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# QUALITY AND STABILITY OF THE NATURAL YOGURT PRODUCED FROM MILK CONDENSED WITH WHEY PROTEIN CONCENTRATE

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#### ABSTRACT

Increase of dry substances in the processed milk, made by introducing various milk protein agents, has an influence on the properties of produced yogurt and its shelf life. This study examined the effect of adding 5% of powdered skim-milk and whey protein concentrate, and their mixtures in proportions 4:1 and 3:2, on quality and stability of natural yogurt. Titratable acidity was determined and active acidity, apparent dynamic viscosity and syneresis susceptibility were measured in yogurts directly after production, and after 7, 14 and 21 days of storage in temperature 8±1 °C. The produced yogurts were assessed organoleptically. Introduction of 5% addition of powdered skim-milk with whey protein concentrate mixed in above mentioned proportions resulted in essential increment of apparent dynamic viscosity and reduced syneresis susceptibility in the yogurts, comparing to separate use of the both components. These yogurts were also indicated as the most desired. During 3 weeks of storage, the yogurts show adverse changes in analyzed parameters, which leads to deterioration of their quality.

Key words: set yogurt, whey protein concentrate, quality, shelf life.

# INTRODUCTION

Variety of forms in which yogurts are available (drinks, gels, pastes, dry or frozen) may be achieved due to specific technologies that are also based on suitable preparation of the source material. When intended curd properties are reached, this may ensure sufficient level of dry matter in milk to be used for yogurt production.

Among wide range of technological additives applied in processing milk condensation process, also the additives that form the curd structure are mentioned. Substances that are used to form a structure of many foodstuffs include animal derived protein preparations (e.g. from milk) and plant protein preparations (e.g. from leguminous plants), which are not considered as food additives, but as its ingredients. Increment of dry matter in the processing milk designed for yogurt production is usually reached by adding: condensed milk (that contains dry matter in amount not less than 25%), powdered skim or whole milk, all milk proteins concentrate, whey proteins concentrate or casein derivatives. These agents differ with not only varied content of total protein, which is ranging from several to almost 90%, but also with proportions between basic milk proteins, i.e. casein, and whey proteins. Milk protein agents introduced to the processed milk affect natural proportions between casein and whey proteins, which determines functional properties of the milk during processing as well as firmness and syneresis susceptibility of the received curd. According to Modler et al. [12], predominant amount of casein comparing to whey proteins effects in rough and coarse-grain structure of yogurt, whilst balanced level of the both proteins gives too loose and too soft curd. Too much whey proteins (over 2%) additionally bring unfavorable taste and smell of yogurt, which is a result of high lactose content in these preparations [17]. Yogurt made from processing milk to which only sodium casein was added, had a firm curd with lower syneresis susceptibility than the yogurt made from milk, to which only whey proteins were added. However, the latter had smoother curd and better look [18]. That is why, addition to the processing milk of any agents that in majority contain casein or whey proteins must be carefully balanced.

This study was aimed to analyze physical, chemical and organoleptic characteristics of the natural yogurt made from processing milk that was condensed by adding whey protein concentrate and powdered skim-milk, directly after production and throughout storage in cold conditions.

#### MATERIALS AND METHODS

Material used in the study was natural set yogurt received in semi-commercial scale of production. The yogurt was made from pasteurized homogenized milk. Source material was pre-selected raw milk having basic chemical constitution corresponding to constitution of milk collected domestically [8], density not less than 1.028 g/cm<sup>3</sup> and freezing temperature not higher than -0.512°C, titratable acidity from 6.0 to 7.5°SH and active acidity with pH value from 6.6 to 6.8 [15]. While, the total count of microorganisms in the milk designed for processing was less than 100 000 cfu/cm<sup>3</sup>, and somatic cells number below 400 000 in 1 cm<sup>3</sup> of milk [3]. Processing of raw milk was absolutely conditioned on the absence of any antibiotics or other inhibitive substances. Powdered skim-milk or whey protein concentrate WPC-60, or their mixture in proportions 4:1 and 3:2, were introduced to the processing milk. Both the powdered skim-milk and whey protein concentrate or their mixtures were introduced in the amount of 5% of the processing milk volume, which allowed to condense dry matter in the processing milk from 12.67 to 17.02%. The introduced powdered skim-milk contained 96.2% of dry matter, including 33.40% of protein and 51.64% of lactose. While, the whey protein concentrate contained dry matter at the level of 95.9%, including 53.60% of protein and 7.12% of lactose. Raw milk was pasteurized together with additives or their mixtures in the temperature of 86°C for 3s. After pasteurization the milk was cooled down to 43°C and inoculated with low aromatizing bacterial culture comprising traditional strains: Lactobacillus delbrueckii subsp. bulgaricus and Streptococcus thermophilus, in the ratio of 1:1. The applied cultures were in lyophilized form designed for direct input to processing milk, type Direct Vat Set (DVS). They were added in the amount of 50 activity units for each 250 cm<sup>3</sup> of the processed milk, which corresponded to adding 2% of activated working starter. The cultures came from the collection of Chr.Hansen's Laboratorium Danmark A/S. Inoculated milk was distributed into unit packages of capacity equal to 150 cm<sup>3</sup>. Natural yogurt was incubated in the air at the temperature of 43°C. After 6 hours, the incubation was interrupted by immediate decrease of the temperature to  $6^{\circ}$ C, and the produced yogurt matured in this temperature for the following 18 h. The received natural yogurt was stored for 21 days in the temperature of 8±1°C.

