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Total Count of Lactic Acid Bacteria in Goats and Cows Milk Yoghurt using Starter *S. thermophilus RRAM-01, L. bulgaricus RRAM-01* and *L. acidophilus IIA-2B4*

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ABSTRACT

Yoghurt is a well-known fermented dairy product which produced using a combination of lactic acid bacteria (LAB) of *Streptococcus thermophilus*, *Lactobacillus bulgaricus* and *L. acidophillus* as fermentation starters. Cow milk is usually used as a raw ingredient. The LAB-based local yoghurt starter (*S. thermophilus* RRAM-01 (ST), *L. bulgaricus* RRAM-01 (LB) and *L. acidophilus* IIA-2B4 (LA)) were previously isolated from milk and meat, nevertheless had not been extensively attempted to be used in yoghurt production. This study aimed to evaluate the characteristics of cow and goat milk based yoghurt produced by using a single local strater of *S. thermophilus* RRAM-01 (ST) or *L. bulgaricus* RRAM-01 (LB) or *L. acidophilus* IIA-2B4 (LA). The yoghurts were produced through addition of the starter (3% v/v each) with 10^{10} CFU mL^{-1} of initial population, and then fermented at room temperature for 24 hr. The result revealed that initial population of LAB in goat's milk yoghurt fermented by ST or LB were significantly higher than that of by LA. Yet, after 24-hour of storage at room temperature, the total population of LA increased and reached final population which was higher than LB or ST. Meanwhile, cow's milk yoghurt fermented by LB had the highest population of at the initial day (D0), while after fermentation the highest population were observed on LB or LA cow's milk yoghurt. Overall goat's milk yoghurt had significantly lower pH values than the cow's milk yoghurt. These were accompanied by higher the total titrated acid (TTA) of goat's milk yoghurt than that of cow's milk yoghurt. Based on pH and TAT values, it was found that ST bacteria produced significantly higher total acidity goat's milk yoghurt, followed by LB and LA. However, the type of culture had no effect on total acidity of cow's milk yoghurt.

Keywords: Cow milk, Goat milk, LAB, Yoghurt

Introduction

Yoghurt is a fermented dairy product produced by using the combination of *Streptococcus thermophiles* and *Lactobacillus bulgaricus*, or other lactic acid bacteria (LAB) (BSN, 2009). The selection of LAB used in yoghurt production influences the quality of final product (Hutkins, 2006). In general, the starter culture consist of two or more different types bacteria, such as *Streptococcus thermophiles* and *Lactobacillus bulgaricus* (Zuriati *et al.,* 2011; Fatmawati *et al.,* 2013), *Streptococcus thermophilus, Lactobacillus bulgaricus*, and *Lactobacillus acidophilus* (Harjiyanti *et al.,* 2013). Each LAB has distinct characteristics in terms of its ability on breaking down complex molecule into simple ones.

S. thermophilus grows and initiates lactic acid production more rapidly, thus decrease pH value faster. It produces CO_2, formic acid, and diacetyl, giving creamy and buttery flavors (Tamime and Robinson, 2007). *L. bulgaricus* produces 1.2 to 1.5% of lactic acid and acetaldehyde which are part of flavors components (Hui, 1993). Furthermore, other strains of *L. d. bulgaricus*, i.e. *L. bulgaricus* G LB44 synthesize bacteriocin (Tamime and Robinson, 2007). *L. bulgaricus* degrades all casein (mainly β-kasein), while *S. thermophiles* has lower proteolytic activity, unaffected by casein hydrolysis (Makinen and Bigret, 1998). *L. acidophilus* is able to synthesize lactase, vitamin K, and anti-bacterial compounds such as acidotin, acidophiline, bacteriocin, and lactocidin (Goldin and Gorbach, 1992). Those compounds affect the pH level and the growth of those bacteria itself.

Cow milk-based yoghurt dominates the yoghurt market as it becomes the most consumed yoghurt all over the globe (Granato *et al.,* 2010).

The high demand of dairy products other than fresh cow milk is a result of the allergic response phenomenon towards cow milk (Farnsworth *et al.,* 2007). The allergic occurrence is an undesired reaction caused by the failure of immune system towards cow's milk protein. β-immunoglobulin (β-Ig) is the main antigen causing the allergic reaction on infants (El-Agamy, 2007). Goat's milk has less allergic response and higher digestibility than cow's milk (Haenlein, 2004), greater contents of short chain fatty acids and antibacterial compounds (Slacanac *et al.,* 2010). The lactose content of goat's milk (5.21%) (Budiarsana and Sutama, 2001) is higher than cow's milk (4.7%) (Sinuhaji, 2006).

