Supporting Information

Effect of the supramolecular interactions on the nanostructure of halloysite/biopolymer hybrids: a comprehensive study by SANS, Fluorescence Correlation Spectroscopy and Electric Birefringence.

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Table 1s. Density and scattering length density of solvent, biopolymers and HNT used in the SANS data analysis.

	δ / g cm^{-3}	SLD / 10^{10} cm^{-2}
HNT	2.5316	3.62
D_2O	1.1099	6.37
chitosan	1.401	1.64
alginate	1.601	4.47
HPC	1.170	0.767

Table 2s. Scattering length density of the nanotube's shell for HNT/biopolymer hybrids based on their composition from Guinier analysis for a weight ratio biopolymer/HNT of 0.1.

	SLD / 10^{10} cm^{-2}
HNT/chitosan	3.34
HNT/alginate	3.45
HNT/HPC	3.48

Table 3s. Fitting parameters obtained by the analysis of FCS curves for DTAF/alginate and HNT/DTAF alginate suspensions through the triplet sate model.

	α	G(0)	τ_c	T	τ_T
DTAF/alginate	0.81 ± 0.04	0.119 ± 0.003	1.80 ± 0.29	0.17 ± 0.04	0.05 ± 0.02
HNT/DTAF/alginate	0.72 ± 0.13	0.078 ± 0.007	3.5 ± 1.0	0.36 ± 0.09	0.0048 ± 0.0015

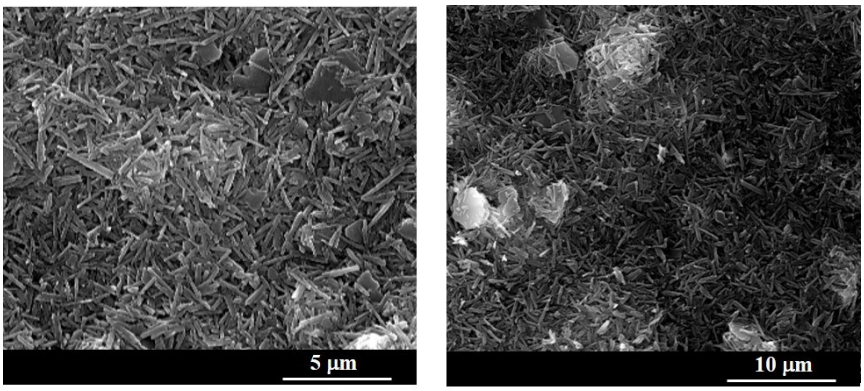

Figure 1s. SEM images of halloysite nanotubes.

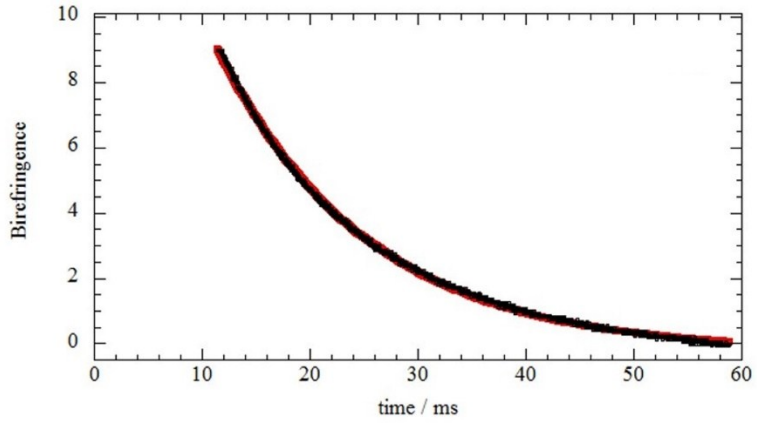

Figure 2s. Decay of the electric birefringence as a function of time after the termination of the voltage for aqueous dispersion of HNT/HPC (1:10).The HNT concentration was 0.15 g dm^{-3}. Red line represents the fit according to the equation $\Delta n = \Delta n_0 \exp(-t/\tau)$, where τ is the characteristic relaxation time of the nanotubes, while Δn_0 represents the birefringence magnitude at the instant of electric field termination.

