

The characterization of physicochemical, microbiological and sensorial red ginger yogurt during fermentation

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Abstract

This study was aimed to determine the effect of red ginger extract concentration (0.2, 0.4, 0.6, 0.8, 1% v/v) and fermentation time on the quality and acidity of yogurt. Yogurt was made from fresh cow milk, *Lactobacillus bulgaricus* and 2% v/v *Streptococcus thermophilus*. Operation conditions included fermentation time of 60 hrs and data retrieval every 12 hrs. The results of this study had shown that pH of yogurt was decreasing, while titratable acidity increased during the fermentation time. The addition of red ginger extract did not give a significant effect on the protein content of yogurt. Yogurt with higher concentration of ginger extract increased the spicy aroma and flavor, however its color was similar to the commercial yogurt. At higher concentrations, red ginger displayed antimicrobial and antifungal characteristics, as shown by the decreasing microorganisms counts.

1. Introduction

Yogurt is a fermented milk product consumed widely worldwide. It could be made from homogenized milk, low-fat milk or skimmed milk (Abubakar *et al.*, 1998; Baba *et al.*, 2018). During the fermentation process, lactose in milk is converted to lactic acid by lactic acid bacteria *Streptococcus thermophilus* and *Lactobacillus bulgaricus* (Amadou *et al.*, 2018). Furthermore, sugars in milk are also hydrolyzed by those bacteria, resulting in conversion of lactose to lactic acid. Meanwhile, *S. thermophilus* is responsible for its flavor (Adolfsson *et al.*, 2004; Tamime and Marshall, 2007). Yogurt contains protein, probiotics, minerals and vitamins that are essential for the human body (Buttris, 1997; IHEMEJE *et al.*, 2015). Perdigeon *et al.* (1998) stated that yogurt consumption promotes good health, including reduces the risk of intestinal disease and promotes gut health. Heymans (2000) and Leblanc and Pedrigo (2004) also supported the study further by concluding that yogurt also lowers cholesterol levels in the blood, increases body immunity and assists in protein, calcium and iron assimilation.

The quality of yogurt depends on its physicochemical, microbiological and sensorial

characteristics (Yang *et al.*, 2012). Natural flavors (commonly fruits) are preferable compared to synthetic ones as it further increases the health benefits of consuming yogurt. Fruit and yogurt have been identified individually as indicators of healthy dietary patterns. Fruits are relatively low in energy density and are an excellent source of antioxidants and prebiotic fibers and polyphenols, which can promote digestive health. Combining the intake of yogurt and fruit could provide probiotics, prebiotics, high-quality protein, important fatty acids, and a mixture of vitamins and minerals that have the potential to exert synergistic effects on health (Fernandez, 2017). One potential natural flavor is red ginger (*Zingiber officinale var rubrum*) which is commonly used as a natural flavoring agent in the food industry and also as a bioactive component in the pharmaceutical industry (Tajkarimi, 2010; Rajathi *et al.*, 2017). Red ginger is famous for its antimicrobial, antifungal and antioxidant capabilities (Srivastava *et al.*, 2015) which could help delay the degradation in yogurt. This study aims to investigate the effect of varying concentration on red ginger extract addition and fermentation time on yogurt characteristics.

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2. Materials and methods

2.1 Materials

Fresh cow milk and red ginger obtained from the local market in Semarang, Indonesia. Chemicals used were *L. bulgaricus* and *S. thermophilus* fermentation starters (Merck, Germany), NaOH (97%, Merck, Germany), formaldehyde solution (37%, Merck, Germany), potassium oxalate monohydrate (99.9%, Merck, Germany) and phenolphthalein (Merck, Germany).

2.2 Characterization of fresh cow milk

Fresh cow milk was organoleptically tested to determine the taste, smell and color. The pH value of fresh cow milk was determined using a digital pH meter. The pH meter was firstly calibrated at pH 4 and 10 with buffer standards solution. Milk as much as 50 mL was then placed in a beaker, the calibrated pH meter was then inserted and the obtained pH value was recorded. Viscosity was measured using a Brookfield viscometer (Brookfield DVII, USA) with spindle no.4 and 3 rpm rotation speed, results recorded in centipoise (cP) after 60 seconds of shearing. The density of milk was measured using pycnometer 25 mL (IWAKI pyrex). Protein and fat contents of the milk were also investigated for further characterization.

2.3 Characterization of red ginger extract

Grated red ginger was hand-squeezed to obtain its extract. The extract was organoleptically tested for its taste, odor and color. Further characterization included moisture and ash content tests.

2.4 Yogurt preparation

Yogurt was prepared by heating fresh cow milk to 70°C and later cooled to 25°C. The milk was added with red ginger extract of varying concentrations (0.2, 0.4, 0.6, 0.8, 1% v/v) and 2% v/v fermentation starters. Anaerobic fermentation took place to form yogurt for 60 hrs. The fermentation was carried out in sterilized reactor.