The following determinations and measurements were made in the produced yogurts: acidity (titratable and active), rheological measurements (apparent dynamic viscosity and flow curves) and syneresis susceptibility. Organoleptic assessment included such attributes like appearance, color, consistency, taste, smell and over all acceptability. Titratable acidity was determined in accordance with conditions described in applicable standard [7]. Active acidity was measured in the yogurt using pH-meter type CP - 315 by Elmetron, provided with combined electrode type ESAgP-301W by Eurosensor, that consisted of glass half-cell and saturated chlorosilvery half-cell. Rheological properties of the natural yogurt were measured by means of rotational rheometer type Brookfield RVT II by Brookfield Engineering Lab. Inc. Stoughton, using Couette's flow between two coaxial cylinders [5,13]. Measurements were made in the temperature of  $10\pm1^{\circ}$ C, in shear rate  $\gamma$  ranging from 2.2 to 111.4 s<sup>-1</sup> [20]. Calculations of shear rate and shear stress were made using Kiljański's method [9]. Flow curves for yogurt were drawn from measured values of shear stresses and apparent dynamic viscosity. For

comparative assessment, the value of apparent dynamic viscosity calculated for one-point shear rate equal to  $\gamma 10$  s<sup>-1</sup> was used [1,11]. Assessment of the yogurt syneresis susceptibility was made according to Dannenberg and Kessler [4]. Organoleptic assessment of yogurts was rated using 9-point scale [2]. The organoleptic assessment was made by trained group of seven persons [10]. To perform statistical analysis of the results the Excel spread sheet and statistical software SPSS/PC+ were used in this study [14]. The confidence intervals were determined at the established significance level of p = 0.05 [19]. To determine significance of arithmetic average differences for results achieved in the examined experiment groups, the analysis of variance and statistical verification using the NIR test were carried out. Determinations and measurements were made directly after yogurt production and after 7, 14 and 21 days of storage. Six production batches of yogurt were analyzed, and each measurement or determination was made in 3 repetitions.

## **RESULTS AND DISCUSSION**

In yogurts produced with participation of powdered skim-milk and whey protein concentrate, used separately and in mixture, the highest titratable acidity directly after production was found in yogurt with only powdered skim-milk and in mixture with whey protein concentrate in the proportion 4:1 (tab. 1). It was demonstrated that along with the time of storage the titratable acidity increased from 37.60 (directly after production) to 44.65°SH (after 3 weeks of storage). Whilst, it was found that statistically significant increment of titratable acidity in the examined yogurts occurred only in the first 2 weeks of storage. After the end of storage, the highest titratable acidity was still found in the samples taken from the milk that was condensed with only 5% additive of powdered skim-milk and with the mixture with whey protein concentrate in the proportion 4:1. Statistical analysis demonstrated significant effect of powdered skim-milk and whey protein concentrate additive, used separately and in mixture, on active acidity in the examined natural set yogurts (Table 1). It was demonstrated that the yogurts, which contain 5% of powdered skim-milk and the mixture with whey protein concentrate in the proportion 3:2, are characterized by significantly higher active acidity than the yogurts that contain in this mixture bigger of powdered skim-milk, or yogurts made solely on the base of powdered skim-milk or whey protein concentrate. After 3 weeks of storage, active acidity in the examined yogurts increased significantly (by 6.9%) comparing to acidity measured directly after production. The demonstrated active acidity increments in yogurts made from the milk that was condensed with mixtures are smaller than the increments found during storage in yogurts produced from the processing milk, whose dry matter had been condensed using individual components of that mixture.

