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Development of formulation and technology of non-dairy soy-coconut yogurt

V. Kosterina[1], A. Yakovleva[2,*], V. Koniaeva[1] and N. Iakovchenko[1]

[1]ITMO University, Faculty of Food Biotechnologies and Engineering, Lomonosova street 9, RU191002, Saint Petersburg, Russia
[2]Saint-Petersburg University of State Fire Service of Emercom of Russia, Department of Economics and Law, Moskovsky prospect 149, RU196105, Saint Petersburg, Russia
[*]Correspondence: frack@mail.ru

Abstract. Yogurt provides an organism with probiotics, which can help digestion. However, many people do not consume dairy for a lot of reasons such as lactose intolerance, milk protein allergy, environmental and ethical concerns. The aim of the research was to develop formulation and technology of plant-based yogurt made of coconut and soy milk. The possibility of using the composition of coconut and soy milk was investigated. The effect of using different ratios of coconut and soy milk on rheological and sensory acceptability was studied. Soy milk containing 20, 30, 40 and 50% coconut milk were used in the production of soy yoghurt using commercially available yoghurt starter. The physico-chemical composition, water activity, rheological and organoleptic properties, fatty acid composition and microorganism viability were investigated. Presented production procedure enables the manufacture of a product with satisfactory functional properties and organoleptic properties.

Key words: soy milk, coconut milk, kudzu, lecithin, soy yoghurt, coconut yoghurt.

INTRODUCTION

People do not consume dairy products because of a number of reasons such as lactose intolerance and malabsorption, milk protein allergy, environmental, ethical and religious concerns.

About two-thirds of the world's population is lactose intolerant (Bayless et al., 2017; Storhaug et al., 2017) and varies widely per country. The lactose malabsorption and intolerance are defined by gastrointestinal symptoms: diarrhea, flatulence, nausea, gut distension, cramps, abdominal pain and discomfort, and systemic symptoms such as headache (Nicklas et al., 2009; Storhaug et al., 2017).

Milk protein allergy is a common problem among infants and young children (Skripak et al., 2007). Besides gastrointestinal symptoms milk protein allergy also involves anaphylaxis symptoms such as skin and respiratory tract allergies or other system disorders (Bahna, 2002).

Globally, 9% of population, on average, decides to cut animal products from their diet and follow a vegan diet (Nielsen, 2016). They are standing against animal cruelty and exploitation, environmental safety and for personal health.

Soy milk is the most nutritionally identical to cow's milk. It is a rich source of proteins, which includes all essential amino acids, low in carbohydrates and fats and does not contain cholesterol. Soy milk contains minerals – iron, calcium, potassium, zinc, phosphorus, copper, manganese, and vitamins B-group, E, K (Messina, 2016). Soy proteins have the highest digestibility compared with other proteins of herbal origin and are rich in lysine, threonine, and tryptophan (Degola et al., 2019).

Isoflavones, founded in soybean, have many health benefits such as cardiovascular protective effects, including reduction of cholesterol and menopause symptoms, prevention of osteoporosis and reduction of risk of developing of certain cancers (prostate and breast cancer). They also possess antioxidant properties and have antiviral and hepatoprotective activities (Garcia et al., 2009; Wu & Kang, 2011; Kant & Albrecht Broadway, 2015). Taking into account the foregoing, food manufacturers are encouraged to incorporate more soy components in food formulations (Nadtochii et al., 2015).

Coconut milk has a sweet taste, delicate coconut flavor and creamy texture. It contains potassium, phosphorus, magnesium, manganese, iron, calcium, zink, vitamins C, E and B-group (Kothalawala et al., 2018; Amirah et al., 2019). Coconut milk is a rich source of antioxidants, low in carbohydrates and high in fiber (Alyaqoubiet et al., 2015).