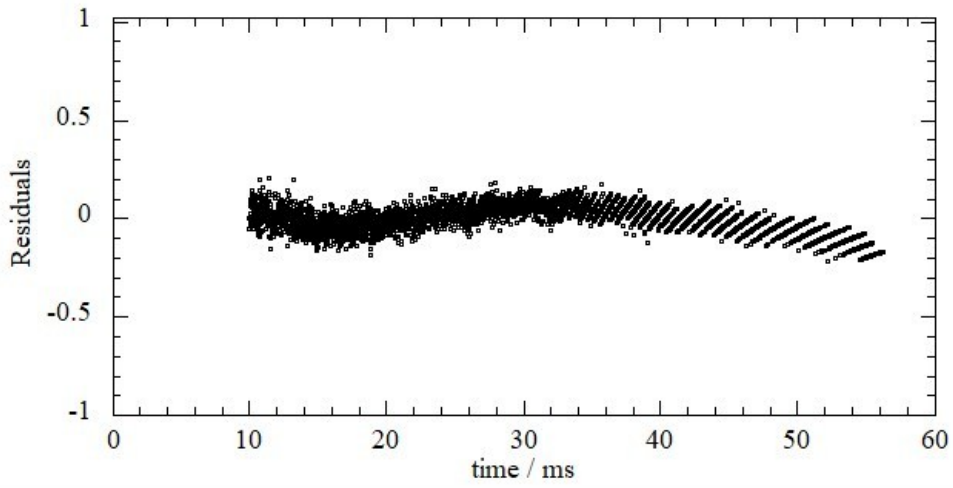

Figure 3s. Residuals for the fit of Δn vs t trend of the aqueous dispersion of HNT/HPC (1:10) reported in Figure 1s. Residuals are calculated as $Res = \Delta n_i - \Delta n_{i,fit}$ with the measured electric birefringence Δn_i and the fitted value $\Delta n_{i,fit}$.