Natural yogurt made	Percentage in mixture	Storage time (days)				
condensed using 5% of following additives		0	7	14	21	
Titratable acidity (°SH)						
Powdered skim-milk	100	38.61 <sup>bA *</sup>	40.04 <sup>bB</sup>	44.26 <sup>bC</sup>	44.80 <sup>bC</sup>	
Whey protein concentrate (WPC 60)	100	37.14 <sup>aA</sup>	39.61 <sup>aB</sup>	43.82 <sup>aC</sup>	44.51 <sup>aC</sup>	
Powdered skim-milk mixed with whey protein concentrate (WPC 60)	80 : 20	37.54 <sup>bA</sup>	39.96 <sup>bB</sup>	44.16 <sup>bC</sup>	44.68 <sup>bC</sup>	
	60 : 40	37.10 <sup>aA</sup>	39.65 <sup>aB</sup>	43.96 <sup>aC</sup>	44.60 <sup>aC</sup>	
Active acidity pH						
Powdered skim-milk	100	4.46 <sup>cC</sup>	4.46 <sup>cC</sup>	4.32 <sup>CB</sup>	4.10 <sup>bA</sup>	
Whey protein concentrate (WPC 60)	100	4.48 <sup>cC</sup>	4.45 <sup>cC</sup>	4.29 <sup>cB</sup>	4.07 <sup>bA</sup>	
Powdered skim-milk mixed with whey protein concentrate (WPC 60)	80 : 20	4.36 <sup>bC</sup>	4.34 <sup>bC</sup>	4.20 <sup>bB</sup>	4.14 <sup>bA</sup>	
	60 : 40	4.26 <sup>aC</sup>	4.23 <sup>aC</sup>	3.97 <sup>aB</sup>	4.03 <sup>aA</sup>	

Table 1. Titratable and active acidity in set yogurts directly after production and during their storage in cold conditions

a-c, A-C various small letters in columns entered for the same determination and various capital letters in rows represent statistically significant differences at the level of p = 0.05

Apparent dynamic viscosity in the analyzed yogurts was in much extent determined by composition of the proteins that were introduced to condense dry matter in the processing milk (Fig. 1). Replacement of powdered skim-milk in 20% by whey protein concentrate affected in statistically significant increment of the apparent dynamic viscosity in the yogurts. However the yogurts, in which next 20% of powdered skim-milk were replaced by whey protein concentrate showed statistically lower apparent dynamic viscosity than the yogurts that were made with participation of these additives in the proportion of 4:1. Among all analyzed samples, the lowest value of apparent viscosity was demonstrated in the yogurts made from milk condensed with 5% addition of only whey protein concentrate. The found relations and trends sustained in the samples during the whole storage period. Rheological parameters, determined using the Herschel-Bulkley model (yield stress, consistency coefficient and flow behavior index) of flow curves drawn for selected yogurts that were used in the experiment are presented in Table 2. It was found that among the selected examples the highest value of yield stress is represented by yogurts made with participation of powdered skim-milk in mixture with whey protein concentrate in the proportion of 4:1. While the lowest value of yield stress were found in yogurts made from the milk that was condensed using 5% additive of WPC 60 or the same amount of powdered skim-milk. Rheological properties of this type of yogurt correspond to equations established for Newtonian fluids almost perfectly [16]. It was also demonstrated that along with lowering of whey protein concentrate, the consistency coefficient is rising, which characterizes fluid consistency at shear rate  $\gamma=1$ , thus increasing viscosity and hardness of curd with stronger thicksotropic tendencies. Condensation of the processing milk with 5% addition of only WPC 60 and the same amount of powdered skim-milk effected the lowest consistency coefficients in yogurts made from that milk. Because the flow behavior index, which is a rheological parameter that reflects departure of the fluid from Newtonian properties, was, as demonstrated, the highest in yogurts made using 5% addition of WPC 60, therefore these yogurts show the least number of non-Newtonian properties (for Newtonian fluids n = 1). Hence the flow curve for this yogurt is closest to linear. The determined flow behavior indexes in all examined yogurts were much below 1, that is why they belong to pseudoplastic fluids. The most susceptible likely to be pseudoplastic fluids were the yogurts with a fraction of powdered skim-milk mixed with WPC 60 in the amount of 20%. Guinee et al. [6] when investigating flow curve in natural yogurts described by involutive model demonstrated the value of consistency coefficient of 13.8 Pas<sup>n</sup> for yogurts with 5% additive of powdered skimmilk, and within 1.8 to 13.9 Pas<sup>n</sup> for yogurt with whey protein concentrate of varied protein content. Whilst the authors found higher values of flow index in yogurts with addition of whey protein concentrate (n < 0.435) than in yogurt with powdered skim-milk (n = 0.155).