Despite the fact that coconut milk is full of saturated fats, they are medium-chain triglycerides (MCFAs). It means that they can easily be absorbed and used by organism for energy, improve cognitive function and treat from memory loss (Alyaqoubi et al., 2015). MCFAs are quickly oxidized by the liver and thus less obesogenic than low chain fatty acids (LCFAs). Unlike LCTs, MCTs go straight to the liver where they are either used for immediate energy or turned into ketones (an alternate source of energy for the brain) (Schönfeld & Wojtczak, 2016).

Due to lauric acid content, coconut milk possesses antiviral, antibacterial and antioxidant activities. Including coconut milk in a diet can help to reduce the risk of heart disease, stroke and control hunger (Paul et al., 2019). Fats in coconut raise the high-density lipoprotein (HDL) levels while reducing the low-density lipoprotein (LDL) (Ekanayaka et al., 2013).

Other studies confirmed that addition of coconut milk to soy milk improved the sensory characteristics of the yoghurt, helped mask bitter beany flavour (Kolapo & Olubamiwa, 2012).

Yogurt has many advantages. Fermentationis not only one of the oldest ways to preserve food and beverage, but it also improves nutritional and organoleptic quality, physicochemical characteristics of a product, providing an organism with beneficial microorganisms (Bell et al., 2018).

Soy milk fermentation is not an exception. Microorganisms reduce anti-nutritional factors in soy such as proteinase-inhibitors, phytic acid, urease, oxalic acids, oligosaccharides and increase the bioavailability of bioactive components by means of increasing the number of free isoflavones and peptides (Sanjukta et al., 2015). Fermentation can also reduce beany flavor and improve texture (Niyibituronsa et al., 2018).

According to Nielsen's Global Health and Ingredient-Sentiment Survey, 70% of global respondents say they actively make dietary choices to help prevent health diseases and make dietary changes for health reasons (Nielsen, 2016). Therefore, the aim of this study is to investigate the possibility of using the combination of coconut milk and soy milk in the manufacture of yogurt without artificial ingredients for people who does not consume dairy products.

MATERIALS AND METHODS

Soy milk preparation

Soymilk was prepared using blending method. Soybeans were washed and soaked in clean tap water for 12 h. The swollen soybeans were drained and blended with water at a bean-to-water ratio of 1:10 in a blender at low speed for 10 minutes until smooth. The homogenized mass was strained through a double-layered cheesecloth to separate milk from residue.

Yoghurt Manufacture

Soy-coconut yoghurts were formulated to contain: soy milk containing 20% coconut milk (Aroy-D, Thailand); soy milk containing 30% coconut milk; soy milk containing 40% coconut milk; soy milk containing 50% coconut milk. Hereinafter groups are referred as SMCM20, SMCM30, SMCM40 and SMCM50, respectively. The prepared mixtures were pasteurized at 85 ºC and placed in water bath to cool down to 42 ± 1 °C. The cooled mixtures were inoculated with the starter culture at a rate of 3% (*Lactobacillus delbrueckii subsp. bulgaricus* and *Streptococcus thermophilus*) and fermented at 42 ± 1 °C for 7 hours. The starter was prepared using soy milk. The yogurt samples were incubated to reach the titratable acidity value equals to 0.585% lactic acid. To avoid the coconut milk to rise on the top and stabilize the mixture kudzu root starch (Mitoku Co.) and lecithin (Cargill Inc., Germany) were used in concentrations 2% and 0.5%, respectively. After incubation, the yoghurt samples were cooled and stored at refrigerator at 5 ± 1 °C until used.

Methods

pH values were measured using pH–meter pH–410 with combined glass electrode (Scientific Production Association 'TECHNOKOM', Russia).

Titratable acidity (TA) was measured by titration of 10 g of sample with 0.1 N NaOH solution and expressed in % lactic acid.

Moulds and yeasts were determined according to GOST 10444.12–12013.