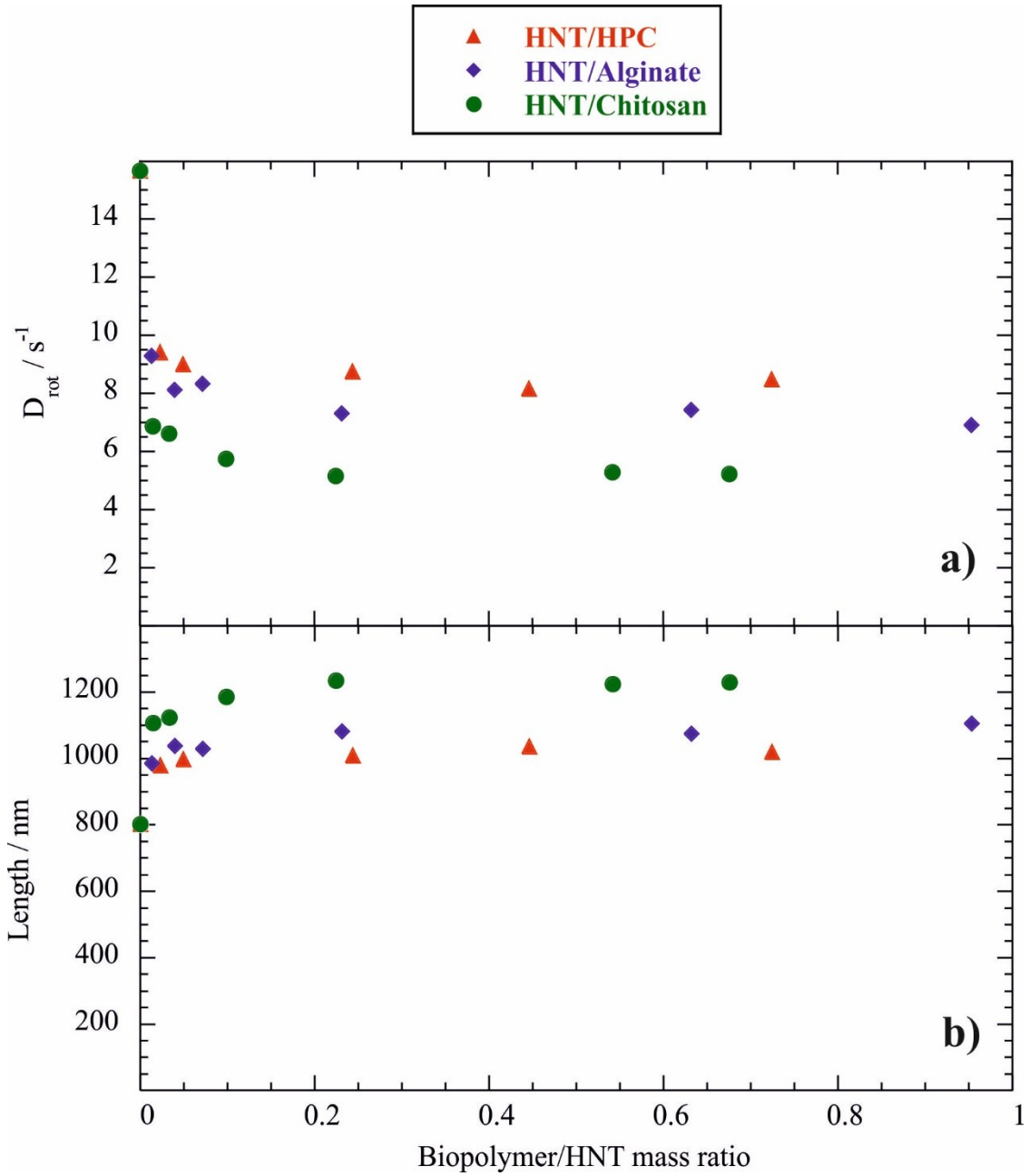

Figure 4s. The rotational diffusion coefficient (a) and the corresponding length (b) determined by the analysis of EBR data as a function of the biopolymer/HNT weight ratio for a HNT concentration of 0.15 g dm^{-3}. The length was calculated by the Broersma theory for rigid thin rods with a radius of 80 nm. Details for the calculation are presented below.

Calculation of the length from rotational diffusion coefficient

According to Broersma theory[47], the theoretical D_{rot} of rigid rods can be calculated as

$$D_{rot} = \frac{3k_BT}{\pi\eta L^3}\left(\ln\left(\frac{L}{d}\right) + \delta\right) \quad (1)$$

$$\text{with } \delta = -0.662 + 0.917\left(\frac{L}{d}\right)^{-1} - 0.050\left(\frac{L}{d}\right)^{-2} \quad (2)$$

where d is the diameter of the rods, η is the viscosity of the solvent, T is the temperature and k_B is the Boltzmann constant.

Equation 1 can be used for rods with $2 \leq \left(\frac{L}{d}\right) \leq 30$. Thus, the Broersma theory is appropriate to study the rotational mobility of cylinders with the average HNT sizes (L = 1000 nm and d = 180 nm)

Fluorescent labeling of biopolymers

Alginate labeling with 5-[(4,6-dichlorotriazin-2-yl)amino]fluorescein (DTAF)

Alginate was fluorescently labeled with DTAF as reported elsewhere.[1,2] Briefly, we prepared a $NaHCO_3$ aqueous solution (concentration = 50 mmol dm^{-3}), which was used as solvent to solubilize alginate through magnetically stirring at 25 °C for 2 h. The alginate concentration was fixed at 10 g dm^{-3}. Once alginate was solubilized, the pH of the polymer solution was adjusted to 9 by adding 1.0 mol dm^{-3} NaOH. On the other hand, we prepared a DTAF solution in dimethyl sulfoxide (concentration = 10 mg cm^{-3}) by magnetically stirring at 25 °C for 2 h. The alginate and DTAF solutions were mixed overnight under magnetically stirring at 25 °C. The alginate/DTAF ratio was set at 1:0.4 v/v. Then, the unbound DTAF was removed by dialyzing the reaction mixture in 10 kDa cutoff dialysis tubing against phosphate-buffered saline (PBS). To this purpose, 20 cm^3 of the alginate/DTAF mixture were dialyzed against 500 cm^3 of PBS solution for 24 hours. The dialysis procedure was repeated three times. According to literature, the average labeling density should be 2.5 moles of DTAF per mole of alginate.[2]

