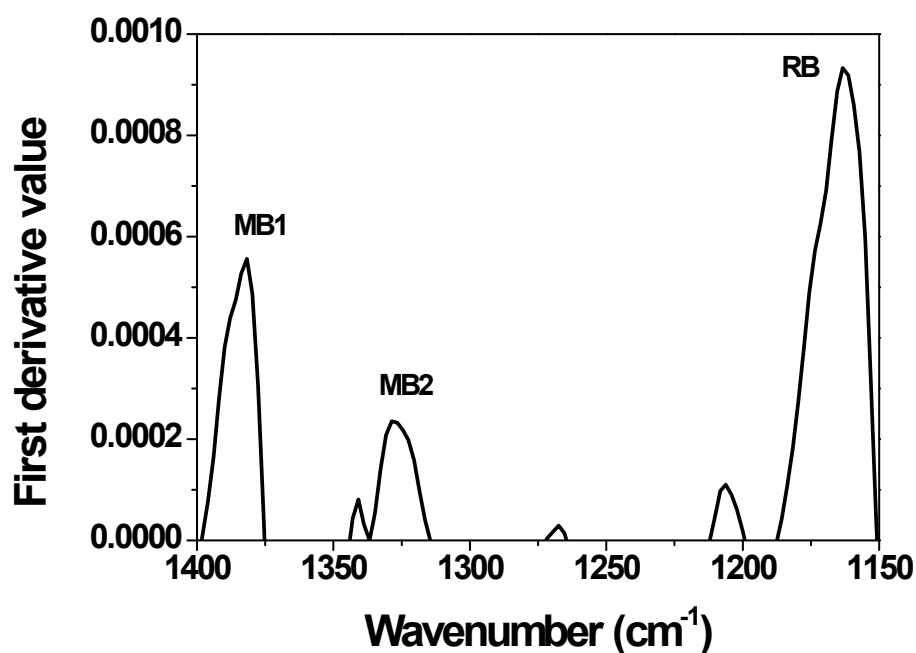


SUPPLEMENTARY MATERIAL

SM1 - The viscometric average molar mass ($\overline{M}_v$) was determined from the empirical equation of Mark-Houwink-Sakurada using a Ubbelohde viscometer (Schott, Germany) with internal diameter of 0.63 mm and constant of 0.01 mm².s⁻², which was coupled to an automatic AVS 350 Schott (Schott, Germany) equipment. Chitosan solutions were prepared at concentrations of 10, 20, 30, and 40 mg/dL by dilution in NaAc/HAc pH 4.5 buffer (0.25M/0.25M)¹. The flow times of chitosan and buffer solutions were measured in triplicate to determine the relative viscosity at 23 ± 1 °C.



SM2 - The degree of deacetylation (DD) of the chitosan sample was estimated at 25 °C by infrared vibrational spectroscopy in the attenuated total reflectance (FTIR-ATR) using an Alpha FTIR-ATR equipment (Bruker, Germany) and a diamond crystal. The spectra were recorded with an accumulation of 64 scans at a resolution of 2 cm⁻¹. The determination of the DD using infrared techniques might suffer from the strong influence of air humidity, which hinders the amide I band at 1655 cm⁻¹. Therefore, different strategies have been used to circumvent the problem of the presence of water in determining the DD of chitosan samples. Here, we applied the method proposed by Beil *et al*². In this method, the first derivative of the transmittance infrared spectrum and three main bands are used to calculate the degree of acetylation: a band at 1163 cm⁻¹ used as an internal standard (RB) referring to the vibration of the oxygen bridge of the glycosidic bond and the amide III bands at 1327 cm⁻¹ (MB2) and the CH deformation of the N-acetyl group at 1383 cm⁻¹ (MB1). The FTIR-ATR spectra for CHI and its first derivative spectra are presented below. The DD was estimated as ~ 76%.