Rheological measurements were carried out using Rheotest 2 type rotating viscosimeter (VEB–MEDINGEN, Germany) with the coaxial cylinder device S_2. The rheological measurements were performed at controlled temperature of 20.0 ± 0.5 °C. The shear stress was estimated as the areas between upward flow curve and downward flow curve.

Apparent viscosity η was calculated using the following formula:

$$\eta = \frac{\tau}{\gamma} \quad \text{(Pa s)} \qquad (1)$$

where γ – shear rate, s^{-1}; τ – shear stress, Pa.

The storage time was estimated according to MUK 4.2.1847–04 as described by Dubrovskii et al. (2019). To determine the shelf life of the yogurt, samples were packed in glass containers and stored at 4 ± 2 °C.

Fatty acid composition of yogurt was determined by gas chromatograph (GC-2010, Shimadzu, Tokyo, Japan) equipped with a flame ionization detector and a capillary column DB-23 (60 m × 0.25 mm × 0.25 µm) (Agilent Technologies, Santa Clara, CA, USA). Injector and detector temperatures were set as 250 ºC and 280 ºC, respectively.

Amino acid content was analyzed by HPLC method using HPLC system LC-20AD (Schimadzu, Japan) according to ISO 13903:2005.

Amino acid score of essential amino acids was calculated using the following formula:

$$Amino\ acid\ score = \frac{g\ of\ amino\ acid\ in\ 100\ g\ of\ test\ protein}{g\ of\ amino\ acid\ in\ 100\ g\ of\ protein\ FAO/WHO} \times 100, \quad (2)$$

Coefficient of distinction amino acid scores (CDAAS) (Lipatov, 1995), difference of amino-acid score of an essential amino acid and a minimum amino acid score, and biological value (BV) were calculated using the following equations:

$$\mathrm{CDAAS} = \frac{\sum_{i=1}^{N}(\mathrm{AAS_i} - \text{the lowest AAS})}{\mathrm{N}}, \quad (3)$$

where N is essential amino acids content, and AAS_i is amino acid score of the i-th amino acid (%).

$$\Delta\mathrm{DAAS} = C_i - C_{min}, \quad (4)$$

where C_i – score of I – essential amino acid and C_{min} – minimum amino acid score.

$$\mathrm{BV} = 100 - \mathrm{CDAAS}\ (\%) \quad (5)$$

Mixture stability of control and treatment samples of yogurt was determined by visual observation of the height of the coconut milk layer formed at the top of the test tubes after incubation and during the storage period every 5 days. Produced mixtures were subjected to fermentation in 25 mL test tubes at 45 °C. For each sample there was considered three replicates.

The creaming index (CI) values were obtained from the ratio between the total height of cream layer (CL) and the total height of mixture layer (ML).

$$CI(\%) = \frac{CL}{ML} \times 100 \quad (6)$$

CL and ML were measured directly into a storage test tubes with the help of a graduate scale.

Lactobacillus delbrueckii subsp. Bulgaricus and *Streptococcus thermophilus* were counted according to GOST 33951-2016.

Sensory evaluation based of the finished product on flavour, colour, taste, texture and overall acceptance was conducted by the panel of 24 students and staff of Faculty of Food Biotechnology and Engineering (50% of participants were female from 20 to 70 years old and 50% of participants were male from 21 to 70 years old) by using 9 point hedonic scale (9 = like extremely; 5 = neither like nor dislike; 1 = dislike extremely) according to the method as described by Clark et al. (2009). Yogurt samples were placed in cups which were randomly coded with 3-digit number. The panelists were allowed to use spring water and unsalted cracker for palate cleansing between the samples.

Statistical analysis

The significance of differences in results of each test values was determined using *Student's t-test*. Differences were considered significant at the $P < 0.05$ level. All statistical analyses were carried out with the BioStat 6.9 (AnalystSoft Inc.) and one-way analysis of variance (ANOVA). The results were expressed as mean values and standard deviations (SD).

