Research Article



Physicochemical, Microbiological, and Sensory Characteristics of Yoghurt Produced by Back-Sloping Fermentation

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Abstract | Yogurt is a functional food that has recently become popular in Indonesia. Making yogurt requires a starter culture, which can be found in liquid or powder form in microencapsulated form. This powdered yogurt starter will be applied using back-sloping fermentation for the practicality of the yoghurt-making process. This research aimed to determine how many cycles the back-sloping fermentation using microencapsulated yogurt (MY) starter was applied and evaluate the quality of yogurt in each back-sloping fermentation cycle. The lactic acid bacteria contained in MY starter were *Lactobacillus plantarum* TMW 1.1623 and *Streptococcus thermophilus* in a 1:1 ratio. The research results showed that yogurt made from ultra-high temperature milk UHTM had a water content of 84.18–86.12%, protein content of 3.52–3.78%, a fat content of 3.34–3.38%, pH 4.72–5.05, titratable acid (TTA) 0.74-1.24%, total lactic acid bacteria (LAB) 7.69–12.07 log CFU/mL, viscosity 285.15–2417.9cP, and Syneresis 0.00-0, 14%. Yoghurt made using fresh cow's milk (FCM) had the following characteristics: water content 84.02–86.18%, protein content 3.43–3.74%, fat content 3.32–3.38%, pH 4.13–4.44, TTA 1.77-2.06%, total LAB 10.32–10.76 log CFU/ml, viscosity 2430.15–4497.55cP, and Syneresis 0.00–0.07%. This research concluded that using MY starter for making yogurt using the back-sloping fermentation method could be used for up to seven and nine cycles for yogurt made from UHTM and FCM. The yogurts produced all meet probiotic drink standards with LAB content exceeding WHO standards, namely LAB content of more than 7 log CFU/ml.

Keywords | Starter, Lactic acid bacteria, Yogurt, Back-sloping fermentation

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INTRODUCTION

One of the most popular functional foods today is yogurt, which contains probiotics. Probiotics are dietary supplements in the form of live microbes that benefit their hosts' health (Lee *et al.*, 2022). In addition, probiotics can have a physiological impact on health, such as preventing coronary heart diseases (Pato *et al.*, 2005), various digestive tract problems (Howarth and Wang, 2013), and lowering triglycerides (Kassaian *et al.*, 2019). The positive function of probiotics in maintaining health, it is suggested the need to add probiotics to various foods or drinks (Göçer *et al.*, 2023). To meet these criteria, the minimum therapeutic level of LAB is 10⁶ CFU per mL or g of a food product when consumed (Ajlouni *et al.*, 2021).

Among many variations of probiotic drinks, the most popular and long-known is yogurt, with its distinctive aroma and sour taste (Sarkar, 2019). Yogurt can be made from various types of milk, such as goat's milk, and cow's milk (Ribeiro *et al.*, 2023; Aritonang *et al.*, 2019), soy milk (Delgado *et al.*, 2019), with the addition of a pure culture or a mixed culture of lactic acid bacteria/LAB (Utami *et al.*, 2020). The bacteria commonly used in yogurt starters are *Lactobacillus bulgaricus*, *Streptococcus thermophilus* (Chen and Hang, 2019) or LAB strains such as *L. plantarum* TMW 1.1623 (Rossi *et al.*, 2021; Riftyan *et al.*, 2022).

Starter cultures are available in various forms, such as liquid and powder. However, the liquid one requires special treatment and has a relatively high possibility of contamination, while using powdered starter culture may facilitate treatment and can be used at any time (Huang *et al.*, 2014). This bacterial culture can help fermentation so that changes in chemical composition occur, resulting in distinctive sensory properties. The starter culture of yogurt powder can be produced with a combination of cryoprotection methods by adding a coating material and freeze-drying to produce a microencapsulated starter. The microencapsulated starter can be recultured and used as a starter in making yogurt with the Back-Sloping fermentation method, thus saving the cost and time of yogurt production.