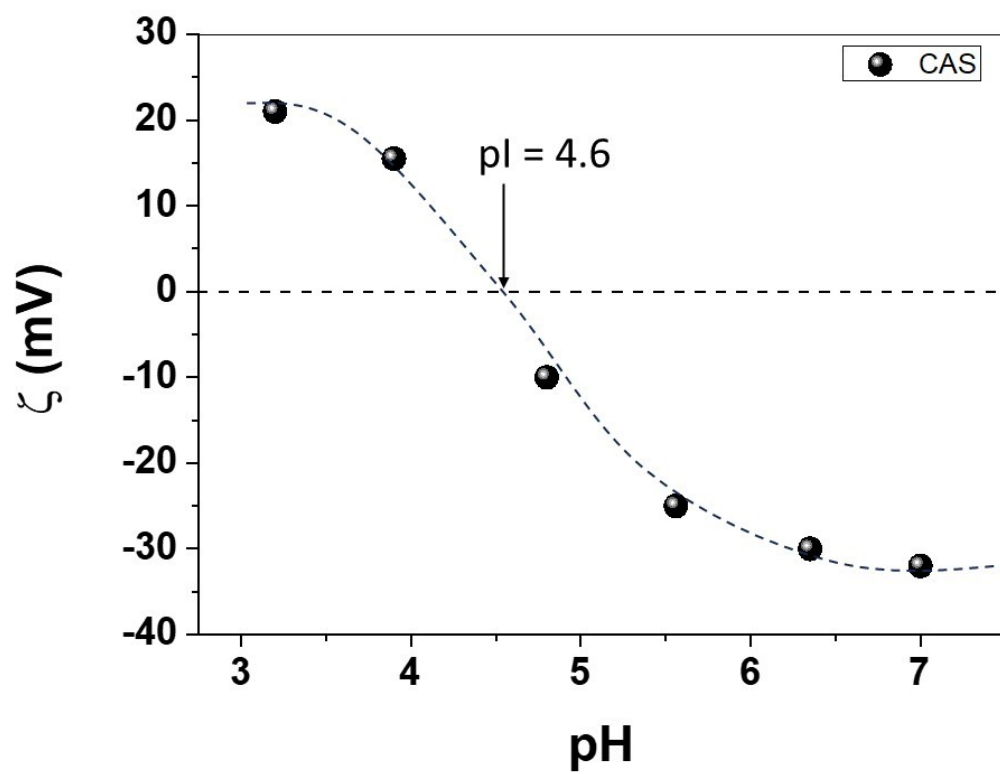
Designation	Wavenumber (cm ⁻¹)	Value
MB1	1383	5.6e-4
MB2	1327	2.4e-4
RB	1163	9.3e-4

$$(MB1 + MB2 / RB) = 0.0157 \text{ DA} + 0.487$$

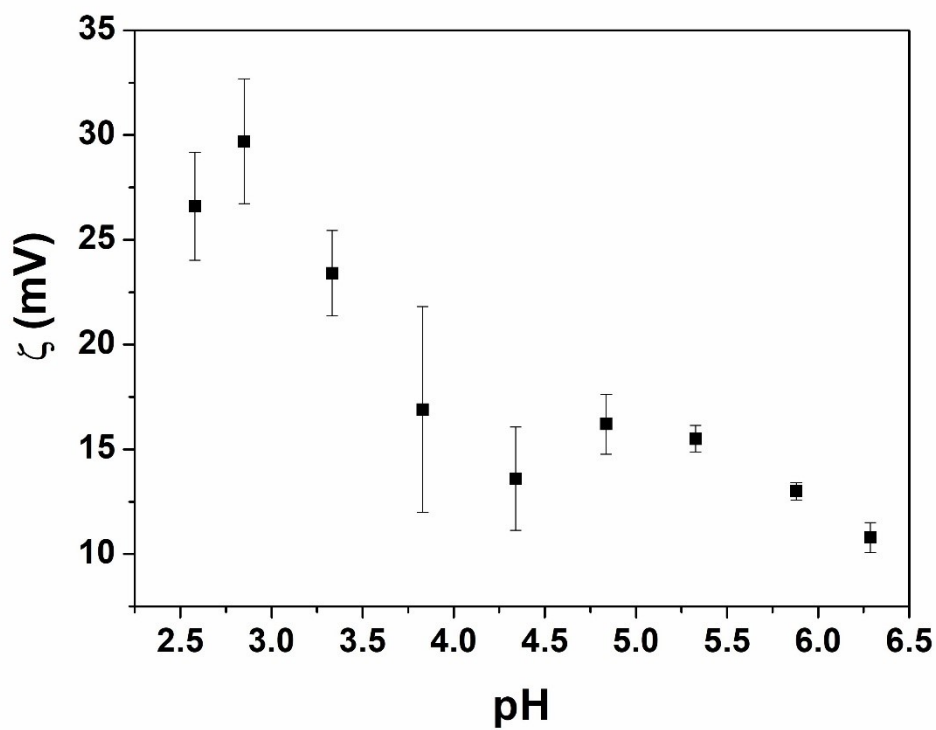
$$\text{DA} = 22.77\%$$

$$\text{DD} = 76.23\%$$

SM3 – Isoelectric titration of (a) CAS that estimated the isoelectric point at pH 4.6, and (b) CHI.



