Agronomy Research **18**(S3), 1897–1905, 2020
https://doi.org/10.15159/AR.20.111

Acceptance of low-sugar yoghurt among Latvian teenagers

J. Zagorska*, I. Ciprovica, E. Straumite and K. Majore

Latvia University of Life Sciences and Technologies, Faculty of Food Technology, Rigas iela 22, LV3004 Jelgava, Latvia
*Correspondence: jelena.zagorska@llu.lv

Abstract. Over a thousand year history, yoghurt has become one of a widely consumed product in the world. Its reputation as a healthy food has been undermined recently by concerns over the high sugar content. The majority of consumers expects and prefers yoghurts to be sweet. However, governments across Europe are calling for significant cuts in the amount of added sugar used in yoghurt production. The aim of the study was to evaluate the acceptance of low-sugar yoghurt produced by different commercial β-galactosidases by teenagers. Standardised milk with fat content 2.0% (SC Tukuma piens) was pasteurized at 95 ± 1 °C 5 min, cooled down till 43 ± 1 °C and fermented with β-galactosidase and starter YC-X11 (Chr. Hansen, Denmark) and fermented till pH 4.50 ± 0.20. Different commercial β-galactosidases: Nola™ Fit 5500, Ha-Lactase 5200 (Chr. Hansen, Denmark), GODO-YNL2 (Danisco, Denmark) and BrennZyme (Brenntag PolskaSp, Poland) were used. Fermented samples were gently mixed and cooled down till 6 ± 1 °C and 5% (w/w) of sugar was added to each sample. Sensory evaluation of the yoghurt's samples was performed by teenagers (14–18 years, $n = 50$) at Aizputes Secondary School (Latvia). Lactose and monosaccharides concentration prior to sugar addition was detected by HPLC (Shimadzu LC 20 Prominence, Japan).
The lactose hydrolysis into glucose and galactose by the use of β-galactosidase helps to increase sweetness through an occurrence of natural sugars in milk. During sensory evaluation, teenagers admitted the yoghurt with reduced sugar as sweet, significantly sweeter ($P < 0.05$) was yoghurt sample with Nola™ Fit 5500. The results demonstrated that it is possible to reduce sugar in yoghurt production and to gain consumer acceptance through the occurrence of glucose and galactose, but it is problematic to offer lactose-free or reduced lactose products to consumers without lactose intolerance.

Key words: acceptance, sweetness, yoghurt, β-galactosidase.

INTRODUCTION

Over a thousand-year history, yoghurt has become one of a widely consumed product in the world. Its reputation as a healthy food has been undermined recently by concerns over the high sugar content. Many studies updated the relationship between sugar consumption and health concerns: obesity (Li et al., 2015), dental caries, type 2 diabetes, and cardiovascular diseases (Lluch et al., 2017). In Europe (Ginder Coupez et al., 2017) and in Latvia the majority of consumers expects and prefers yoghurts to be sweet. Dairy products, comparing to other food products, are the leaders in high sugar concentration, still children and teenagers prefer sweet yoghurts and dairy desserts

(Azaïs-Braesco & Maillot, 2017). In Latvia sucrose concentration in yoghurts ranged from 6 to 25 g per 100 g. Dairy manufacturers often add sugar in higher doses to compensate for the taste of post acidification, finding the balance between sweet and sour. An average citizen, according to Latvia Statistics, consumes 80 grams of sugar per day. This is almost three times higher than 25 grams recommended by WHO (Guideline…, 2015). However, governments across Europe are calling for significant cuts in the amount of added sugar used in yoghurt production. The Dietary Guidelines in Latvia has recommended to reduce sucrose concentration in yoghurt till 5% for children at educational institutions (Cabinet Regulation 172..., 2012). Sugar reduction negatively influences yoghurt consumption (Lluch et al., 2017), and this goal can be achieved differently: replacing by steviol glycosides (Li et al., 2015), by adding natural sweetening flavours, the use of oligo-fructose and applying lactose hydrolysis (Rogenhofer & Hauß, 2019), as well as novel starters (Chr. Hansen, 2019). The application of commercial enzymes in yoghurt production allows create sweetness hydrolysing lactose and yielding sweeter monosaccharides (McCain et al., 2018). Relative sweetness of lactose is low (16), comparing to monosaccharides: glucose (70) (Tiefenbacher, 2017) and galactose (65) (Hobbs, 2009), therefore splitting of lactose to monosaccharides sums up higher product sweetness. The aim of the study was to evaluate the acceptance of low-sugar yoghurt produced by different commercial β–galactosidases by teenagers.