Several studies on making yoghurt using the back slopping method have been carried out. According to research by Olukotun et al. (2021) back slopping yoghurt using Streptococcus thermophilus and Lactobacillus bulgaricus was able to survive up to the 3rd cycle based on pH value, total LAB, lactose content, syneresis, and yoghurt sensory. According to research by Syafitri et al. (2022) back slopping yoghurt using Streptococcus thermophilus and Lactobacillus bulgaricus stored for 24 hours at a temperature of 27–30°C produces yoghurt with a pH of 5, white in color, slightly sour in taste, and thick in texture. The use of microencapsulated Streptococcus thermophilus and Lactobacillus plantarum TMW 1.1623 as a yoghurt starter using the back sloping method has not yet been carried out. In addition, starter cultures must contain certain probiotic microorganisms that play a role in fermentation (Salminen and Wright, 2011). This research aimed to determine how many backslopping fermentation cycles of MY starter can be applied for producing yogurt and to evaluate the quality of yogurt in each back-slopping fermentation cycle on UHTM and FCM.

MATERIALS AND METHODS

STRAINS AND CULTURES CONDITIONS

The modified deMan Ragosa Sharpe (MRS) broth (Indonesian Patent registered No.: S00202107566)

MICROENCAPSULATION YOGHURT STARTER PRODUCTION

pure cultures were then incubated for 24 hours at 37°C.

To prepare the starter, 100 ml of low-fat milk was placed in an Erlenmeyer flask and covered with aluminum foil. It was then sterilized at a temperature of 115°C for 15 minutes and subsequently cooled to a temperature of approximately 37°C. *Lactobacillus plantarum* TMW 1.1623 and *S. thermophilus* were inoculated separately in jars, with each bacteria constituting 3% of the total. The jars were then incubated at a temperature of 37°C for 24 hours.

The freeze-drying process refers to a modification by (Rossi *et al.*, 2021). Following homogenization, 200 ml of yogurt starter (consisting of a 1:1 ratio of *L. plantarum* TMW 1.1623 and *S. thermophilus*) was mixed with skim milk (25% w/v). The mixture was then frozen at -20°C for 24 hours and subsequently dried using a freeze vacuum drier (Buchi: Lyovapor L-200) at a pressure of 0.036 psi for 96 hours. The desiccated starter was subsequently pulverized using a blender to produce the powdered starter.

YOGURT PRODUCTION

The preparation of yogurt refers to (Rossi *et al.*, 2021) which was modified, where UHTM milk or fresh cow's milk (FCM) was put into an erlenmeyer as much as 200 mL then, the milk was pasteurized at 7585°C for 15 minutes and then cooled to a temperature of 35–37°C. The pasteurized milk was then added with 5% (b/v) skim milk 7% (b/v) granulated sugar and 5% (v/v) activated yogurt starter, homogenized, and then incubated in an incubator at 37°C until it reached pH 4.0–4.8. The resulting yogurt is partially used as a starter (F1) for the next yogurt-making (back-sloping fermentation). yogurt making is stopped if the yogurt does not meet the quality requirements of a probiotic drink.

The initial fermentation sample (C0) was used to inoculate pasteurized UHTM milk and fresh milk in separate batches. This new batch was then fermented for 8 hours, resulting in the production of another batch cycle called back slope sample 1 (C1). Batch fermentation was repeated until the n^{th} cycle (Cn). This back-sloping cycle was stopped if the quality characteristics of the yogurt were not achieved.

PARAMETER OBSERVED DETERMINATION OF MOISTURE, PROTEIN AND FAT CONTENT

The moisture, protein and fat content of yogurt were

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measured using methods described in AOAC (2016). The gravimetric method developed was used to determine the moisture content. The Kjeldahl method used to analyze protein content involves quantifying the overall nitrogen content present in the sample. The fat content was determined using Soxhlet extraction using benzene as a fat solvent.

DETERMINATION OF PH

The pH of the yoghurt in all batches, including the control, was assessed using a portable pH meter equipped with a glass electrode after 8 hours of fermentation (Rossi *et al.*, 2022).

TOTAL TITRABLE ACID

The TTA value was determined using the AOAC (2016) recommendations. A 10 ml sample was transferred into an Erlenmeyer flask, and then the PP indicator was added and mixed by shaking. Subsequently, the solution was subjected to titration using a 0.1 N solution of sodium hydroxide (NaOH). The titration proceeded until the color transition occurred, indicating the equivalence point. The recorded volume utilized for the titration was documented.

TOTAL LACTIC ACID BACTERIAL

Total LAB Colonies were carried out as follows (Riftyan *et al.*, 2022). One mL of the sample was put into 9 mL MRS Broth and vortexed until it turned homogeneous. 0.1 mL dilution was taken into an Eppendorf tube containing 0.9 mL of MRS Broth media. The dilution was carried out up to a dilution of 10-7. At the last dilution, 0.1 mL was taken and then planted by the spread method onto MRS Agar media and flattened with a hockey stick. The sample was then stored in an anaerobic jar and incubated at 37°C for 48 hours. The colony calculation results were multiplied by 10.