S. thermophilus RRAM-01, *L. bulgaricus* RRAM-01, and *L. acidophilus* IIA-2B4 are local LAB isolated from milk and meat, yet have not be used widely in yoghurt production. Hence, this study aims to evaluate the characteristics of cow and goat milk based yoghurt, fermented using *Streptococcus thermophilus RRAM-01* or *Lactobacillus bulgaricus RRAM-01* or *Lactobacillus acidophillus IIA-2B4*. Hopefully, this study will provide information regarding characteristic of yoghurt produced by using those LAB as single starter.

Materials and Methods

Main ingredients used on this study are culture starter, cow's milk, and goat's milk. Etawah (PE) goat milk and Friesian Holstein (PHF) milk were obtained from Fapet Farm at Institut Pertanian Bogor. Culture started used on this study are *Streptococcus salivarius* subsp *thermophilus* RRAM-01, *Lactobacillus delbruecki* subsp *bulgaricus* RRAM-01, and probiotic *Lactobacillus acidophilus* IIA-2B4, available at Laboratory of LAB, Department of Animal Production and Technology, Faculty of Animal Science, Institut Pertanian Bogor.

LAB culture preparation

Inoculum were first grown on Nutrient Broth (NB) media for 25 hours at 37ºC (Arief *et al.,* 2014) to adapt the LAB on the media and increase their viability, indicated by the thicker color of the media. The results were then inoculated (2%) on 10% of sterile skim milk, incubated at 37ºC for 48 hours. Results of the process were then named as parent culture. The process were conducted for several times until obtaining inter and main culture. The main culture were then counted on plate count agar (PCA) to evaluate its initial number. Culture can be used as main culture if it its meet the minimum population of ≥ 10^8 log CFU mL^{-1}.

Yoghurt production

By using autoclave, milk were pasteurized at 95 ºC for 35 minutes, then cooled until reached 43 to 45 ºC. 3% (v/v) of culture starter with 10^{10} CFU mL^{-1} of population were added to pasteurized milk. The culture starter-added milk were than incubated for 24 hours until coagulation formed. The total count of LAB were evaluated at h-0 and h-24 of the incubation period. Meanwhile, the pH level and total titrated acid (TTA) were evaluated at h-0, h-1, h-2, h-3, h-4, h-5, h-18, h-20, h-22, and h-24 of incubation period.

This study used completely randomized design with 6 treatments and 3 replications. The treatments were types of yoghurt (goat and cow), manufactured by the addition of single culture of *Streptococcus salivarius* subsp *thermophilus* RRAM-01 or *Lactobacillus delbruecki* subsp *bulgaricus* RRAM-01 or *Lactobacillus acidophilus* IIA-2B4. Data were analyzed by using variance test. Differences between groups were then subjected to further Tukey test.

Total count of LAB

25 mL yoghurt sample were diluted into 225 mL buffered peptone water (BPW) to obtain 10^{-1} dilution. 1 mL of diluted sample were than diluted into 9 mL of BPW to achieve 10^{-2} dilution. Sample from previous dilution were then diluted by using the same method until reaching 10^{-10} of dilution. 1 mL of sample from 10^{-8} to 10^{-10} of dilution were then transferred to petri dish, mixed with 15 mL of plate count agar (PCA) medium. Those petri dish were then incubated at 37 ºC for 48 hours. The LAB colony grown on the medium were then counted by using colony counted according to standard plate count (SPC) method (BAM, 2001).

pH test

pH value were determined by using electronic pH meter (Schoot Instrument Lab 850, Germany). The end tip of cathode indicator was washed by using distilled water and dried with a tissue swap. The pH meter was then calibrated by tipping the cathode end into pH 4 and 7 of buffer solution. The pH value of samples were then determined by tipping the cathode end into samples. Once the pH meter indicated "ready", the value shown on the display was recorded as the pH value of the sample (AOAC, 2005).

Total titrated acid (TTA) determination

Total titrated acid (equivalent with the lactic acid concentration) was determined by titration (Nielsen, 2003). 10 mL of yoghurt sample were diluted with distilled water (1:1; v/v), added with 3 drops of phenolphthalein (PP) (1%), and titrated with 0.1 N NaOH until stable pink color formed.

