How the type of exopolysaccharide affects the rheological properties of fermented products

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ABSTRACT

In situ produced exopolysaccharides (EPS) influence processing steps and properties of the fermented products. Fermentation broths from starter culture production and fresh cheese were used to investigate the effects of different types of EPS with physical and rheological methods. A strong influence of the type of EPS was found on yield stress, viscosity and sedimentation behaviour, hence processing steps and product properties can be controlled by a proper selection of starter cultures.

INTRODUCTION

Some lactic acid bacteria (LAB) are capable of synthesising exopolysaccharides (EPS), which show texture-enhancing effects[1]. These strains are therefore more and more used for the manufacture of fermented dairy products (e.g. yoghurt, cheese). However, the *in situ* produced EPS increase also medium viscosity during production of the starter culture itself, impeding efficient cell separation.

Influence of *in situ* produced EPS on properties of dairy products are already acknowledged, but little is known on the effects of a particular type of EPS. EPS can be distinguished by their location into free EPS (fEPS) present in the medium and capsular EPS (cEPS), attached to the bacterial cells, and by the effect they evoke into ropy and non-ropy EPS[2].

To shed more light on this issue we selected two strains with different types of EPS and determined the rheological and physical properties of the fermentation medium from starter culture production and of fresh cheese produced by them.

MATERIAL AND METHODS

Cultures

Two *Streptococcus thermophilus* (ST) strains were selected from a screening trial based on occurrence of ropiness and ability to produce cEPS: ST-E, produces ropy fEPS and ST-G, produces non-ropy fEPS and cEPS. Fermentation broth from starter culture production was defrosted at 4 °C for 24 h. Cell-free samples were obtained by centrifugation at 19000 g for 15 min at 4 °C and decantation of the supernatant.

Fresh cheese production

Reconstituted skim milk (dry matter 14.5 g/100 g) was prepared by dissolving low heat skim milk powder (Sachsenmilch Leppersdorf, Wachau, Germany) in deionized water. After storage for 24 h, the milk was heated to 90 °C for 10 min, subsequently cooled to 40 °C and inoculated with 1 mL/100 g precultured ST-E or ST-G. pH was continuously monitored and fermentation stopped at pH 4.6 by cooling in ice water to 20 °C. The milk gels were further processed by shearing with an Ultra-Turrax® T50 (Ika Werke, Staufen, Germany; G45F, 6400 rpm, 1 min), heating

(60 °C, 5 min) and centrifugation at 40 °C. Centrifugal force was adjusted to obtain 40 g fresh cheese from 100 g milk gel after whey separation[3].

Rheological characterisation

Flow curves of the fermentation broth were recorded in a double gap geometry (d_o = 44 mm, d_i = 41 mm, $d_{i,stator}$ = 40 mm, h = 59.5 mm) of an AR-G2 rheometer (TA Instruments, New Castle, USA) at 20 °C. Shear rate $\dot{\gamma}$ [1/s] was increased from 0.1 to 1000/s in a logarithmic ramp with 5 points per decade and 30 s per point.

Fresh cheese was examined after 14 d of storage at 4 °C. Shear stress τ [Pa] was determined at 15 °C with a plate geometry (d = 40 mm) as a function of $\dot{\gamma}$, which was increased from 0.001 to 100/s with 10 points per decade and 20 s per point. Herschel-Bulkley model parameters were obtained after fitting the flow curves ($\dot{\gamma}$ = 0.1 - 100/s).

Sedimentation behaviour

Phase separation of sheared and heated milk gels was investigated with an analytical optical centrifuge (LUMiSizer® 610, LUM GmbH, Berlin, Germany). Centrifuge tubes were filled with ~ 1.75 mL milk gel and closed with an in-house developed plug to prevent movement of the sample in the horizontal direction. Transmission was recorded at 40 °C and a centrifugal force of 900 g.

RESULTS AND DISCUSSIONS

Viscosity curves of cell containing starter culture medium revealed a slight shear thinning behaviour for both strains with η decreasing above $\dot{\gamma}$ ~ 10/s (see Fig. 1). However, viscosity of the fermentation medium with ST-E was higher compared with ST-G, reflecting the viscosity increasing effect of the ropy fEPS. After removing the cells the viscosity of the medium from ST-E slightly decreased. In contrast, ST-G cell-free medium showed a much lower viscosity (η at 10/s = 1.4 mPa·s) compared to the medium with cells (η at 10/s = 2.0 mPa·s), indicating that the non-ropy fEPS do not influence medium viscosity. ST-G also produces cEPS, which are removed with the cells, and this caused a significant viscosity decrease. Cells from ST-E are not covered with cEPS and, consequently, their removal did not result in such a strong effect. This means that both types of exopolysaccharides do have an influence on the medium viscosity and consequently also on the efficiency of cell separation during starter culture production.

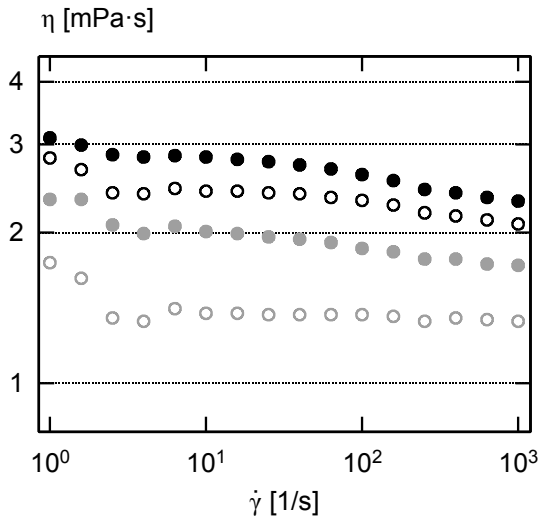

Figure 1. Viscosity curves of fermentation broth of ST-E (black) and ST-G (grey) with cells (closed) and cell-free (open).