VISCOSITY

Viscosity testing refers to Riftyan *et al.* (2022). Yogurt viscosity was measured using a Brookfield viscometer by dipping the cleaned spindle no 6 into a glass jar containing 100 ml of sample. The spindle will rotate and measure the viscosity of the sample. The results of the viscosity value can be seen directly on the screen with units of cP.

Syneresis

The syneresis test refers to (Tavakoli *et al.*, 2018). A 20 g sample of yogurt was poured into a centrifugal tube and put into a centrifuge at 500 rpm for 5 minutes. The transparent supernatant was then transferred into a small beaker and the volume was determined using a pipette.

SENSORY EVALUATION

Sensory evaluation was carried out descriptively. The

panelists who tested were semi-trained panelists totaling 30 peoples who had passed standard tests for sensitivity to smell, taste, vision, and verbal ability. The panelists were semi trained who were lecturers and students, male and female from the Agricultural Products Technology Study Program at Riau University. The panelists were requested to evaluate each sample (20 ml) using the questionnaire form provided.

EXPERIMENTAL DESIGN AND DATA ANALYSIS

This study used a completely randomized design (CRD) with three replications for each treatment. The treatment refers to Olukotun *et al.* (2021), where yogurt was made using MY starter as the parent starter (cycle 0) to the nth derivative (cycle). The data obtained from the observation were analyzed statistically using a one-way analysis of variance (ANOVA). If the calculated F count was greater than or equal to the F table, proceed with Duncan's Multiple Range Test at the 5% level. The data were analyzed using SPSS version 23 software.

RESULTS AND DISCUSSION

MOISTURE, PROTEIN AND FAT CONTENT

The observation of yogurt made using MY starter with a back-sloping fermentation method (Table 1) had a significant effect (P<0.05) on the moisture, protein and content of yogurt using UHTM milk or fresh cow's milk (FCM). On the other hand, the fat content of yogurt was not significantly influenced (P>0.05) by the back-sloping fermentation cycle.

The average moisture content of yogurt can be seen in Table 1 ranging from 84.02–86.03%. The use of MY starter up to the sixth cycle with back-sloping fermentation produced yogurt with water content that was not significantly different from the water content of control yogurt (C0) using MY starter for both types of yogurt (UHTM and FCM). The moisture content of this yogurt was almost the same as the moisture content of yogurt from research Riftyan *et al.* (2022).

From Table 1, the average protein content of yogurt made with MY starter control (C0) and derivatived MY starter with back-sloping fermentation method was 3.52-3.78%and 3.43-3.74 respectively for yogurt with UHTM milk and fresh cow's milk as raw materials. The amount of protein in UHTM yogurt made in treatments C0 and C1 was higher (P<0.05) than the amount of protein in yogurt made in treatments C2–C7. On the other hand, the amount of protein in FCM yogurt went down at the start of the 8th back-sloping fermentation cycle up to 9. The more MY starter cycles were used in making yogurt, the protein content decreases. This decrease in protein

Table 1: The nutritional content of the yogurt made by back-sloping fermentation.

Cycle of BSP	Moisture (%)		Pr	otein (%)]	Fat (%)	
	UHTM	FCM	UHTM	FCM	UHTM	FCM	
C0	84.02 ^a ±0.99	84.02ª±0.99	$3.78^{b} \pm 0.28$	$3.71^{\rm b} \pm 0.28$	3.35±0.21	3.38±0.02	
C1	84.08 ^a ±1.02	84.08ª±1.02	$3.71^{b} \pm 0.20$	$3.69^{b} \pm 0.20$	3.37 ± 0.02	3.35±0.07	
C2	84.28ª±0.34	84.28ª±0.34	$3.69^{ab} \pm 0.06$	$3.68^{b} \pm 0.06$	3.35±0.18	3.34±0.12	
C3	84.22ª±0.22	84.22ª±0.22	$3.68^{ab} \pm 0.21$	$3.66^{b} \pm 0.21$	3.38±0.11	3.36±0.18	
C4	84.41ª±0.43	84.41ª±0.43	$3.69^{ab} \pm 0.28$	$3.67^{b} \pm 0.28$	3.35 ± 0.16	3.35±0.22	
C5	$85.25^{ab} \pm 0.24$	84.55 ^a ±0.24	$3.62^{a} \pm 0.20$	$3.65^{b} \pm 0.20$	3.34±0.12	3.35±0.19	
C6	85.46 ^{ab} ±0.46	85.46 ^{ab} ±0.46	$3.64^{a} \pm 0.06$	3.63 ^{ab} ±0.06	3.36±0.09	3.37±0.12	
C7	85.03 ^b ±0.16	86.03 ^b ±0.16	$3.66^{a} \pm 0.21$	$3.60^{ab} \pm 0.21$	3.35 ± 0.02	3.35±0.34	
C8		86.08 ^b ±0.46		3.58ª ±0.06		3.37±0.42	
C9		86.12 ^b ±0.32		3.63ª ±0.21		3.32±0.38	