MATERIALS AND METHODS

Yoghurt preparation

Standardised milk with 2% of fat and 4.6% of lactose (SC Tukuma piens) was pasteurized at 95 ± 1 °C for 5 min, cooled down till 43 ± 1 °C and fermented adding β-galactosidase (see Table 1); after one hour of hydrolysis freeze-dried starter YC-X11 (*Streptococcus thermophilus, Lactobacillus delbrueckii subsp. bulgaricus, Lactobacillus fermentum*), was added 0.02% (w/w) according to the producer recommendations and fermented until pH 4.50 ± 0.20. The optimum pH range for all β-galactosidases is 5.4–8.0 (Table 1) and decreasing substrate pH cause enzyme inactivation (Zhou & Chen, 2001). Yoghurt samples were gently mixed and cooled down till 6 ± 1 °C and 5% (w/w) of sugar (Dansukker, Denmark) was added to each sample. A control sample was prepared according to the previously described technology without the usage of enzyme.

Different commercial food grade β-galactosidases: Nola™ Fit 5500, Ha-Lactase 5200, GODO-YNL2 and BrennZyme DairyLact were used (Table 1). Their characteristics are summarized in Table 1.

Carbohydrates detection

Lactose and monosaccharides concentration was detected by HPLC.

Sample preparation: samples were transferred into 2 mL test tube and deproteinized by adding 50 µL of hydrochloric acid (10% w/w) to 1 mL of sample. Afterwards, samples were centrifuged at 10,000 rpm for 5 min and the supernatant was used for analysis (Samanidou et al., 2017).

Approximately 1.5 mL of filtered whey sample was placed into sampler vials and sealed for HPLC analysis. HPLC (Prominence HPLC system, Shimadzu LC-20, Torrance, CA, USA) was used for sugar determination, refractive index detector

RID-10A; column SUPELCOSILLC-NH2 (250 mm × 4.6 mm) 5 µm column; 35 °C temperature; gradient mobile phase acetonitrile: deionized water (80:20); volume of the injected sample: 10 µL; flow rate: 1.0 mL min^{-1} (Zolnere et al., 2018).

Table 1. β-galactosidases characteristics

Parameter	Enzyme[1,2]			
	BrennZyme DairyLact	Nola™Fit 5500	Ha-Lactase 5200	GODO-YNL2
Specific activity[3]	5400 oNPGU g^{-1}	5500 BLU g^{-1}	5200 NLU g^{-1}	5000 NLU g^{-1}
Recommended dose	0.5–1.2 mL L^{-1}	0.1–3.3 g L^{-1}	0.5–1.0 mL L^{-1}	0.5–1.0 mL L^{-1}
Amount added in the study	0.5 ml L^{-1}			
Temperature range, °C	5–45	35–50	35–45	4–45
Optimal pH	6.0–7.0	5.4–7.0	6.5–8.0	7.5–8.0
Origin	*Saccharomyces marxianus var. lactis*	*Bacillus licheniformis*	*Kluyveromyces lactis*	*Kluyveromyces lactis*
Producer	BrenntagPolskaSp., Poland	Chr. Hansen, Denmark		Danisco, Denmark

[1]Zolnere & Ciprovica, 2017; [2]Zolnere et al., 2018; [3]Units of β-galactosidase activity are defined differently by each manufacturer; BLU – bifido lactase units; NLU – neutral lactase units; oNPGU – o-nitrophenyl-β-D-galactoside units.

Sweetness and reduction of carbohydrates calculation

Sweetness of yoghurt (1) before sucrose addition and carbohydrates reduction (2) were calculated according the following equitation's:

$$Sw = galactose \times 65 + glucose \times 72 + lactose \times 16 \quad (1)$$

where *Sw* – sweetness of yoghurt; *galactose* – concentration of galactose (%); *glucose* – concentration of glucose (%); *lactose* – concentration of lactose (%).

$$R = 100 - \frac{(galactose_y + glucose_y + lactose_y) \times 100}{lactose_m} \quad (2)$$

where *R* – reduction of carbohydrates (%); $galactose_y$ – concentration of galactose in yoghurt (%); $glucose_y$ – concentration of glucose in yoghurt (%); $lactose_y$ – concentration of lactose in yoghurt (%); $lactose_m$ – concentration of lactose in milk (%).

