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The role of exopolysaccharide-producing cultures and whey protein ingredients in yoghurt

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ABSTRACT

Physical stability and textural properties of eighteen low fat stirred yoghurts, differing in type of added dairy protein ingredients and in exopolysaccharide (EPS)-producing cultures, were studied by instrumental and sensorial measurements. Image analysis was performed to quantify the amount of grains and the amount of EPS. Enhanced textural properties and physical stability was observed in systems containing whey protein concentrate or particulated whey proteins as well as EPS-producing cultures. Irrespective of the culture used, casein and fractionated whey protein did not contribute to the textural properties. Sensory analysis confirmed the results obtained by instrumental measurements. Oral gel firmness was highly correlated to G′, viscosity at low shear rate, and instrumental measured firmness ($R \approx 0.82$). Mouth thickness was well described by viscosity at high shear rate ($R = 0.81$), as well as by the hysteresis loop area and ropiness ($R \approx 0.81$). Measured syneresis showed a significant relationship with visually evaluated syneresis ($R = 0.76$), and the amount of grains also correlated with visual evaluated graininess ($R \approx 0.81$). The effect of the cultures on the sensory properties was revealed by multilevel simultaneous component analysis (MLSCA-P).

1. Introduction

The textural attributes and physical stability of stirred yoghurt define the initial acceptance and degree of product quality for the consumers and are mainly evaluated as firmness on the spoon and in the mouth, visual and oral viscosity, smooth and shiny surface, plus creaminess and mouth thickness. Good physical stability of low fat stirred yoghurt is related to no or negligible syneresis, and absence of visual or perceived grains. Textural properties and physical stability can be enhanced by fortifying the initial milk with ingredients such as milk proteins and/or stabilizers. Skim milk powder (SMP) is often used but whey protein concentrate (WPC), whey protein isolate (WPI), and sodium caseinate are viable alternatives. Other options are the use of stabilizers such as gelatin, starch or other hydrocolloids, which improve the functional properties, but they can also be the cause of off-flavors (De Vuyst & Degeest, 1999). Consumers' expectations and wishes are moving toward products containing fewer additives, and there has thus been focus on the *in situ* production of EPS produced by lactic acid bacteria (LAB). EPS can either be composed of long linear chains or have a more complex structure with side-chains (De Vuyst & Degeest, 1999). The type of EPS including monomer composition, molecular weight, branching, linkage type, and charge are specific for the producing strain. EPS-producing cultures are known to augment the final viscosity/mouth feel, ropiness, smoothness, and to reduce syneresis. Inconsistent findings have however been reported on viscoelastic parameters, firmness, cohesiveness and maximal penetration force (Folkenberg, Dejmek, Skriver, Skov Guldager, & Ipsen, 2006; Girard & Schaffer-Lequart, 2007; Hahn et al., 2014; Purohit, Hassan, Bhatia, Zhang, & Dwivedi, 2009). The effect on rheological parameters very much depends on the type of EPS and on the possible interactions with milk proteins (Gentès, St-

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Gelais, & Turgeon, 2011, 2013). Interactions between milk proteins and EPS are known to enhance firmness and viscosity of fermented milk and to reduce syneresis and graininess (Ayala-Hernández, Hassan, Goff, & Corredig, 2009; Girard & Schaffer-Lequart, 2007). However, a clear understanding of the mechanisms involved in the interactions, and how those in turn influence structure formation and final texture, is still lacking. In addition, studies attempting to correlate sensorial texture attribute to instrumental analysis mainly focus on a limited set of specific sensorial and/or physical properties, varying either the composition of the bacterial culture or the composition of the milk (Folkenberg et al., 2006; Krzeminski et al., 2013; Skriver, Holstborg, & Qvist, 1999). The present study focused on the effect that different commercial EPS-producing cultures and whey protein ingredients have on the physical and sensorial properties of a varieties of yoghurt systems. This was done to contribute to a better understanding of the interactions occurring between the biopolymers, and of the relationship between sensorial attributes and instrumental measurements. The latter was studied using a novel data modeling method, MLSCA-P (Timmerman, 2006).