The superscript letters in the same column indicate significant differences (p < 0.05). Value was expressed in mean ±standard deviation (n=3) UHTM=Ultra high-temperature milk, FCM= Fresh cow milk, BSF= back-sloping fermentation, C=cycle.

Table 2: The pH, TTA and total LAI	of the yogurt made	by back-sloping	fermentation.
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Cycle of BSF	рН		Total tritable acid (%)		Total LAB (log CFU/mL)	
	UHTM	FCM	UHTM	FCM	UHTM	FCM
C0	$4,72^{a} \pm 0,11$	4,13 ^b ± 0,03	$1,24^{\circ} \pm 0,09$	$2,05^{\text{b}} \pm 0,10$	$12,07^{\circ} \pm 0,06$	$10,32^{a} \pm 0,07$
C1	$4,79^{\rm b} \pm 0,09$	4,12 ^b ±n0,04	$1,22^{\circ} \pm 0,05$	$2,01^{ab} \pm 0,10$	$11,82^{de} \pm 0,33$	10,76 ^a ± 0,24
C2	$4,84^{\circ} \pm 0,11$	4,20 ^b ± 0,03	$1,19^{\circ} \pm 0,05$	$2,06^{\text{b}} \pm 0,09$	$11,74^{de} \pm 0,15$	$10,76^{a} \pm 0,25$
C3	$4,84^{\circ} \pm 0,10$	4,20 ^b ± 0,06	$1,17^{\circ} \pm 0,07$	$2,01^{ab} \pm 0,10$	$11,71^{de} \pm 0,03$	$10,74^{a} \pm 0,13$
C4	4,85° ± 0,11	$4,18^{\rm b} \pm 0,05$	$1,17^{c} \pm 0,01$	$2,05^{\text{b}} \pm 0,09$	$11,43^{d} \pm 0,05$	$10,40^{a} \pm 0,13$
C5	$5,02^{d} \pm 0,06$	4,19 ^b ± 0,06	$0,92^{\rm b} \pm 0,02$	1,96 ^{ab} ± 0,09	10,61° ± 0,35	$10,52^{a} \pm 0,13$
C6	$5,05^{d} \pm 0,05$	$4,20^{\text{b}} \pm 0,01$	$0,77^{a} \pm 0,05$	1,99 ^{ab} ± 0,05	$9,73^{\rm b} \pm 0,50$	10,73 ^a ±0,23
C7	$5,03^{d} \pm 0,07$	$4,42^{a} \pm 0,00$	$0,74^{a} \pm 0,04$	$1,86^{a} \pm 0,05$	$7,69^{a} \pm 0,14$	10,64 ^a ±0,65
C8		$4,41^{a} \pm 0,01$		$1,89^{a} \pm 0,16$		$10,47^{a}\pm0,28$
C9		$4,44^{a} \pm 0,09$		$1,77^{a} \pm 0,10$		10,53ª ±0,39

Note: The superscript letters in the same column indicate significant differences (p < 0.05). Value was expressed in mean ±standard deviation (n=3) UHTM= Ultra high-temperature milk; FCM= Fresh milk; BSF= back-sloping fermentation; C=cycle.

may be due to changes in the composition of nutritional content and metabolites due to metabolic activity, so that the growth and development of LAB will be disrupted (Lee snd Salminen, 2019). Lactic acid bacteria were single cells, and each cell's dry weight contains the most organic compounds, namely proteins (Nelson and Cox, 2005).