RESULTS AND DISCUSSION

Yogurt samples with different ratios of coconut milk and soy milk were characterized by their thixotropicbehavior. Fig. 1 shows the flow curves for yogurt. Among the four prepared yoghurt samples, the obtained hysteresis loops showed significant difference. Hysteresis loop was assumed to be the difference between the energy required for structural breakdown and samples possibilities of being reconstructed.

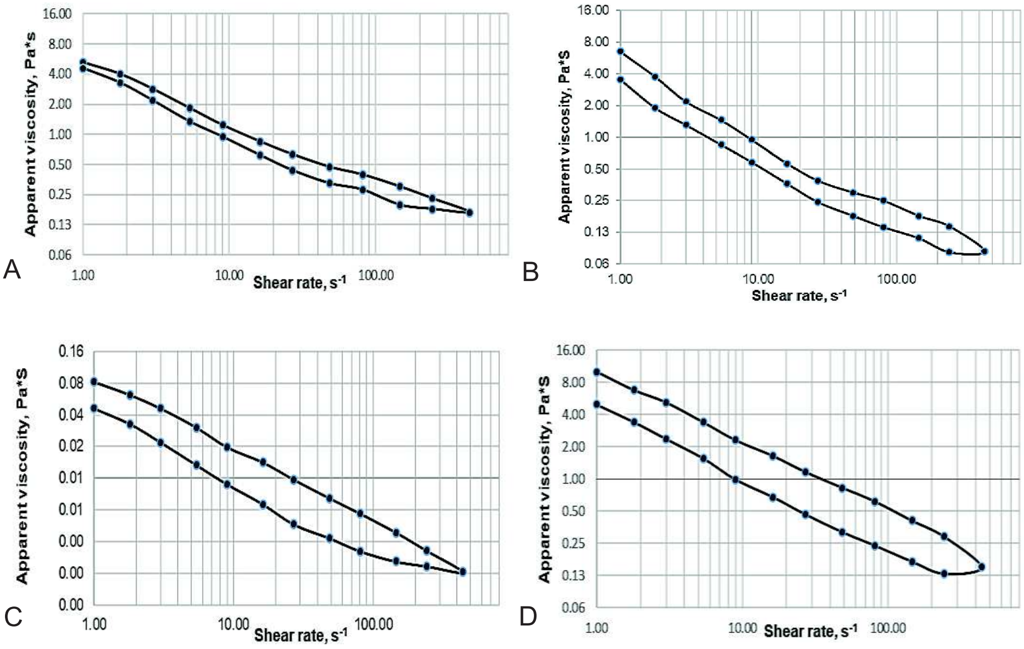

Figure 1. Thixotropy loop of the yoghurt samples: A is yoghurt containing 20% coconut milk and 80% soy milk; B is yoghurt containing 30% coconut milk and 70% soy milk; C is yoghurt containing 40% coconut milk and 60% soy milk; D is yoghurt containing 50% coconut milk and 50% soy milk.

Data presented showed that the thixotropy properties of yogurt samples decreased with the increase in coconut concentration in the mixture. Yaakob et al. (2012) have also found that viscosity of the yogurt is increased with increasing coconut milk concentration.

The hysteresis loop area of the yoghurt sample prepared from with 50% of coconut milk was the largest, while that of the yoghurt sample prepared with 20% of coconut milk was the smallest. The reason of the higher hysteresis area in this sample may be explained by the higher viscosity of the sample containing 50% of coconut milk in contrast to the sample containing 20% of coconut milk. Hence, these samples provided a firmer product because more energy is required to break its structure. The yoghurt sample prepared with 20% of coconut milk tended to have better structural reversibility than that prepared with 50% of coconut milk.

In this work, the indicators that characterize the sustainability of the structure yogurt samples to destruction during mechanical action and its ability to thixotropic recovery, namely, viscosity loss coefficient (VLC), coefficient of mechanical stability (CMS) and indicator of structural recovery (SR) were determined. The data obtained are shown in Table 1.