2.5 Characterization of yogurt product

The product was characterized for its pH, density, titratable acidity value, protein content and lactic acid bacteria count. pH and density were measured using pH meter and pycnometer respectively. The titratable acid (TA) of yogurt was analyzed by diluting 10 g of yogurt with 10 mL of distilled water, then titrating by 0.1 N NaOH solution and methyl red as an endpoint indicator (Krisnaningsih *et al.*, 2019). Titratable acidity value was calculated using the following equation:

$$\text{Titratable Acidity} = \frac{\text{Vol.NaOH used (mL)} \times 0.1 \text{ N NaOH} \times \text{milliequivalent factor} \times 100}{\text{mass of sample (g)}}$$

Where V is the volume of titrant (mL), F is the concentration of NaOH (mol/L), M is the mass of yogurt titrated (g) and 0.9 belongs to the conversion factor of lactic acid.

Protein analysis was carried out using Sørensen formol titration, where yogurt was titrated with potassium hydroxide in the presence of formaldehyde (Levy, 1932). Lactic acid bacteria count was conducted using total plate count method. The organoleptic test was also carried out to determine its color, odor and taste. Organoleptic test using quantitative descriptive analysis (QDA). The procedure for the ginger yogurts was similar to that used by Yang *et al.* (2012) for peach-flavored yogurt drinks. Because semi-trained assessors have been found to be appropriate for studying major characteristics of products, we organized the panel with 10 semi-trained assessors (5 females and 5 males in the age range of 18-55 years). The assessors were trained in 6 sessions and for 2 hrs each. In the first 3 sessions, they were trained for developing the vocabulary of descriptors, and in the next 2 sessions, the assessors established the references of each attribute. All 5 samples were served to the assessors in 100-mL plastic cups with lids labeled with 3-digit random codes and the evaluations were repeated 4 times.

3. Results and discussion

3.1 pH and titratable acidity

Effects of different ginger extract concentration on pH value and acidity of yogurt samples were investigated and the results were presented in Figure 1 and 2. It is seen that the time of fermentation and the level of the added ginger extract was reducing the pH (Figure 1) and increasing the acidity (Figure 2). The titratable acidity is expressed as a percentage of lactic acid present in the yogurt samples (Matela *et al.*, 2019). This phenomenon as a result of the production of lactic acid by fermentative organisms responsible for the fermentation. But there was no significant difference between the lower or higher addition of ginger extract. According to IHEMEJE *et al.* (2015), fermentation of sugars in milk (specifically lactose) into lactic acid will increase the acidity of milk and hence decreases pH value. This statement was also supported by Marshall (1987), who concluded that the acidic taste of yogurt was caused by lactic acid formation. *Streptococcus thermophilus* will first reduce the initial pH to less than 5.5 by synthesizing formic acid. *Lactobacillus bulgaricus* growth was simultaneously stimulated by formic acid and further lower the pH as *Streptococcus thermophilus* bacterial activity decreases (Bourlioux and Pochart, 1988). The acidity of yogurt samples in this study was 1.3-3.1%, still within the Indonesian National Standard of 0.5-2% (SNI,

2009).

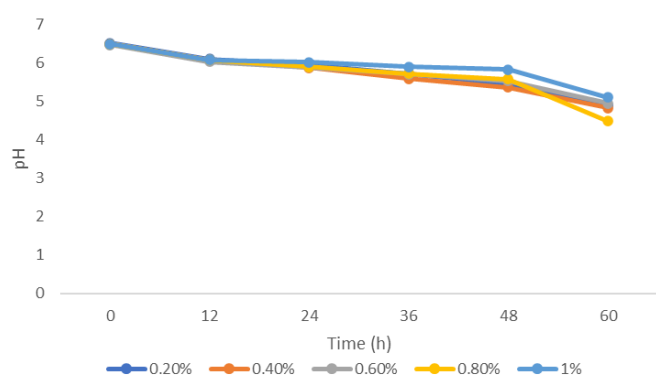


Figure 1. pH of yogurt with various red ginger extract concentration

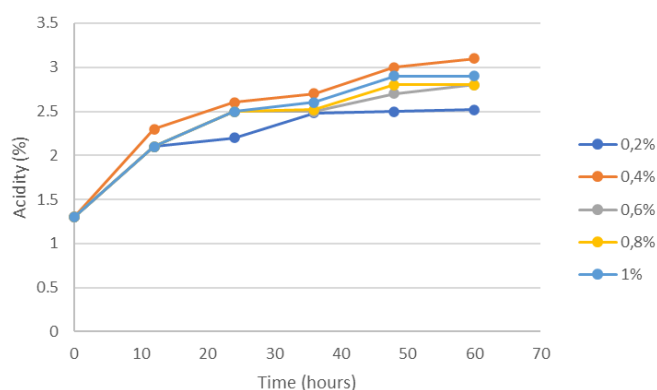


Figure 2. Titratable Acidity of yogurt with various red ginger extract concentration