Table 1 showed that different back-sloping fermentation cycles produced fat levels that were not significantly different (P>0.05) between treatments for both UHTM and FCM yogurt. The increase in the cycle did not change the fat content, where the main starter components are LAB and milk, the main compound of which is protein. The fat content in this study ranged from 0.30-0.41%, which is relatively the same as the Indonesian national standard (SNI) for yogurt No. 01-2981-1992 whose fat content ranges from 3.0-3.8%. This fermentation method can cause changes in the microbial composition of the initial to final product, making it difficult to maintain

physicochemical and nutritional characteristics over time (Kim et al., 2018).

POTENTIAL HYDROGEN TRITABLE ACID, AND TOTAL OF LACTIC ACID BACTERIA OF YOGHURT

The values of pH, TAT, and total LAB yogurt made from UHTM and FCM with different cycle starters of BSF were presented in Table 2. The average pH value of yogurt made from UHTM milk ranged from 4.72-5.03, while the pH of yogurt made from fresh cow's milk ranged from 4.13-44. In both types of yogurt, it shows that the pH value is relatively almost the same between the control (C0) to the fifth cycle (C1-C5) of BSF, for the next BSF cycle, there is an increase in pH value. The change in yogurt pH due to BSF was almost the same as Olukotun *et al.* (2021) research, which used the BSF method up to the fourth cycle. The back-slopping method can accelerate the fermentation process by introducing an active LAB source into fresh milk. This results in a faster decrease in

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pH compared to spontaneous fermentation. According to Hakimi *et al.* (2018), many factors affect the pH of yogurt, including incubation time and fat concentration. The pH of yogurt made from FCM is almost the same as the pH of yogurt from Labbo *et al.* (2021) research using back-sloping fermentation with a pH of 4.35_4.72.

The average TAT of yogurt formed in this study was 0.74-1.19% and 1.77-2.05% respectively for yogurt made from UHTM and FH milk. The TAT value in the C0-C5 treatment was significantly different (P<0.05) from the TAT in the C6 and C7 treatments in yogurt from UHTM milk. The same pattern can also be seen in TAT of yogurt made from FCM. The pH of yogurt has an inverse relationship with its TTA, indicating that the acidity of yogurt is due to the formation of many organic acids from the fermentation process. The TTA value in this study is in line with the results of Matela *et al.* (2019) the research which was 0.69–1.81, the TTA value was directly proportional to the acidity of yogurt.

The total LAB of yogurt made from UHTM milk in treatment C9 (7.69 log CFU/mL) was the lowest total LAB compared to the other treatments. The low total LAB in yogurt from UHTM milk in treatment C7 was due to the seventh cycle of using MY starter; there may be changes in the ratio of LAB and the number of primary metabolites produced. The changes in yogurt in previous cycles impacted the total LAB in this yogurt. Yogurt made using FCM, had a total LAB content that was not significantly different (P>0.05) for all treatments, with an amount of 10.32_10.76 log CFU/mL. The difference in total LAB results between the two yogurts was due to the biological response of each LAB contained in the starter to the substrate, which is different in different cycles. All treatments in this study, either using MY starter as a

control (C0) or using MY starter with the back-sloping method, resulted in a total LAB greater than 7 log CFU/mL. The number of LAB >7 log CFU/mL has met the standard for probiotic drinks (FAO/WHO, 2006).

VISCOCITY DAN WATER HOLDING CAPACITY YOGHURT

The viscosity and syneresis values of yogurt made from UHTM and FCM milk by the back-sloping fermentation method were presented in Table 3. The results of this study showed.

That the use of MY starter (control/C0) and MY starter with different back-sloping fermentation cycles produced yogurt with significantly different viscosity and syneresis values (P<0.05). Table 3 showed that the viscosity of yogurt from UHTM milk in the C0 treatment was significantly different (P<0.05) from the viscosity in the other treatments, with an average viscosity of 285.15– 2417.23 cP. The difference in viscosity of yogurt was caused by the biological response of different LAB, besides the factors that affect viscosity include pH and fermentation time (Routray and Mishra, 2011).

Water holding capacity (WHC) was a characteristic that affects the texture and consistency of yogurt. The WHC of yogurt is the ability to hold water due to the formation of curdles during the fermentation process. The WHC of yogurt from UHTM milk and FCM milk was significantly different (P<0.05) between treatments (C0-C7). The lowest WHC in yogurt from UHTM milk and FCM milk was found in treatment C0, which was 16.11 and 26.02% for yogurt made from UHTM milk and FCM milk, respectively. The viscosity increases as the WHC decreases, which was attributed to the duration of fermentation and the rate of acidity (Saccaro *et al.*, 2009).