Analyzing the data presented in Table 1, it can be seen that viscosity loss increased with the increase in coconut milk concentration. The maximum thixotropic recovery showed the sample with 20% of coconut milk.

The data given in Table 1 also show that the decrease in the coconut concentration had no significant influence on the resulting coefficient of mechanical stability of the yoghurt samples, but might have enhanced the indicator of structural recovery of yogurts.

Table 1. Structure stability characteristics of yogurt samples

Coconut milk: soy milk ratio	Indicators		
	VLC(%)	CMS	SR(%)
20:80	9.8 ± 1.2	1.11 ± 0.11	96.7 ± 2.5
30:70	12.5 ± 1.1	1.11 ± 0.11	93.1 ± 2.7
40:60	13.0 ± 1.2	1.16 ± 0.11	86.8 ± 2.7
50:50	23.1 ± 1.2	1.31 ± 0.11	76.9 ± 2.9

Data from the Fig. 2. present the organoleptic characteristics of yogurt samples. It can be seen, that the mean scores for overall acceptance and taste was higher for SMCM30 sample than in the others. Products with higher percent of soy are often less accepted by consumers due to its flavour, identified as bean-likeflavour. Addition of coconut milk in yoghurt formulation improved the flavour evaluation scores. Flavour and texture of SMCM30 and SMCM40 samples presented the same degree of liking by the participants of the organoleptic characteristic's analyses. In terms of colour all yogurt samples were well accepted by the panelists. The result of the sensory evaluation from this study has shown that coconut-soy yoghurt could become a more acceptable to the consumers if an appropriate amount of coconut milk is added to the formula.

Based on the results of the studies of rheological and organoleptic characteristics, for further studies SMCM30 sample was chosen.

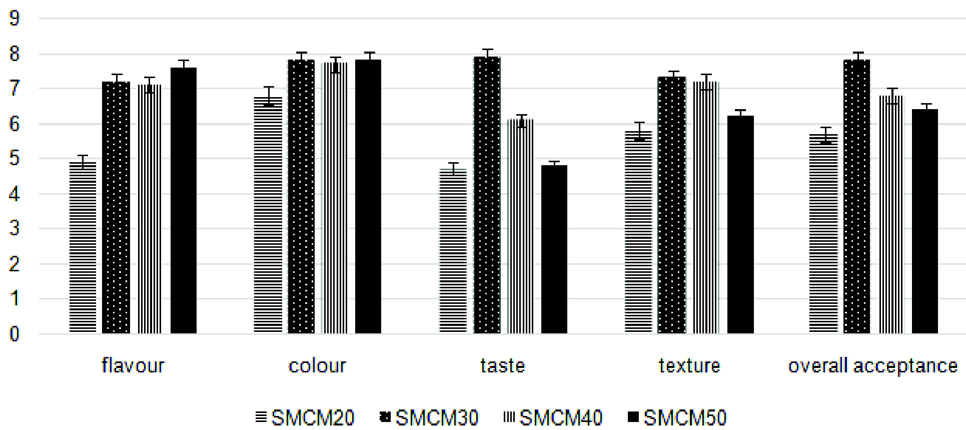

Figure 2. Hedonic scale for flavor, colour, taste, texture and overall acceptability of the samples with different ratios of coconut: soy milk. Values are means ± standard deviation. Data represent significant differences within each sensory attribute between the yogurt samples ($P < 0.05$).

The analyzed samples presented 11 fatty acids, which were identified and quantified (Table 2).

Fatty acids were grouped as follows: short-chain saturated fatty acids (C4-C10, SCFA), medium-chain saturated fatty acids (C12-C15, MCFA), long-chain saturated fatty acids (C16-C24, LCFA), monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA). A total of 11 fatty acids comprised of both saturated and unsaturated fatty acids. As shown in Table 2, 91.3% of the fatty acid were saturated, 8.5% were monounsaturated and 0.3% were polyunsaturated.