(a)

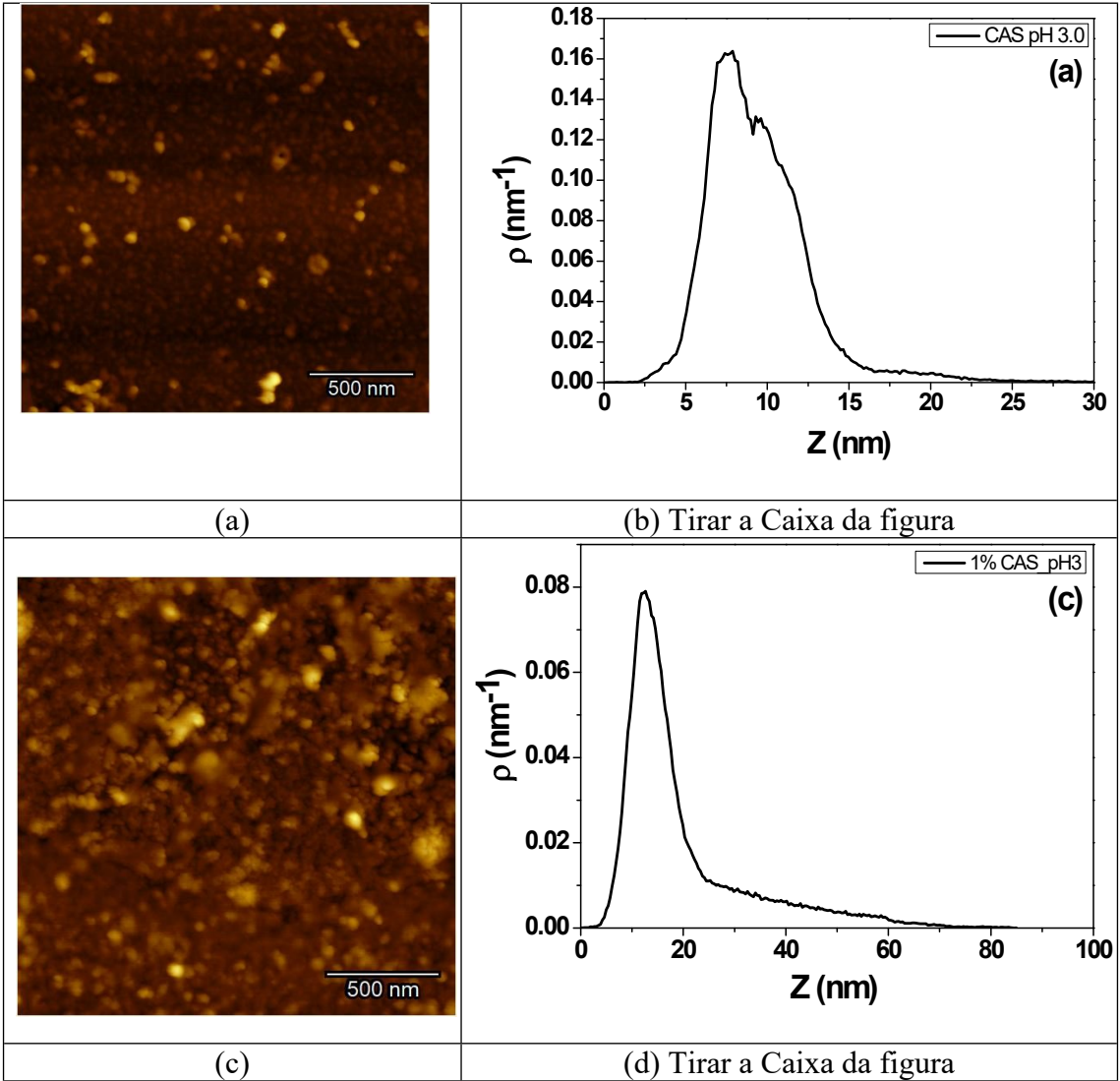


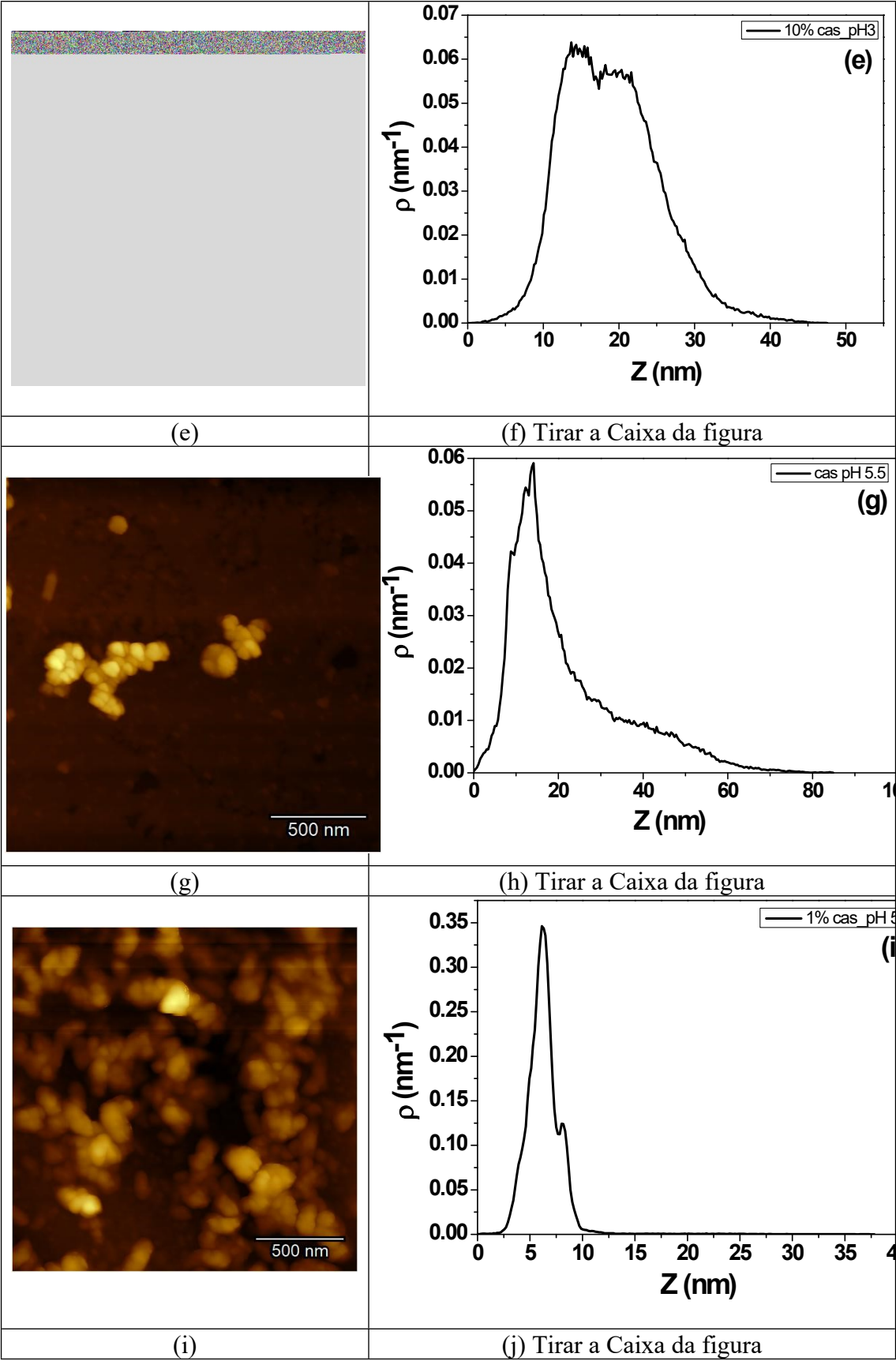
(b)

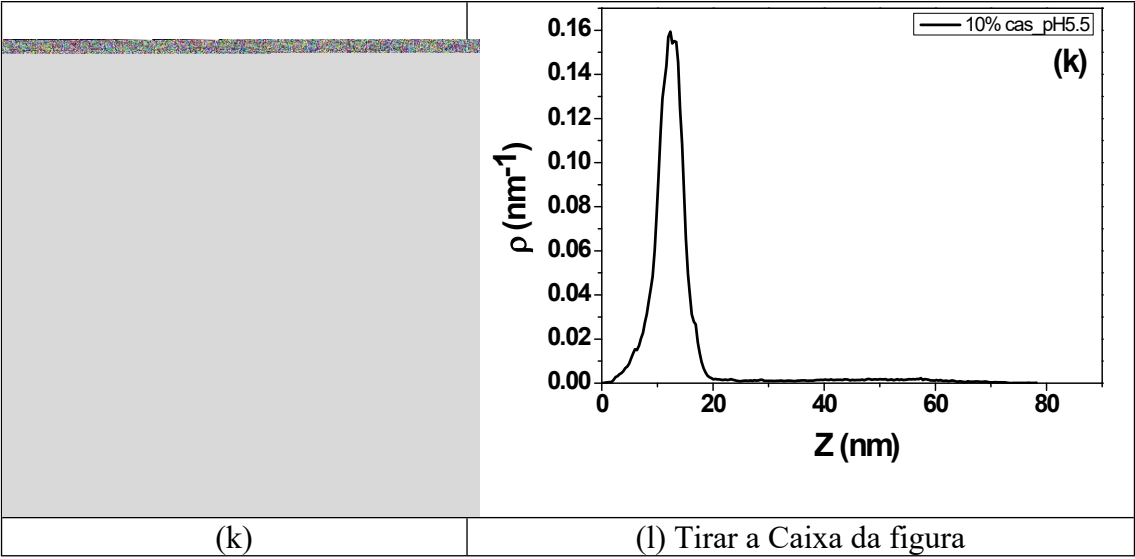
SM4 –AFM topographical images (2 μm x 2 μm) and their respective statistical distribution of pixel heights across the analyzed surface. The Probability Density Function (PDF) describes the likelihood of encountering a particular height value on the surface. Unlike a simple count, the PDF indicates the density of probability per unit height interval. A higher value on the y-axis at a given height signifies a greater concentration of pixels within that specific height range, while the x-axis indicates the pixel height (Z) in nanometers (nm). To obtain the actual probability (or fraction of the surface area) for a given height range, the PDF value must be integrated over that range. The total area under this curve sums to 1, representing 100% of the surface pixels.