Table 3: Viscosity	and water holding	capacity of the yogurt	made by back-slopir	ng fermentation.
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Cycle of BSF	Viscosi	ty (%)	WHC (%)		
	UHTM	FCM	UHTM	FCM	
C0	$2417.23^{f} \pm 0.38$	4497.55° ± 0.95	16.11 ^a ± 0.57	$26.02^{f} \pm 0.89$	
C1	1109.03° ± 0.67	$3628.70^{d} \pm 0.10$	$19.00^{\rm b} \pm 0.68$	33.11°± 1.73	
C2	$753.07^{d} \pm 0.91$	2656.30 ⁱ ± 1.90	38.79° ± 0.61	$30.85^{de} \pm 0.85$	
C3	$418.70^{\circ} \pm 0.92$	$3550.50^{\rm f} \pm 0.10$	$44.26^{d} \pm 0.46$	41.24 ^a ± 0.82	
C4	285.08 ^a ± 0.58	$2430.50^{j} \pm 0.70$	$44.39^{d} \pm 0.93$	$32.20^{cd} \pm 0.41$	
C5	285.98 ^{ab} ± 0.78	$2872.70^{\rm h} \pm 0.50$	$45.10^{d} \pm 0.46$	$39.61^{b} \pm 0.11$	
C6	$287.00^{b} \pm 0.30$	3615.20° ± 1.70	46.31° ± 0.57	$29.50^{\circ} \pm 0.16$	
C7	285.15 ^a ± 0.55	$4070.00^{\circ} \pm 0.00$	46.73° ± 0.93	41.45ª± 0.87	
C8		4265.55 ^b ± 1.25		$30.19^{\circ} \pm 0.17$	
C9		$2926.50^{\text{g}} \pm 0.50$		23.99 ^g ± 0.55	

Note: The superscript letters in the same column indicate significant differences (p < 0.05). Value was expressed in mean ±standard deviation (n=3), BSF= back-sloping fermentation; C=cycle; UHTM= Ultra high-temperature milk; FCM= Fresh cow milk; WHC=water holding capacity.

Table 4: Sensory evaluation of the yogurt made by back-sloping fermentation.

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Cycle of BSP	Color		Flavor	Flavor		Taste		Texture	
	UHTM	FCM	UHTM	FCM	UHTM	FCM	UHTM	FCM	
C0	$3.20^{b} \pm 0.88$	$3.17^{ab} \pm 0.99$	$3.92^{b} \pm 0.58$	4.00°±0.45	$3.16^{b} \pm 0.48$	2.53±0.82	4.00°±0.62	4.07±0.58	
C1	2.68ª±0.81	$3.56^{b} \pm 0.78$	$3.92^{b} \pm 0.50$	3.88°±0.45	2.96ª±0.20	2.60±0.58	$3.52^{b} \pm 0.51$	3.88±0.58	
C2	2.72ª±0.69	2.84ª±0.99	3.16ª±0.54	$3.52^{ab} \pm 0.78$	$3.00^{b} \pm 0.29$	2.52±0.59	3.32 ^b ±0.48	3.52 ± 0.66	
C3	2.40ª±0.49	$3.20^{ab} \pm 0.49$	2.84ª±0.67	3.24ª±0.51	2.80ª±0.40	3.04±0.20	2.80ª±0.40	3.36±0.49	
C4	2.36ª±0.48	2.92ª±0.89	3.16ª±0.37	3.32ª±0.55	2.62ª±0.50	3.12±0.43	2.53ª±0.57	3.36±0.48	
C5		$3.04^{ab} \pm 0.87$		$3.52^{ab} \pm 0.70$		2.96±0.53		3.64±0.62	
C6		$3.32^{ab} \pm 0.79$		$3.52^{ab} \pm 0.85$		3.08±0.48		3.48±0.57	
C7		$3.40^{ab} \pm 0.75$		3.92°±0.84		2.96±0.45		3.20±0.75	
C8		$3.40^{ab} \pm 0.80$		3.40 ^{ab} ±0.69		3.16±0.73		3.92 ± 0.74	
C9		3.20 ^{ab} ±0.69		3.36ª±0.62		3.20±0.49		3.60±0.63	