Table 2. Fatty acid composition of the vegan yogurt

Fatty acid profile	Group	Fatty acid content from total amount of fatty acids (%)
caproic acid (6:0)	SCFA	0.81 ± 0.07
caprilic acid (8:0)	SCFA	8.13 ± 0.62
capric Acid (10:0)	SCFA	6.02 ± 0.45
lauric acid (12:0)	MCFA	46.04 ± 2.96
myristic acid (14:0)	MCFA	18.12 ± 1.13
palmitic acid (16:0)	LCFA	8.92 ± 0.48
stearic (18:0)	LCFA	3.4 ± 0.21
oleic (18:1)	MUFA (omega-9)	5.8 ± 0.31
linoleic acid (18:2)	PUFA (omega-6)	2.6 ± 0.14
linolenic (18:3)	PUFA (omega-3)	0.3 ± 0.03
arachidic (20:0)	LCFA	0.1 ± 0.01

As can be seen from the Table 2, saturated fatty acids were the predominant fatty acids. The level of caprilic acid (8:0), lauric acid (12:0), myristic acid (14:0) and palmitic acid (16:0) in fermented product were the highest among all fatty acids. The proportion of MCFA in yogurt sample was the highest, followed by SCFA, LCFA, MUFA and PUFA. Increasing of SFA in coconut milk doesn’t lower its health benefits (Bawalan & Chapman, 2006) cleared that coconut oil is unique as it contains the highest percentage of medium chain fatty acids which are metabolized differently in the human body to other saturated and unsaturated fats or oils. They are similar in structure to the fats in mother's milk that provide babies immunity. There are also similar beneficial effects in adults (Kabara, 2000).

The amino acid composition and Amino acid score of the yogurts are presented in Table 3.

Calculated quality characteristics of protein component of soy-coconut yogurt are presented in Table 4.

As can be seen from the Table 3, the developed yogurt contains all essential and non-essential amino acids and the scores for all essential amino acids, except lysine amino acids, are higher than 100%. The low amount of lysine amino acid in yogurt is a consequence of lack lysine in coconut milk (Sousa & Kopf-Bolanz, 2017). Calculation of amino acid scores (Table 4) showed the high biological value of soy-coconut yogurt protein.

Table 3. Amino acid content and score of the coconut-soy yogurt

Amino acid		FAO/WHO, 2007, g 100 g^{-1} of protein	g 100 g^{-1} of protein	Amino acid score of yogurt (%)
essential amino acids, incuding:	amino acids total	27.7	34.9	133.7
histidine		1.5	2.06 ± 0.15	137.2 ± 9.9
isoleucine		3.0	3.46 ± 0.27	115.4 ± 9.2
leucine		5.9	6.38 ± 0.30	108 ± 4.9
lysine		4.5	4.61 ± 0.34	102 ± 7.5
methionine + cysteine		2.2	1.76 ± 0.29	79.8 ± 13.4
phenylalanine +tyrosine		3.8	7.01 ± 0.26	184 ± 6.67
threonine		2.3	3.54 ± 0.29	154 ± 12.6
tryptophan		0.6	1.26 ± 0.26	210 ± 34.5
valine		3.9	4.42 ± 0.30	112.8 ± 7.5

The data given in Table 5 shows that the viability of *Streptococcus thermophilus* and *Lactobacillus delbrueckii subsp. Bulgaricus* decreased in all yogurt treatments at the end of storage periods compared with their counts when fresh. No mould and yeasts were detected in soy-coconut yogurt samples during the first 15 days of refrigerated storage. Analyzing the data, it can be concluded that quality indicators during the shelf life of the samples do not exceed the established parameters for yogurt. The results revealed that considering the reserve ratio according to MUK 4.2.1847–04 the recommended period of storage soy-coconut yogurt is 20 days at 4 ± 2 °C.

