Supporting Information

Understanding lignin aggregation processes. A case study: budesonide entrapment and stimuli controlled release from lignin nanoparticles

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Contents

10 pages including nine Supplementary Figures



Figure S-1. Effect of ethanol concentration on solubility of wheat straw soda lignin. Solubility was determined after 20 h stirring at room temperature. The suspensions were centrifuged and the soluble amount of lignin was determined by UV-vis spectrophotometry at 280 nm (ϵ =21.4 L g⁻¹ cm⁻¹ in 0.1 M sodium hydroxide)



Figure S-2. Effect of 1-6 g L⁻¹ initial lignin concentration on aggregation of LNPs. Final ethanol concentration was 13%.



Figure S-3. Optimization of the loading ratio of budesonide for entrapment in LNPs. SEM images of crystals and particles formed when the initial solution mixture contained 100%...5% of budesonide relative to total amount of dissolved material (lignin+budesonide).



Figure S-4. UV-Vis absorbance spectra of mixtures and pure substances of budesonide and lignin in ethanol-water 7:3 v/v. (**a**) Spectra of the crude and resin-purified permeate fractions from budesonide entrapment in LNPs. Crude permeate exhibits absorbance typical for lignin at 280–400 nm, while in the resin-treated (Amberlite IRA-900 Cl) permeate the absorbance in this region is lower. The absorbance values were corrected for dilution to enable direct comparison. (**b**) Spectra of pure budesonide and GreenValue wheat straw soda lignin at 0.04 g L⁻¹ concentration in 70% ethanol (aq.).



Figure S-5. (a) HPLC chromatograms of budesonide released from LNPs after 10 h stirring at pH 7.4. (b) HPCL and UV spectrophotometry determinations of dissolution kinetics of budesonide at pH 2, 5.5, and 7.4 in 0.05 M buffer solutions at 37 °C. (c) SEM images of budesonide-LNPs at pH 7.4 without SDS (c1, 48 h and c2, 25 h) and after SDS supplementation (c3, 31 h).



Figure S-6. Digital photographs of budesonide-LNP dispersions after 96 h incubation at 37 °C.



Figure S-7. Dissolution of budesonide from the unpurified budesonide-LNPs that contained 73% of free budesonide while the remaining part was entrapped in LNPs. The solubility limit is higher (20 mg L⁻¹) and is reached faster in the presence of lignin compared to the dissolution of the pure active under saturation conditions at pH 7.4. Data points with error bars indicate mean values \pm average deviation of two replicates. Other data points are single determinations.



Figure S-8. HPLC calibration curves for budesonide.



Figure S-9. Calibration curves used in the ABTS antioxidant assay.