The same two *S. thermophilus* strains were used to obtain milk gels for further processing into fresh cheese. Their sedimentation behaviour was determined by using an optical centrifuge. Figure 2 shows the progress of the transmission profiles during forced sedimentation. A minimum in transmission at cell position 26 mm indicates the sample height. During measurement the transmission increased and the phase boundary between sediment and supernatant moved towards 0 mm. A faster increase in transmission within the first

profiles (grey lines) was detected for milk gels fermented with ST-E compared to ST-G, indicating a faster initial phase separation. Moreover, a clear phase boundary (recognizable by a straight vertical line) was formed earlier for ST-E than for ST-G.

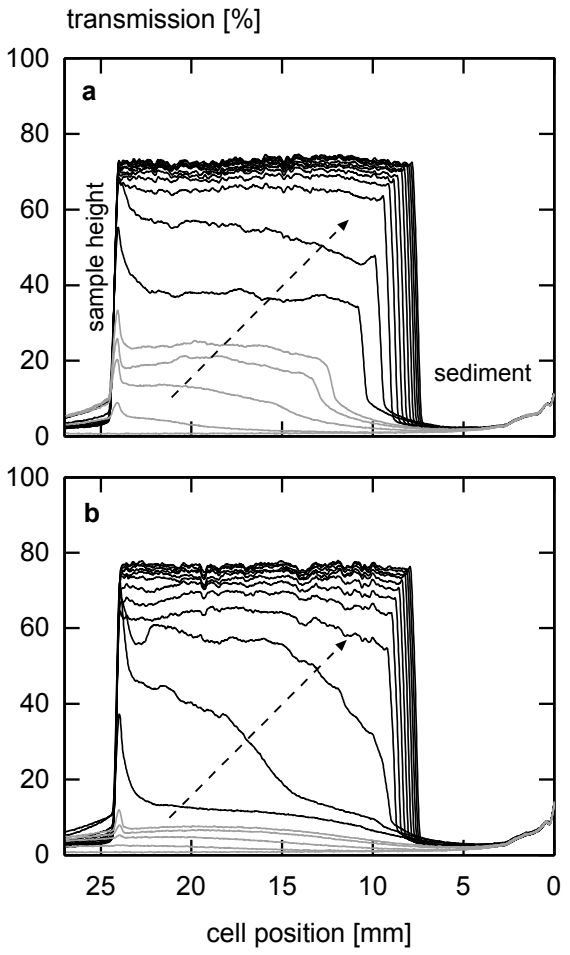

Figure 2. Transmission as function of cell position at selected time points during centrifugation of milk gels fermented with ST-E (a) and ST-G (b). 10 s (grey) and 100 s (black) intervals between transmission profiles. Arrow indicates time progress.

With progressive measurement a decrease in the distance between the consecutive phase boundaries was observed, suggesting a so-called zone sedimentation and the sediment was compressed[4]. Increase in transmission during compression indicates the clearance of the supernatant from fine particles[5]. The begin of both sediment compression and clearance occurred later for milk gels fermented with ST-G. However, these samples showed also slightly curved transmission profiles above the phase boundary, which points to polydisperse sedimentation of fines[4].

This leads to the conclusion that milk gels containing ropy fEPS are less stable against forced sedimentation compared to non-ropy fEPS and cEPS. In the context of fresh cheese manufacture a lower stability means better phase separation, hence a higher cheese yield. Quantitative analysis of these experiments is under progress.

Flow curves of fresh cheese samples showed an increase in τ with rising $\dot{\gamma}$ until a maximum, which was followed by a plateau region and a further increase (see Fig. 3).

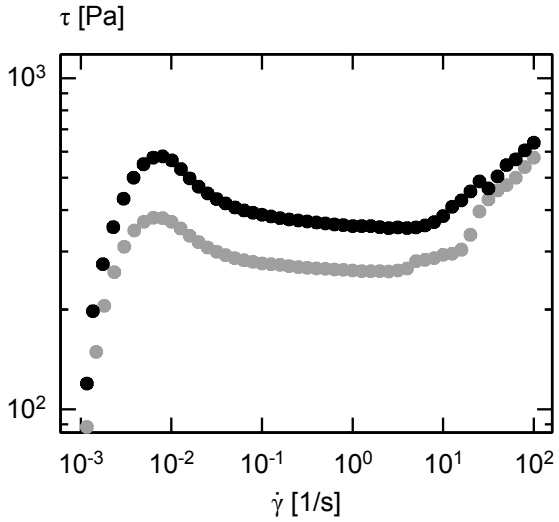

Figure 3. Flow curves of fresh cheese produced with ST-E (black) and ST-G (grey).

The shear stress maximum τ_{max} at low $\dot{\gamma}$ can be attributed to a static yield stress[6,7], which is followed by a shear-induced break-up of aggregates. Fresh cheese with ST-E showed higher τ_{max} = 582 Pa compared to ST-G (τ_{max} = 396 Pa). Curve fitting with the Herschel-Bulkley model gave generally lower absolute yield stress values (τ_0)

compared to static yield stress (τ_0 = 358 Pa for ST-E and τ_0 = 260 Pa for ST-G). The non-ropy fEPS and cEPS producing strain (ST-G) led to a lower yield stress than the ropy fEPS (ST-E), which corresponded to findings with stirred yoghurt[8,9]. Further research on physical and rheological properties is currently under progress.

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