At pH 3.0, AFM images of CAS:CHI (a) 100:0, and (b) the corresponding PDF; (c) 100:0.24, and (d) the corresponding PDF; (e) 100:1.27 and (f) the corresponding PDF.

At pH 5.5 AFM images of CAS:CHI (g) 100:0, and (h) the corresponding PDF; (i) 100:0.24, and (j) the corresponding PDF; (k) 100:1.27 and (l) the corresponding PDF.



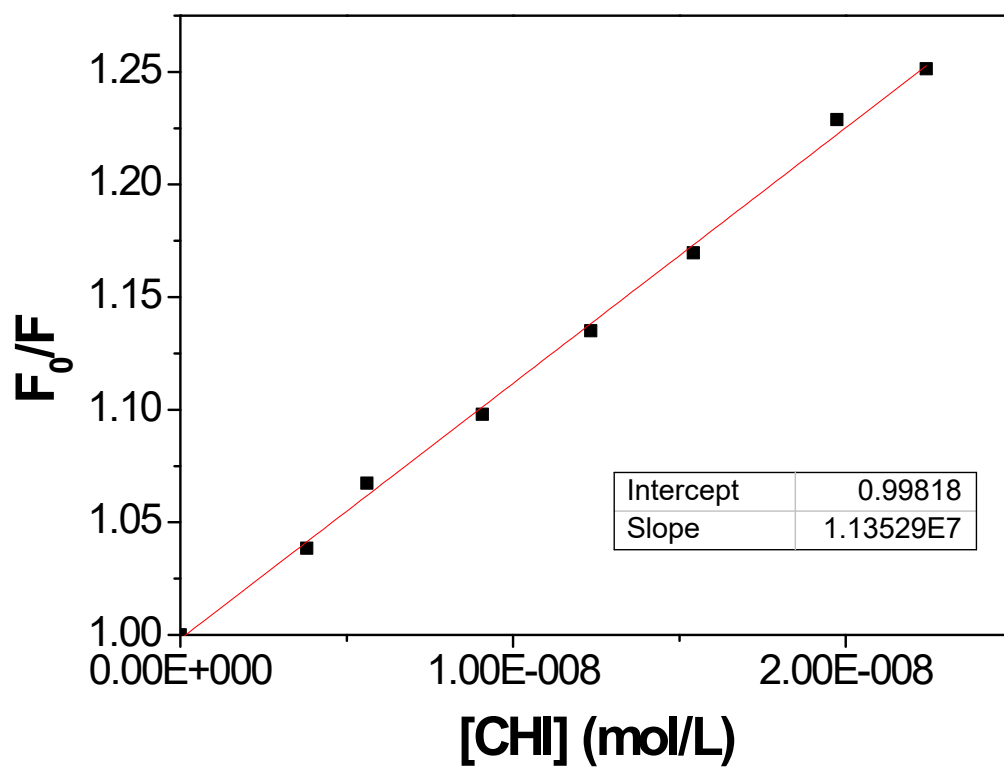




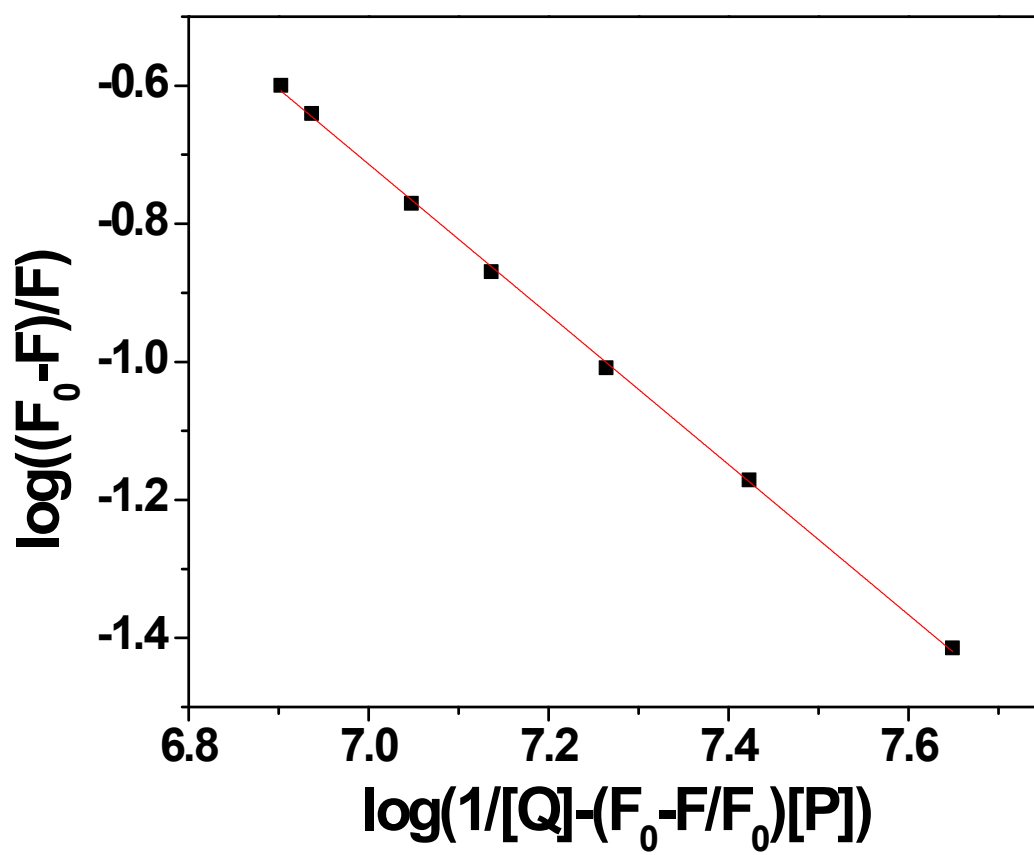
SM5 - Zeta potential, mean size, polydispersity index values determined for different CAS:CHI systems at pH 4.6, in the absence and in the presence of NaCl.