2. Materials and methods

2.1. Yoghurt manufacturing

Eighteen low fat stirred yoghurts, which differed both in protein ingredients and in culture used, were manufactured at pilot plant scale. Medium heat SMP (Arla Foods Ingredients, Viby, Denmark) was dissolved in demineralized water to a final protein concentration of 3.5% and 4.5% for the controls, and 2.5% for samples with added ingredients. The final volume of the milk base was 3 L. The milk bases were fortified with 1% protein using four different whey ingredients (Arla Foods Ingredients) chosen to give a high degree of variation in textural properties. They included: whey protein concentrate (WPC), particulated whey protein (PWP), whey fraction (WF), and a product with a casein to whey protein ratio of 1:1 (WC). The ingredients were added simultaneously with the SMP, and the obtained solutions were stirred at 600 rpm for 3 min (Janke & Kunkel, Germany). The milk bases were stored at 5 °C overnight for protein hydration. A thermal treatment, 90 °C for 20 min in water bath, followed. Three different yoghurt starter cultures (Ch. Hansen A/S, Hørsholm, Denmark) were selected in order to confer high variation in amount and type of EPS released into the yoghurt. They included two cultures known to produce high levels of EPS (based on the manufacturer's information), Yo-Flex Premium-1 (Premium-1) and YF-L901, and one culture known to produce low levels of EPS, YF-3331. Fermentation was performed at 43 °C using 0.02%(v/v) culture inoculum. Fermentation was terminated at pH 4.55. Subsequently, the coagulum was broken by a perforated disk, cooled to 18 ± 1 °C in a plate heat exchanger, and then smoothed by passing it through a pipe system with a back pressure of 2 bars (FH Scandinox A/S, Tarm, Denmark). The yoghurts were stored at 5 °C in 250 ml plastic containers for instrumental measurements, and in 155 ml transparent plastic containers for sensory evaluation. Instrumental measurements were performed after 3 days of storage, whereas sensory evaluation was performed 1 week after production. A minimum of 2 productions were conducted for each system.

2.2. Rheological measurements

2.2.1. Small deformation rheology

Dynamic oscillatory rheology and flow tests were performed using a rheometer (AR G2, TA Instruments, US based) equipped with a cone-plate geometry (diameter 40 mm; angle 2°; gap 52 μm) set to 10 °C. The samples were manually stirred before loading, and 3 min equilibration applied. Prior to a frequency sweep measurement (0.5% strain, 0.1–100 Hz), the linear viscoelastic region was identified by a strain sweep test (1 rad/s, 1×10^{-3}–$1 \times 10^{+3}$% strain). From the oscillation test the storage modulus G′ and the phase angle δ were analyzed. Flow curves were determined by applying a shear rate from 1×10^{-3} to 300 s^{-1}, and back to 1×10^{-3} s^{-1}, within 10 min for each ramp. From the flow test, the apparent viscosity at 1 s^{-1} (η_1) and at 300 s^{-1} (η_{300}), and the hysteresis loop area (H) were analyzed. The latter represents the ability of the yoghurt network to rebuild after applying shearing, and it was calculated as the area (1/s × Pa) between the flow curves (TA Instruments Ltd, version V5.7.0).

2.2.2. Large deformation rheology

A back extrusion test was performed using a texture analyzer (TA.XT Plus, Stable Micro System, Surrey, UK) with a 35 mm parallel plate. The travel distance was set to 15 mm with a speed of 2 mm/s. The test was performed in the 250 g container, directly after removal from 5 °C storage. From the obtained force versus distance curves, the maximal force (firmness, F, N), the positive area (degree of deformation, D, N × mm), the maximal negative force (ropiness, R, N) were analyzed.

2.3. Syneresis and water holding capacity

Syneresis (S) was determined according to a method reported by Amatayakul, Halmos, Sherkat, and Shah (2006) with minor modification. Five days after manufacture, the yoghurt cups were placed at room temperature with an inclination of 45°; after 1 min the whey on the surface was weighed and indicated as %(w/w). Water holding capacity (WHC) was calculated as %(w/w) by weighting the supernatant formed after centrifugation of 21 *g* of yoghurt at 5 °C for 10 min at 3913 *g*.

2.4. Particle size distribution

Particle size distribution was measured by static light scattering (Malvern Mastersizer, Malvern Instruments Ltd., Worcestershire, UK). The refractive index indicating the real part was set to 1.52, the imaginary part was set to 0.1, and refractive index of water, 1.33, was used for the dispersant phase. The parameter $D_{0.5}$ was used for further analysis.

2.5. Graininess

Graininess (G) was evaluated by image analysis, and the sample preparation was conducted according to Remeuf, Mohammed, Sodini, and Tissier (2003). Images were obtained by a VideometerLab instrument (Videometer A/S, Hørsholm, Denmark). The spatial resolution of the images was 74 μm per pixel. The RGB images were imported into MATLAB (Mathworks, Natick, MA, USA). A background area was selected and used for color/intensity correction between different images. For this, the first image of the full batch was taken as reference, in such a way that the average of the color of the background reference was calculated. This generates a vector P_{ref} (3 × 1) containing the average for the three color channels (RGB). Assuming that the intensity differences between the reference image and the subsequent images are linear, the least square regression between P_{ref} and a vector containing the average of background area of a new image (P_{im}) can be calculated as:

$$L = \left({P_{ref}}^{T} P_{ref}\right)^{-1} {P_{ref}}^{T} P_{im} \quad (1)$$