Table 4. Quality characteristics of the yogurt

The name of the quality characteristics	The value of the quality characteristics
minimum amino acid score (%)	79.8
ΔDAAS (%)	485.2
CDAAS (%)	53.9
BV (%)	46.1

Table 5. Quality indicators of yogurt

Indicators	Soy-coconut yogurt				
	Storage time, days				
	0	8	15	20	26
titratable acidity, % lactic acid	0.59 ± 0.03	0.62 ± 0.03	0.67 ± 0.02	0.79 ± 0.03	0.81 ± 0.03
Streptococcus thermophilus, CFU mL^{-1}	6.5×10^8	4.6×10^8	2.2×10^8	5.6×10^7	1.5×10^7
Lactobacillus delbrueckii subsp. Bulgaricus	4.8×10^6	2.3×10^6	1.0×10^6	3.6×10^5	2.3×10^5
Yeasts and molds, Log CFU mL^{-1}	-	-	-	0.5 ± 0.1	0.9 ± 0.1

The results of the yogurt stability during the storage period are shown in Fig. 3. Regarding the creaming index data, the lowest CI rates were observed for the formulation of coconut-soy yogurt with addition of kudzu starch and lecithin at the rate of 2% and 0.5%, respectively. Separation of the two phases was almost complete in less than 8 days in all samples. After 25 days, lower CI was observed for the sample with kudzu starch and lecithin and characterized by high stability.

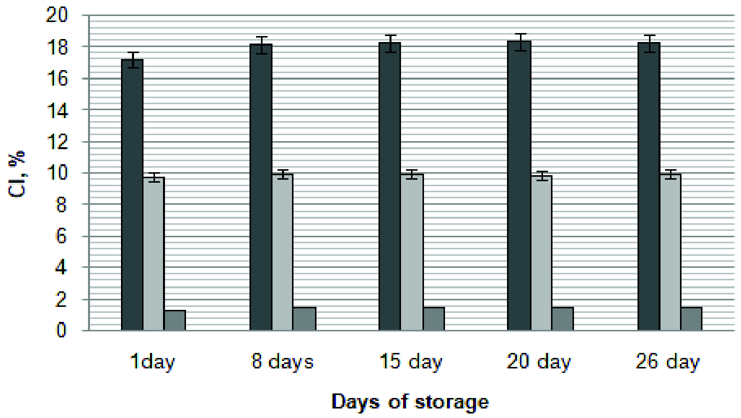

Figure 3. Creaming index versus days: – control sample without kudzu starch and lecithin; – treatment sample with use of kudzu starch; – treatment sample with use of kudzu starch and lecithin. The values represent the mean of three determinations ± standard deviation. The differences were significant ($P < 0.05$).

CONCLUSIONS

Plant-based milk alternatives are a rising trend. In general, they can serve as inexpensive substitutes to cow's milk to those who cannot afford cow's milk because of high cost and limited availability or to those who are allergic to cow's milk. The present research efforts were carried out to create healthy plant-based beverage with high overall acceptability as an alternative to bovine milk which is palatable as well as nutritionally adequate to meet the present demands of consumers.

The results of the study showed that it is possible to produce plant-based yogurt to satisfy organoleptic needs of consumers, especially to people who are lactose intolerant or follow the vegan diet. The developed coconut-soy yogurt is a good source of amino and fatty acids. It is rich in lauric acid which is helpful in boosting immune system and maintaining the elasticity of the blood vessels.

The recommended storage period of yogurt without significant changes in viability of microorganisms is 20 days. Kudzu starch in the amount of 2% and lecithin in the amount of 0.5% can be used to improve the textural properties and stability of yogurt during the storage period without affecting the flavour of the final product.

The results of the study showed that blending two types of plant-based milk allows to obtain the product with sufficiently high nutritive and biological values and expand prospective for health food market.

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