Questionnaire

Teenagers completed the questionnaire, which consisted of: demographic questions, information about lactose intolerance, yoghurt consumption frequency, teenagers' preference in yoghurt choice.

Sensory evaluation

Sensory evaluation was performed by 50 teenagers as potential low-sugar yoghurt consumers, at the age from 14 to 18, (60% – female, 40% – male), 13% of teenagers have lactose intolerance.

The sensory evaluation has been performed at Aizputes Secondary School (Latvia) in a class with individual tables, the temperature of yoghurt was 16 ± 1 °C.

Before sensory evaluation panellists were instructed about evaluation procedure. Panellists received approximately 30 mL of low-sugar yoghurt sample at 16 ± 1 °C temperature in cups with volume 50 mL, coded with three-digit random numbers. Warm tea was provided to panellists for cleansing their palates between samples.

Five-point JAR (just-about right) method was used to determine mean overall liking of yoghurt samples sweetness. In the five-point JAR scale: 1 – not sweet, 2 – somewhat too weak sweetness, 3 – just about right sweetness, 4 – too much sweet, 5 – somewhat too much sweet.

Seven-point hedonic scale (ISO 11136:2014) was used to determine yoghurt overall liking. In the seven-point scale – 1 – dislike very much; 4 – neithr neither like nor dislike, 7 – like very much.

The data collection and statistical interpretation of data were processed with FIZZ Aquistion Ver.2.51 software (Biosystemes, France).

Data processing

All results were reported as mean ± standard deviation. Statistical analyses were performed using analysis of variance (ANOVA) and mean concentrations of parameters were carried out by Duncan's multiple range test. Differences were considered statistically significant with a confidence interval of $P < 0.05$.

RESULTS AND DISCUSSION

Our previous research (Zolnere & Ciprovica, 2019) confirmed that various concentrations of monosaccharides were obtained after lactose hydrolysis with different β-galactosidases. The lactose hydrolysis data are shown in Table 2.

Contradictory results were obtained with galactose, a significantly higher ($P < 0.05$) concentration of galactose was established in sample with GODO-YNL2, and significantly lower in sample with HA-Lactase 5200.

Lactose splitting by β-galactosidase provides higher carbohydrates (mostly lactose and glucose) reduction rate during fermentation (see Table 2) compared to control. In all samples carbohydrates reduction during hydrolysis and fermentation was significantly higher ($P < 0.05$).

Table 2. Characteristics of fermentation products, their values

Parameters	Control	Nola™Fit 5500	Ha-Lactase 5200	GODO-YNL2	BrennZyme
glucose, g L^{-1}	2.59 ± 0.05^{a}	22.20 ± 0.12^{c}	21.00 ± 0.11^{bc}	18.45 ± 0.11^{b}	21.60 ± 0.08^{bc}
galactose, g L^{-1}	2.84 ± 0.05^{a}	13.90 ± 0.10^{c}	11.70 ± 0.05^{b}	14.00 ± 0.09^{d}	13.10 ± 0.10^{c}
lactose, g L^{-1}	33.01 ± 0.11^{d}	0.007 ± 0.001^{a}	0.09 ± 0.001^{c}	0.04 ± 0.001^{b}	0.03 ± 0.001^{b}
reduction of carbo-hydrates during fermentation, %	16.4^{a}	21.5^{b}	28.7^{d}	29.4^{d}	24.5^{c}
pH	4.7 ± 0.05^{c}	4.4 ± 0.05^{a}	4.5 ± 0.05^{b}	4.5 ± 0.05^{b}	4.5 ± 0.05^{b}
sweetness	0.18^{a}	0.68^{c}	0.66^{b}	0.67^{bc}	0.68^{c}

Means marked with a different letter are significantly different ($P < 0.05$).

Lactose hydrolysis into glucose and galactose helps to increase sweetness through an occurrence of natural sugars in milk (Li et al., 2015; Ohlsson et al., 2017; Zolnere & Ciprovica, 2017; Cheng et al., 2020). Sweetness of analysed samples could be compared to 2.5% of sucrose addition (Harju et al., 2012). However, the sweetness of the product depends on the hydrolysis degree, variety and concentration of occurred monosaccharides and oligosaccharides.

A significantly higher sweetness ($P < 0.05$) was established in all samples with β-galactosidase. A higher sweetness was established in samples with Nola™ Fit 5500 and BrennZyme.