Note: The superscript letters in the same column indicate significant differences (p <0.05). Value was expressed in mean ±standard deviation (n=3) UHTM=Ultra high-temperature milk, FCM= Fresh milk, BSF= back-sloping fermentation, C=cycle. Color descriptive score 1= very yellow, 2= yellow, 3= yellowish white, 4= white, 5= very white. Flavor descriptive score 1= Very milky flavored. 2= Milk-scented, 3= Somewhat yoghurt-scented, 4= yoghurt-scented, and 5= Very yoghurt-scented. Taste descriptive score 1= very sour. 2= sour, 3= sweet acidity, 4= sweet, and 5= very sweet. Texture descriptive score 1= very liquid 2= liquid, 3= slightly thick, 4= thick, and 5= very thick.

SENSORY EVALUATION TEST

The descriptive assessment given by the panelists to the color of UHTM and FCM yogurt ranged between 2.36–3.2 and 2.92–3.40 (yellowish-white) and 2.92 and 3.40 (yellowishwhite), respectively, for the color of UHTM and FCM yogurt (Table 4). This data shows that the difference in the back-sloping cycle from 0 to 1 fermentation only slightly affects the yoghurt color from yellowish-white to yellow in UHTM yoghurt. This may be due to the UHTM milk processing process causing relatively small color changes. On the other hand, FCM yogurt has relatively the same color (yellowish-white) in each back-sloping fermentation cycle. Flavor perception is a complex phenomenon and traditionally.

Flavor perception is a multifaceted process and conventionally includes aspects of odor, taste, and touch, as explored extensively by Reineccius (2005). The results showed that the utilization of different cycles of back-sloping fermentation exerted a substantial impact on the flavor of yogurt. The results showed that an increase in the backsloping cycle caused changes in the metabolites produced by LAB, such as organic acid, hidrogen, bacteriocyn, and other metabolites. According to Routray and Mishra (2011) the flavor of fermented dairy products are distinguished by a numerous of volatile bacterial metabolites, many of which were the result of lactic acid fermentation or were generated through other reaction pathways. The changing of yogurt flovor in this study could be due to changes in the LAB population ratio in yogurt. Yogurt flavor was a sophisticated biochemical process that can change depending on the type and composition of lactic acid bacteria (LAB) used as a starter culture (Chen et al., 2017).

The taste and texture scores of UHTM yogurt decreased significantly (P<0.05) with increasing back-sloping fermentation cycles, but for FCM yogurt the taste and texture did not differ in each back-sloping fermentation cycle. Many factors influence the texture of this yoghurt. In general, many factors influence the aroma, taste and texture of yoghurt, including variations in the total amount of LAB for each strain, the nutritional composition of the starter. The MY starter used in back-sloping fermentation for each cycle will change the nutritional composition and total LAB in each strain. Research results like this were also obtained by Eissa *et al.* (2011).

CONCLUSIONS AND RECOMMENDATIONS

This research concludes that the back-sloping fermentation method can be used up to the seventh and ninth cycles for yogurt made from UHTM milk and fresh milk with the same physicochemical and microbiological characteristics as yogurt using microencapsulated yogurt starter. The results of the descriptive sensory evaluation show that back-sloping fermentation can maintain color, flavor, taste, and texture similar to conventionally made yogurt up to the fourth and 9th cycles respectively for yogurt made from UHTM milk and fresh milk. This research showed that back-sloping fermentation offers greater opportunities for the small-scale yogurt manufacturing industry to reduce production costs. By purchasing a microencapsulated yogurt starter once to ferment milk, the resulting fermented milk (yogurt) can be reused to make the next yoghurt until it can be used to make yoghurt for 4-9 cycles.

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NOVELTY STATEMENT

The novelty of this research is the use of microencapsulated *Lactobacillus plantarum* TMW 1.1623 and S. thermophilus as yogurt starters. The back-sloping fermentation method up to 5–9 cycles each for yogurt made from UHT milk and fresh cow's milk still meets probiotic drink standards and still maintains color, aroma, taste, and texture.

AUTHOR'S CONTRIBUTION

ER was involved in the design of this research. The other authors were collaboratively involved in carrying out the research, performing statistical analysis, and composing this article. The completion of this manuscript was accomplished in cooperation with FR and UP.

CONFLICT OF INTEREST

The authors have declared no conflict of interest.

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