3.2 Density

The results of this study indicate increasing fermentation time would decrease yogurt density (Figure 3). Bacteria's activity on yogurt production were fermented lactose (milk sugar) to lactic acid. Longer fermentation time would be increasing lactic acid formation (Turner *et al.*, 2016). Density is the amount of mass over volume, therefore less mass would lead to less density (SERC, 2020). Increasing of lactic acid decreases density of yogurt because of its molecular weight which is the mass of a molecule of a substance. Lactic acid has a molecular weight of 90.08 g/mol (National Center for Biotechnology Information, 2020), which was lighter

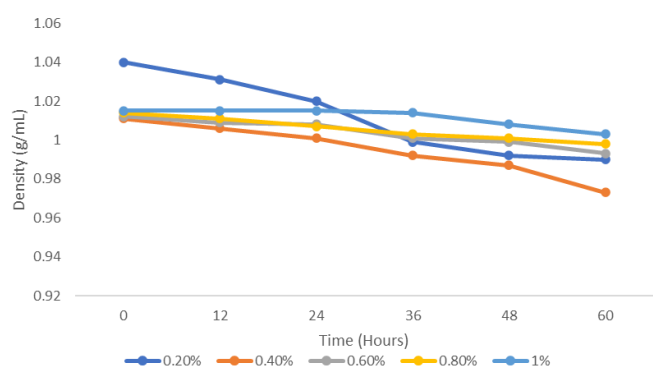


Figure 3. Density of yogurt with various red ginger extract concentration

than lactose that has a molecular weight of 342.3 g/mol (National Center for Biotechnology Information, 2020).

3.4 Protein content

As can be seen in Table 1, there is no significant difference in protein content in various concentration of red ginger addition. Based on the protein analysis, red ginger that use for fermentation doesn't contain protein this causes red ginger extract to yogurt did not give any significant effect to the protein content. The protein content among all concentration of red ginger extract has similar values with no clear trend(s) shown.

Table 1. Protein content of yogurt with various red ginger extract concentration

Time (hours)	% Protein				
	0.20%	0.40%	0.60%	0.80%	1.00%
0	3.50	3.49	3.51	3.50	3.51
12	3.50	3.49	3.51	3.50	3.51
24	3.50	3.49	3.51	3.50	3.51
36	3.49	3.49	3.51	3.51	3.51
48	3.49	3.50	3.52	3.51	3.51
60	3.49	3.50	3.52	3.51	3.51

3.5 Organoleptic analysis

The yogurt produced was organoleptically tested to 10 respondents for its odor, flavor and color. Table 2 shows that scores of yogurt aroma improved with increasing the amount of ginger extract. Yogurt produced have a spicy and pungent ginger aroma, particularly in a higher concentration of red ginger extract. According to Ketaren (1987), the spicy aroma of ginger is caused by zingiberene and zingiberol, 2 essential oil compounds found in red ginger. The addition of ginger extract could take weaken effect on the sweetness but increase the spicy flavor. Red ginger has a natural spicy flavor that comes from oleoresin (Jyotsna *et al.*, 2017). For the color of the yogurt produced, the results stated that it has a yellowish-white color, approximately of similar color to the commercial yogurt.

Table 2. Organoleptic analysis of yogurt with various red ginger extract concentration

Variable	Odour	Color	Flavor	
			Sweet	Spicy
0.20%	1.75	2.15	3.75	2.20
0.40%	1.78	2.16	3.70	2.23
0.60%	1.86	2.20	3.64	2.86
0.80%	1.97	2.22	3.58	3.17
1.00%	2.20	2.22	3.50	3.22

3.6 Lactic acid bacteria count

Table 3 shows the results of lactic acid bacteria count (LABC) at fermentation time 60 h. There was an increasing bacterium count for 0.4% v/v addition of red

ginger extract, then the bacteria count were decreased and had similar value for 0.6%, 0.8%, and 1% v/v ginger extract addition. A study conducted by Srivastava *et al.* (2015) stated that red ginger has the capability to be an antimicrobial and antifungal agent. So, at higher concentrations of red ginger, the antimicrobial and antifungal characteristics began to affect the bacterial activity.

Table 3. Lactic acid bacteria count of yogurt with various red ginger extract concentration at 60 hrs fermentation

Variable	Total BAL (CFU/mL)
0.00%	1.8 x 10 ⁹
0.20%	1.8 x 10 ⁹
0.40%	2.3 x 10 ⁹
0.60%	1.8 x 10 ⁹
0.80%	1.8 x 10 ⁹
1.00%	1.8 x 10 ⁹

4. Conclusion

pH of yogurt was decreasing and titratable acidity increased during the fermentation time in various concentration of ginger extract addition. The addition of red ginger extract did not give a significant effect on protein content of yogurt. Yogurt with a higher concentration of ginger extract increased the spicy aroma and flavor, however, the color was similar to the commercial yogurt. At higher concentrations of red ginger shows antimicrobial and antifungal characteristics, so there's no increase in microorganisms' amount.

Conflict of interest

The authors declare no conflict of interest.

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