According to the findings of different studies (Rosolen et al., 2015), β-galactosidase application in yoghurt production decreases fermentation time, this fact was proved by pH set during the current study. In all samples with enzyme pH was lower, comparing to the control one (see Table 2). The availability of a higher proportion of easily fermented monosaccharides (mainly glucose) promotes faster growth rate of lactic acid bacteria (Schmidt et al., 2016).

The focus of the questionnaire is to analyse teenagers' preferences in yoghurt choice, yoghurt dietary pattern, sensory properties, etc.

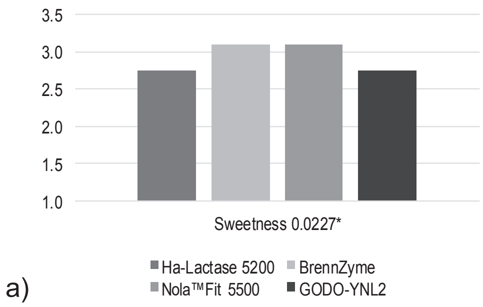
Figure 1. Teenagers' preferences in yoghurt market.

According to the questionnaire results a significant number of teenagers (94%) eat yoghurt at least once per week (59%), 93% of respondents has preferences in sweet strawberry yoghurt consumption (see Fig. 1). Li and co-authors (2015) reported similar results, their study has shown a positive correlation between increased sugar concentration in flavoured milk and dairy product consumption among adults and children. The overall questionnaire results show that teenagers are highly aware of yoghurt consumption. Yoghurt producers should be oriented, particularly on teenagers, by improving product composition, as well as reducing added sugar and lower lactose concentration.

Sensory evaluation of yoghurt samples was done and the results are presented in Fig. 2.

During samples evaluation teenagers admitted low–sugar yoghurt as sweet, significantly sweeter ($P < 0.05$) was yoghurt sample with Nola™ Fit 5500 and BrennZyme (see Fig. 2, a.). The sensory evaluation showed the same tendency in sweetness of yoghurts as in analysed monosaccharides composition and concentration data.

The degree of sweetness correlates ($r = 0.84$) with overall liking degree of yoghurt. The current research results proved the statement that teenagers admitted yoghurt with higher sweetness (Nola™ Fit 5500) as the best one, followed by yoghurt with BrennZyme (see Fig. 2, b).



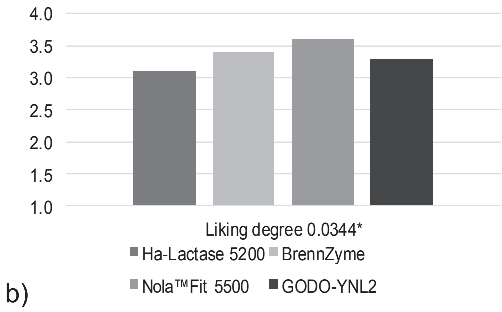


Figure 2. Yoghurt sensory evaluation results: a) Mean overall sweetness liking (JAR); b) Overall liking degree (Seven-point hedonic scale).

*Not significant at 5%.

Taking into account teenagers' preference, obtained results can be evaluated positively, still the largest part of teenagers (85%) has evaluated yoghurts with Nola™ Fit 5500 and BrennZyme as sweet enough. Sugar can be replaced only to a certain extent and the strategical objective should not be a reduction of consumer expectations in terms of sweetness intensity (Rogenhofer & Hauß, 2019).

Lactose hydrolysis could be a possible tool for sugar reduction in yoghurt production from a technological and nutritional point of view. Yoghurt with hydrolysed lactose is sweeter, faster fermented and more readily absorbed from intestine (Rosolen et al., 2015). However, the question: 'Could producers offer such product to lactose tolerant consumers?' is still debatable. The solutions could be the commercial starter, which allows to increase sweetness throught higher hydrolysis of lactose, yielding glucose comparing to classical yoghurt starters.

CONCLUSIONS

Sweetness of yoghurt can be regulated by commercial β-galactosidases. A significantly higher ($P < 0.05$) concentration of glucose was determined in yoghurt with Nola™ Fit 5500 and BrennZyme, which reflected in a higher (calculated and sensory evaluated) sweetness of the product. The results demonstrated that low-sugar yoghurt has gained consumer acceptance as sweet enough.

ACKNOWLEDGEMENTS. Project No 18-00-A01612-000012 'Novel solutions for milk products and their processing by products production' supported by ministry of Agriculture and Rural Support Service of the Republic of Latvia.

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