System	ζ (mV)	Size (nm)	PDI
CAS:CHI (100:1.27) pH4.6 (0 mM NaCl)	27.1 ± 0.6	288.6 ± 3.2	0.211 ± 0.015
CAS:CHI (100:1.27) pH4.6 (1 mM NaCl)	9.46 ± 0.4	178.2 ± 2.8	0.184 ± 0.020
CAS:CHI (100:1.27) pH4.6 (10 mM NaCl)	29.4 ± 3.5	636.6 ± 27.6	0.417 ± 0.011
CAS:CHI (100:1.27) pH4.6 (100 mM NaCl)	15.2 ± 1.2	2596 ± 198	0.221 ± 0.039

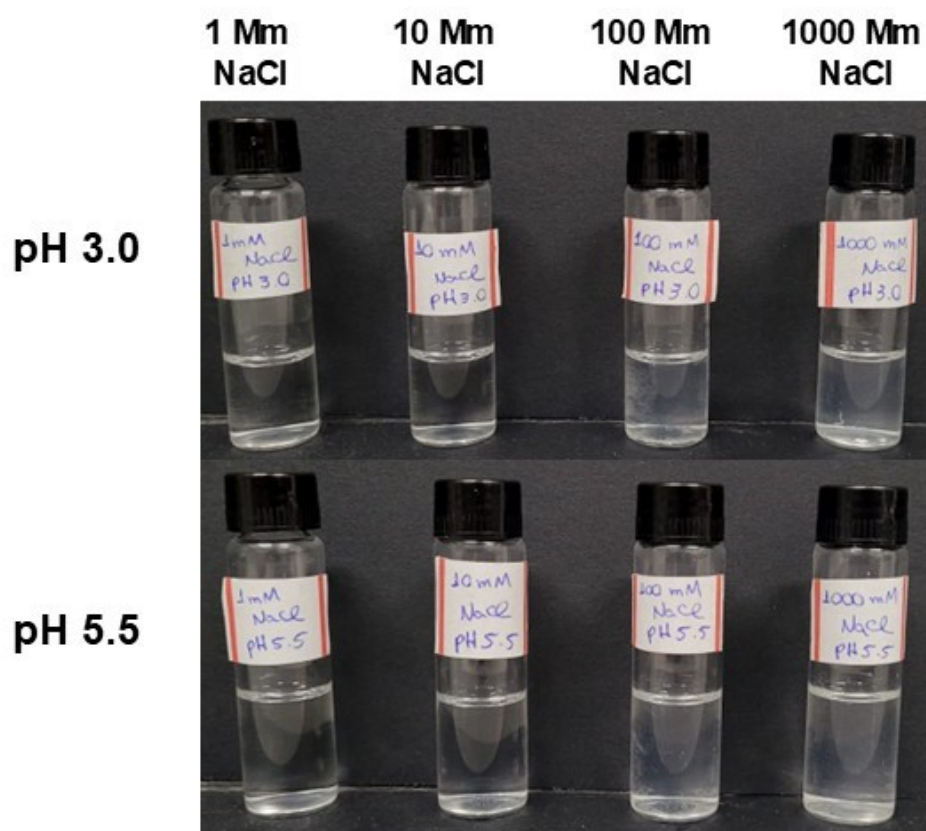
SM6 - Stern–Volmer plot (eq. 3) of the interaction between CAS and CHI at pH 3.0 and 296 K. The slope of the linear fitting yielded the K_{sv} value of $1.13 \times 10^7 \text{ L mol}^{-1}$.