Chitosan labeling with fluorescein isothiocyanate (FITC)

According to literature,[3] FITC was employed as fluorescent probe for chitosan labeling. The covalent attachment of the FITC isothocyanate group with the primary amine groups of chitosan was achieved by using the following procedure. Firstly, chitosan was solubilized in 10 g dm^{-3} acetic acid solution by magnetic stirring at 25 °C for 2 h. The chitosan concentration in the acetic acid solution was fixed at 2.5 g dm^{-3}. On the other hand, we prepared a FTIC solution (concentration = 3.75 g dm^{-3}) using anhydrous ethanol as solvent. FTIC was dissolved in a dark glass bottle by magnetically stirring at 25 °C for 1 h. Then, 6 cm^3 of the FITC solution was mixed with 6 cm^3 of the chitosan solution by magnetic stirring for 2 h at 25 °C in dark. Afterwards, 10 cm^3 of NaOH solution (concentration = 0.1 mol dm^3) were added to the chitosan/FITC mixture in order to precipitate the labeled biopolymer.

The obtained chitosan/FITC precipitate was washed five times with an ethanol/water (70:30 by volume) mixture in order to remove free FITC. Finally, the chitosan/FITC material was dissolved in acetic acid solution (concentration = 10 g dm^{-3}), which was dialyzed in 10 kDa cutoff dialysis tubing against deionized water for 3 days. In detail, 20 cm^3 of the chitosan/FITC solution were dialyzed against 500 cm^3 of water for 24 hours. The dialysis procedure was repeated three times. As reported elsewhere,[3] 1 mole of chitosan should be averagely labeled by 9.5 moles of FITC.

References

(1) Mackie, A.; Bajka, B.; Rigby, N. Roles for Dietary Fibre in the Upper GI Tract: The Importance of Viscosity. *Food Research International* **2016**, *88*, 234–238. https://doi.org/10.1016/j.foodres.2015.11.011.

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(3) Tallury, P.; Kar, S.; Bamrungsap, S.; Huang, Y.-F.; Tan, W.; Santra, S. Ultra-Small Water-Dispersible Fluorescent Chitosan Nanoparticles: Synthesis, Characterization and Specific Targeting. *Chem. Commun.* **2009**, No. 17, 2347–2349. https://doi.org/10.1039/B901729A.

Calculation of the number density and the volume of halloysite nanotubes

The number density (^1N) is defined as the number of scattering objects per unit volume. On this basis, ^1N of the investigated HNTs dispersion can be calculated as

$$^1N = c(HNT)/m(HNT) \quad (1)$$

where c(HNT) is the concentration of HNT dispersion, while m(HNT) represents the mass of a single nanotube.

Given the density of halloysite ($\delta(HNT) = 2.53$ g cm^{-3}), the mass of a single nanotube can be determined by using the following equation

$$m(HNT) = V(HNT) \cdot \delta(HNT) \quad (2)$$

V(HNT) is the volume of a single nanotube that can be calculated as

$$V(HNT) = (R_e^2 - R_i^2) \cdot \pi \cdot L \quad (3)$$

where L. R_e, R_i are the length, external and internal radii, respectively.

L was fixed at 1000 nm, while R_e and R_i were fixed at 80 and 15 nm, respectively. These sizes are the average values reported in literature from microscopy studies.

Determination of the HNT/biopolymer volume ratio of the coated nanotubes

The biopolymer/HNT volume ratio ($RV_{(Biop/HNT)}$) was calculated by the following equation

$$R_{V(Biop/HNT)} = (SLD_{Biop} \cdot C_{HNT})/(SLD_{HNT} \cdot C_{Biop} \cdot \chi) \quad (4)$$

where C_{HNT} and C_{Biop} refer to the concentrations (expressed as g dm^{-3}) in the investigated mixtures for halloysite and biopolymers, respectively, while SLD_{HNT} and SLD_{Biop} are the densities of each components. χ corresponds to the fraction of biopolymer adsorbed onto halloysite.