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Using particle tracking to probe the local dynamics of barley β-glucan solutions

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Abstract

The sol-gel transition of barley isolated β-glucan solutions which undergo gelation with ageing has been studied by conventional bulk rheology, phase contrast microscopy and particle tracking microrheology. Also, characterization of primary structure of the β-glucan isolate was carried out by HPLC. The Brownian diffusion of fluorescent microspheres (0.75 μm diameter, carboxylate-coated particles) was used to probe spatial mechanical properties of the gels at the scale of microns. The potential use of passive particle tracking as a new method of studying food systems that present spatial heterogeneities is explored. For the β-glucan gels cured at 25°C both the microrheology and the bulk rheology revealed that with increasing concentration of the polysaccharide the gelation time decreased, while the gelation rate and gel strength of the barley β-glucan gels increased. Moreover, the melting point increased with increasing concentration of the β-glucans indicating a better organization of the ordered domains in the network structure. The particle tracking method had higher sensitivity and could map molecular ordering and structural heterogeneities at a micro level. Furthermore, this method could detect changes in the structuring of the system before such events can be registered by the bulk rheological measurements. For these reasons, the timescales of the microscopic dynamic behavior do not always seem to match the timescales of the overall macroscopic behavior.

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Keywords: β-glucans; Particle tracking microrheology; gelation

1. Introduction

Cell walls of cereal grains, mostly barley and oat, are rich in mixed-linkage (1→3), (1→4)-β-D-glucans (β-glucans), which are linear polysaccharides of D-glucose residues interlinked via β-(1→3) and β-(1→4) linkages; their structure consists of consecutively (1→4)-linked β-D-glucose in blocks (i.e. oligomeric cellulose segments) that are separated by single (1→3)-linkages [1]. Cereal β-glucans display

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all the functional properties of viscous and gel forming food hydrocolloids combined with all the health benefits of soluble dietary fibers, such as reduction of plasma cholesterol and of postprandial serum glucose levels in humans and animals. These physical and physiological properties of cereal β-glucans largely depend on their molecular characteristics and polysaccharide concentration.

Passive particle tracking is a direct method consisting of observing the Brownian motion of tracer particles within a system and interpreting this movement in terms of the local viscoelasticity, or microrheology. In this type of passive microrheology, there is no external driving force applied to the tracer microspheres. The local particle motion is driven solely by Brownian forces generated by the thermal energy k_BT. Therefore, particle tracking can probe spatial variation in mechanical properties at the scale of microns without significantly distorting or altering the microstructure, providing that a sufficiently low concentration of tracer particles of appropriate size is employed. On the other hand, bulk rheological measurements describe the overall mechanical response of a material. That is, it is assumed that the sample is homogeneous and that there is no local variation in the structure. Although this assumption is valid for simple fluids, most colloidal systems are more complex and commonly present spatial heterogeneities [2, 3]. In order to understand the origins of the overall response, it is therefore necessary to probe rheology over shorter length scales and particle tracking provides this possibility.

2. Materials & Methods

In the present study, a barley β-glucan sample was used; this was an isolate from a barley concentrate supplied by CEBA (Lund, Sweden). The purification protocol for this preparation was described in detail by Lazaridou et al. (2003) [4] involving a dual-enzyme digestion with thermostable α-amylase and pancreatin followed by exhausting dialysis and precipitation of the polysaccharide by ethanol. The purity of the β-glucan isolate was estimated by determination of the barley β-glucan and protein contents using an (1→3, 1→4)-β-glucan assay kit purchased from Megazyme International Ltd (Bray, Ireland) and by the method of Lowry et al. (1951) [5], respectively.

The apparent peak molecular weight (Mw^p) of the β-glucan sample was obtained with a high performance size exclusion chromatography (HPSEC) system combined with a refractive index (RI) detector. Calculation of the Mw^p from the peak fraction of main eluting peak of the HPLC chromatogram was based on calibration with β-glucan standards isolated according to Lazaridou et al. (2004) [6] and having Mw^p of $466x10^3$, $300x10^3$, $186x10^3$, $83x10^3$, $33x10^3$ and $15x10^3$, as characterized by a light scattering technique [7]; eluting volumes of peak fractions for both standards and unknown samples were used in this calculation. The distribution of cellulosic oligomers in the chain of β-glucans was determined by treatment with lichenase [(1→3),(1→4)-β-glucan-4-glucanohydrolase, EC 3.2.1.73] and chromatography. High-performance anion-exchange chromatography (HPAEC), combined with a pulsed amperometric detector (PAD), was employed for analysis of oligosaccharides released from β-glucan by lichenase action. Description of the sample preparation and running conditions of both HPLC methods as well as of the determination of limiting viscosities [η] of the β-glucan sample using Ubbelodhe capillary viscometer are described in detail elsewhere [4].

The gel curing–melting events for the β-glucan preparations at different polysaccharide concentrations (2-5% w/w) were performed on a rotational Physica MCR 300 rheometer (Physica Messtechnic GmbH, Stuttgart, Germany) using double gap cylindrical geometry (bulk rheometry) as previously described [4].

Particle tracking: An optical microscope mounted on an Olympus BX61 microscope base, was operated in the fluorescence mode with a 100x oil-immersion objective of numerical aperture 1.25. The fluorescently labeled microspheres were added at a concentration of ~ 0.2 %v/v. The samples were then immediately placed into a welled slide filling it completely. Images were scanned approximately 10-20 μm below the level of the coverslip to minimize hydrodynamic interactions with the coverslip. All the measurements were conducted at 25°C. Trajectories of fluorescently labeled COOH-PS microspheres were recorded using an AVT Pike CCD camera at a frame rate of 30 Hz and short exposure time of