Data analysis

This study was carried out by using completely randomized design, with 6 treatment groups and 3 replications. Variables observed on this study included total LAB count of main culture, total LAB count of yoghurt, pH, and total titrated acid. All data were statistically evaluated on analysis variance (Minitab version 16 computer software).

Result and Discussion

Total LAB count of main culture

The population number of lactic acid bacteria on a yoghurt product determines its microbiological quality. Lactic acid ($C_3H_6O_3$) is the main acid produced on yoghurt production. Yoghurt is a fermented milk product using lactic acid bacteria (LAB) as the main culture. The LAB culture used on this study were *Lactobacillus delbrueckii subsp bulgaricus* RRAM-01, *Streptococcus salivarus subsp thermophilus* RRAM-01, and *Lactobacillus acidophilus* IIA-2B4. *Lactobacillus acidophilus* IIA-2B4 was isolated from fresh Ongole beef at traditional market in Bogor area (Arief *et al.,* 2015). Population of started culture on skim milk (main culture) for 3 types of LAB used on this study is presented on Table 1.

LAB population used on goat's and cow's milk-based yoghurt exceed 10^{10} CFU mL^{-1}. Arief *et al*. (2015) stated that 10^6 CFU mL^{-1} from more than 10^8 CFU mL^{-1} of LAB can reach small intestine and act as probiotic. Starter culture population used on this study has met BSN standar (2009), i.e. min. 10^7 CFU mL^{-1}.

Total count of LAB on yoghurt

Yoghurt is a fermented product from milk and or reconstituted milk using *L. bulgaricus* and *S. thermophilus* or other LAB, with or without food ingredients additions (BSN, 2009). *Lactobacillus acidophilus* IIA-2B4, a probiotic isolated from Indonesian beef meat, shows antimicrobial capacity towards pathogenic bacteria (Arief *et al.,* 2015). Addition of *Lactobacillus acidophilus* IIA-2B4*, Streptococcus salivarius* subsp *thermophilus* RRAM-01*,* and *Lactobacillus delbruecki* subsp *bulgaricus* RRAM-01 on goat and cow milk-based yoghurt can deliver positive health outcome. Wirjantoro and Phianmongkhol (2009) suggested that minimum daily probiotic consumption is 10^8 log CFU mL^{-1} to ensure its health benefit. The total count of LAB on yoghurt is presented on Table 2 and 3. Yoghurt produced on this study can be categorized as probiotic yoghurt.

Table 2 shows significant difference (P<0.05) on the total count of *Streptococcus salivarius* subsp *thermophilus* RRAM-01 between goat and cow milk-based yoghurt. The total count of LAB on goat milk-based yoghurt was higher than cow milk-based yoghurt, influenced by the availability of substrate for the LAB. For a certain period of time, the number of substrate on fermented milk product is available in great amount. The abundant substrate enables LAB to proliferate. However, it will then drops and cause the LAB to be relatively inactive and has passed the logarithmic phase (Sunarlim and Usmiati, 2006). The addition of *Lactobacillus delbruecki* subsp *bulgaricus* RRAM-01did not give significant difference on the total count of LAB. Usmiati (2011) reported that bacteria cells possibly undergo lysis process during storage, causing smaller number during counting process. Addition of *Lactobacillus acidophilus* IIA-2B4 resulted in higher total count of LAB on cow-milk based yoghurt than on goat-milk based yoghurt (P<0.05). There are several factors that can influence LAB growth, i.e. nutrition, temperature, humidity, oxygen, pH, and inhibiting substances. Nutrition as such, is in form of lactose and protein from milk that provide vital carbon and nitrogen for LAB growth. Lactose is the main energy source available on milk, supplying almost half of total calorie (35-45%) (Sinuhaji, 2006). Milk protein can increase LAB growth (Karinawatie *et al.,* 2008). Protein content of goat milk is 4.29 % (Zuriati *et al.,* 2011), 4.1% (Rosartio *et al.,* 2015), 3.71% (Hanum *et al.,* 2016), and 3.8-3.9% (Ratya *el al*., 2017), while cow milk contains 3.71% protein (Hanum *et al.,* 2016) and 3.01-3.59% (Oka *et al.,* 2017). The nutrient content of goat milk meets the requirement for metabolic activity of

Table 1. Total count of lactic acid bacteria (LAB) in main culture

Starter	Total count (log CFU mL^{-1})
Streptococcus salivarius subsp *thermophilus* RRAM-01	11.14±0.02
Lactobacillus delbruecki subsp *bulgaricus* RRAM-01	10.69±0.03
Lactobacillus acidophilus IIA-2B4	10.59±0.01

Data was not statistically tested.