Natural yogurt made	Demonstration	Parameters by I	Correlation		
condensed using 5% of following additives	in mixture	yield stress ı <sub>y</sub> (Pa)	consistency coefficient k (Pa s <sup>n</sup> )	flow behavior index n	coefficient R
Powdered skim-milk	100	18.46	28.80	0.212	0.889
Whey protein concentrate (WPC 60)	100	10.36	22.65	0.239	0.882
Powdered skim-milk mixed with whey protein	80 : 20	50.32	93.08	0.084	0.965
concentrate (WPC 60)	60:40	38.31	63.39	0.131	0.944

 Table 2. Rheological parameters of flow curves in the examined set yogurts directly after production

Statistical analysis of the results collected in this study showed statistically significant effect of the milk condensation method on syneresis susceptibility in yogurts (<u>tab. 3</u>). It was found that introduction of powdered skim-milk and whey protein concentrate in mixture, regardless of their proportions, statistically limits susceptibility of the yogurt to syneresis. Among the analyzed samples, the most susceptible to syneresis were yogurts made from the milk that was condensed with only whey protein concentrate, and the demonstrated differences increased along with the time of storage.

 Table 3. Susceptibility to syneresis (%) in set yogurts directly after production and during cold storage

Natural yogurt made	Dereentage	Storage time (days)			
condensed using 5% of following additives	in mixture	0	7	14	21
Powdered skim-milk	100	4.3 <sup>bA</sup>	4.9 <sup>bA</sup>	6.2 <sup>bB</sup>	7.4 <sup>bC</sup>
Whey protein concentrate (WPC 60)	100	8.4 <sup>cA</sup>	8.8 <sup>cA</sup>	9.5 <sup>cB</sup>	9.6 <sup>bB</sup>
Powdered skim-milk mixed with whey protein concentrate (WPC 60)	80 : 20	1.0 <sup>aA</sup>	1.0 <sup>aA</sup>	1.0 <sup>aA</sup>	1.1 <sup>cA</sup>
	60 : 40	1.0 <sup>aA</sup>	1.0 <sup>aA</sup>	1.0 <sup>aA</sup>	1.1 <sup>aA</sup>

a-c, A-C various small letters in columns and various capital letters in rows represent statistically significant differences at the level of p = 0.05

As it might be expected, the results of measurements and determinations corresponded to organoleptic assessment of the examined yogurts (<u>tab. 4</u>). The results of organoleptic assessment of appearance, color, consistency, taste and smell decided how many points were awarded for over all acceptability of the examined yogurts. According to the evaluating persons the most desirable were the yogurts made from milk condensed with the mixture of powdered skim-milk and whey protein concentrate in proportion of 4:1, while the least desirable were yogurts in which the both components of the mixture were used separately. Such tendency was demonstrated irrespective how long the analyzed sample had been stored. Effect of the storage time resulting in over all acceptability decrement for the examined yogurt was demonstrated only in the last week of refrigerated store.

Natural yogurt made	Percentage in mixture	Storage time (days)			
condensed using 5% of following additives		0	7	14	21
Powdered skim-milk	100	5.7 <sup>aB</sup>	5.7 <sup>aB</sup>	5.5 <sup>aB</sup>	5.1 <sup>aA</sup>
Whey protein concentrate (WPC 60)	100	5.6 <sup>cB</sup>	5.6 <sup>aB</sup>	5.5 <sup>aB</sup>	4.8 <sup>aA</sup>
Powdered skim-milk	80 : 20	8.4 <sup>dB</sup>	8.3 <sup>dB</sup>	8.3 <sup>CB</sup>	7.8 <sup>cA</sup>
mixed with whey protein concentrate (WPC 60)	60 : 40	7.7 <sup>cC</sup>	7.4 <sup>cB</sup>	7.0 <sup>bA</sup>	7.0 <sup>bA</sup>

# Table 4. Over all acceptability of set yogurts directly after production and during cold storage

Footnote see Table 2

#### CONCLUSIONS

Summarizing the results of this study it may be said that:

- substituting the powdered skim-milk part by whey protein concentrate during milk condensation process leads to significant decrement of titratable acidity in the produced yogurts,
- the most proper rheological parameters were seen in the yogurts made from the milk that was condensed using a mixture of powdered skim-milk and whey protein concentrate in the proportion of 4:1,
- use of mixtures, comparing to introducing their components separately, essentially limited susceptibility to syneresis in the yogurts,
- unfavorable alteration of the yogurt quality during the time of storage may be limited by using the mixture of powdered skim-milk and whey protein concentrate in the proportion of 4:1.

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