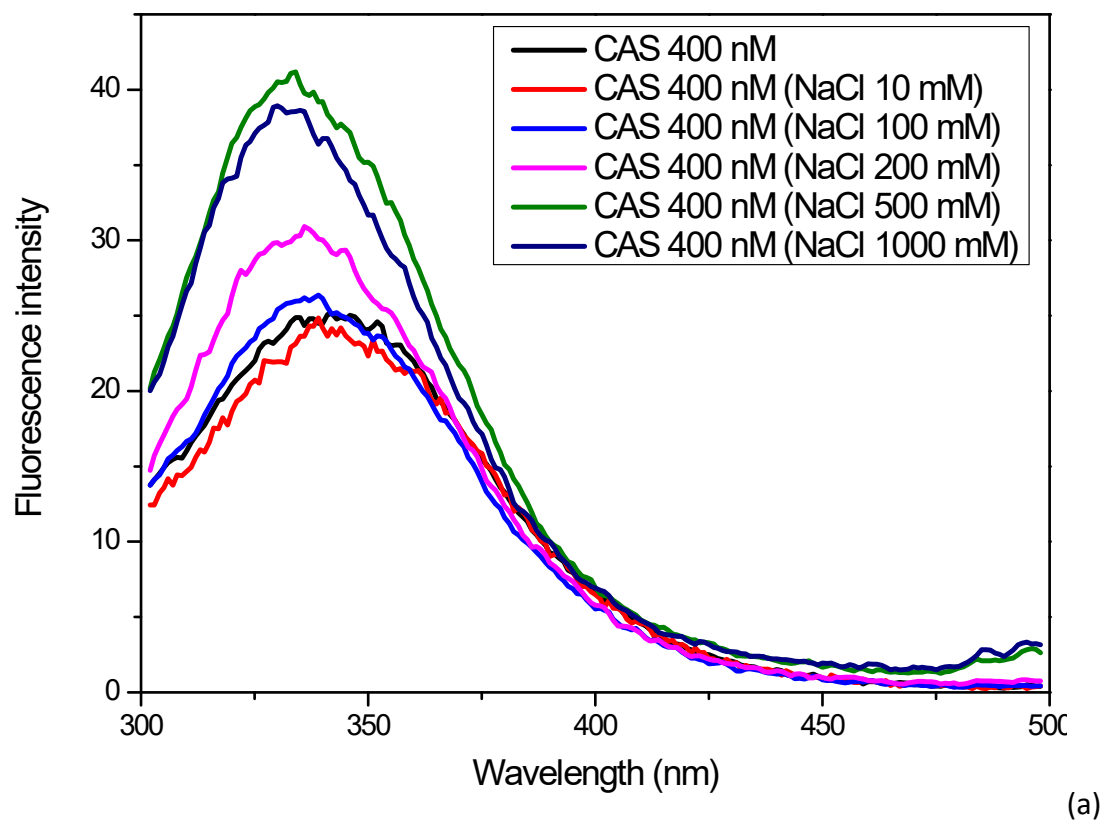
SM7 – Modified Stern–Volmer plot (eq. 4) of CAS-CHI at pH 3 and 296 K.

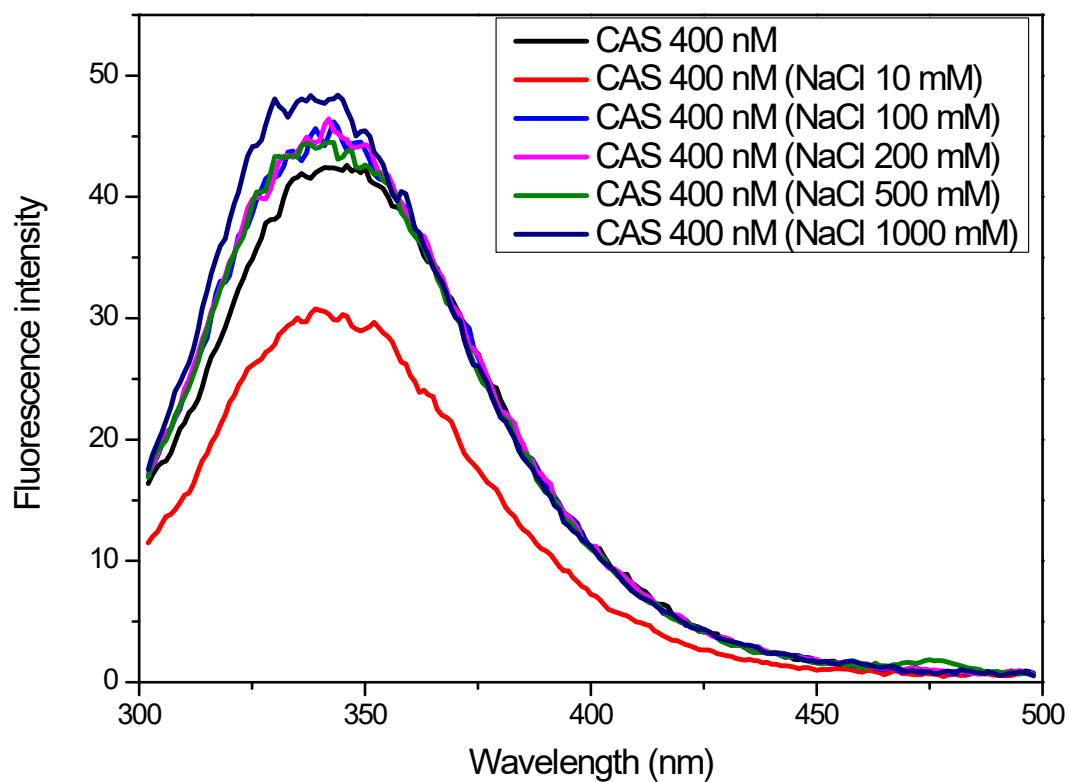


SM8 –CAS:CHI 100 nM:5.5 nM mixtures at pH 3.0 and 5.5 in the presence of 1 mM, 10 mM, 100 mM, and 1000 mM NaCl.



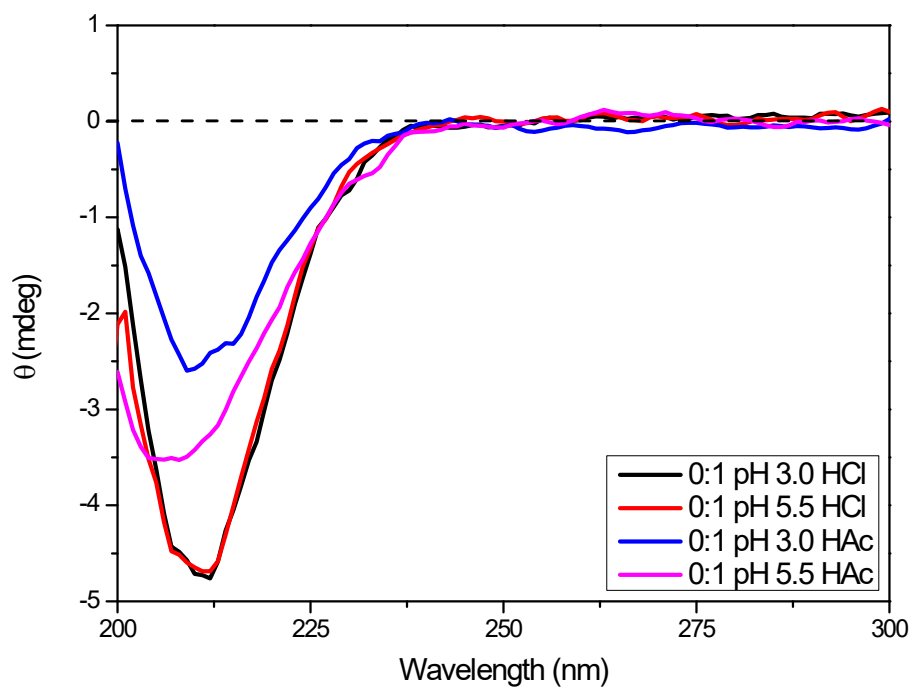
SM9 – Fluorescence of 400 nM CAS at pH 3.0 (a) and pH 5.5 (b) with different NaCl concentrations. $\lambda_{\text{exc}} = 275$ nm. Font size





(b)

SM10 - Circular dichroism spectra measured for CHI dissolved in HCl (pH 3.0 and pH 5.5) and acetic acid (HAc, pH 3.0 and pH 5.5). Font size



	(%)								Tur n	Random
	Helix		AntiP				Parallel			
Component	Helix1	Helix2	Tota 1	Anti1	Anti2	Anti3	Tota 1			
CAS:CHI (0:1) pH 3.0 in HCl	4.5	3.4	7.9	1.9	16.7	16.9	35.4	1.9	12.4	42.3
CAS:CHI (0:1) pH 5.5 in HCl	4.0	3.2	7.2	0.4	14.0	18.4	32.8	3.2	13.0	43.8
CAS:CHI (0:1) pH 3.0 in HAc	5.7	6.6	12.4	2	13.6	15.2	30.9	3.3	12.9	40.5
CAS:CHI (0:1) pH 5.5 in HAc	3,3	7,5	10,8	0	13,8	11,7	25,5	4,8	13,4	45,4

References

- 1 G. Kasaai, M.R., Arul, J., Charlet, *J Polym Sci B Polym Phys*, 2000, 38, 2591-2598.
- 2 S. Beil, A. Schamberger, W. Naumann, S. MacHill and K. H. Van Pée, *Carbohydr Polym*, 2012, 87, 117–122.