Table 2. Total count of LAB (log CFU mL^{-1}) on yoghurt during storage at room temperature (h-0)

Starter	Types of yoghurt (h-0)	
	YSK0	YSS0
Streptococcus salivarius subsp *thermophilus* RRAM-01	9.67±0.05[a]	9.27±0.01[b]
Lactobacillus delbruecki subsp *bulgaricus* RRAM-01	9.77±0.07[a]	9.67±0.05[a]
Lactobacillus acidophilus IIA-2B4	9.07±0.07[c]	9.35±0.03[b]

Different superscripts indicate significant difference (P<0.05) among groups. YSK0: goat milk-based yoghurt at h-0. YSS0: cow milk-based yoghurt at h-0).

Table 3. Total count of LAB (log CFU mL^{-1}) on yoghurt during storage at room temperature (h-24)

Starter	Types of yoghurt (h-24)	
	YSK24	YSS24
Streptococcus salivarius subsp *thermophilus* RRAM-01	11.72±0.02[b]	11.31±0.22[c]
Lactobacillus delbruecki subsp *bulgaricus* RRAM-01	10.94±0.06[d]	12.28±0.01[a]
Lactobacillus acidophilus IIA-2B4	12.48±0.00[a]	12.33±0.01[a]

Different superscripts indicate significant difference (P<0.05) among groups. YSK24: goat milk-based yoghurt at h-24. YSS24: cow milk-based yoghurt at h-24).

Streptococcus salivarius subsp *thermophilus* RRAM-01 and *Lactobacillus delbruecki* subsp *bulgaricus* RRAM-01. Thus, the total count of LAB on goat milk-based yoghurt was observed higher.

The total count of *Streptococcus salivarius* subsp *thermophilus* RRAM-01 between goat and cow milk-based yoghurt were significantly different (P<0.05), see Table 3. Yoghurt produced from milk goat had greater number of LAB than cow milk yoghurt. *S. thermophilus* produce lactic acid and small amount of formic acid. *S. thermophilus* also produce amino acids to support its growth. Chemical content of milk (total solid and fat content) determines starter culture activity (Astawan *et al.,* 2012). The addition of *Lactobacillus delbruecki* subsp *bulgaricus* RRAM-01IIA-2B4 resulted in higher total count of LAB on goat milk-based yoghurt than on cow milk-based (P<0.05). Many fermented milk products are based on high proteolytic starter cultures. Microorganism can break protein down to produce peptides with specific amino acid chains. Each proteolytic microorganism has distinct ability in digesting protein. Hence, the bioactive peptide produced are specific among microorganism, i.e. the ability of *L. delbrueckii subsp. Bulgaricus* to break β-kasein will affect the activity of starter culture (Ashar and Chand, 2004).

Compared to other LAB, *Lactobacillus acidophilus* IIA-2B4 did not deliver significant difference between goat and cow milk-based yoghurts. However, the total LAB count of the yoghurt was higher than yoghurt that produced using *Streptococcus salivarius* subsp *thermophilus* RRAM-01 or *Lactobacillus delbruecki subsp bulgaricus* RRAM-01. Krasaekoopt *et al*. (2003) reported that common starter culture of *L. bulgaricus* and *S. thermophiles* synthesize β-galactosidase on the yoghurt. However, those LABs can not survive and grow on small intestine due to low tolerance for bile salt. *L. acidophilus* has capacity in inhibiting free radical and lipid peroxidation. The protective capacity increases during fermentation (Virtanen, 2007). Peptides and hydrosilate from α-lactalbumin have been reported to have antimicrobial property, presumably by lethally affecting the membrane function of microbes. The high total count of LAB on milk-based yoghurt due to the nutrient availability on the milk to support metabolic activity of *Lactobacillus acidophilus* IIA-2B4. On this study, *Lactobacillus acidophilus* IIA-2B4 grew well on cow milk-based yoghurt added by probiotic. Thus, the total count of LAB increased on that product. Fermented milk products have been recommended as dietary supplementation due to their hypocholesterolaemic benefit for human (El Gawad *et al.,* 2005).

pH value

Result of pH value evaluation is presented on Table 4. According to analysis of variance, the pH values at h-0 of goat milk produced using *Lactobacillus acidophilus* or *Lactobacillus bulgaricus* or *Streptococcus thermophilus* was not significantly different (6.40; 6.38; and 6.40, respectively). The pH kept decreasing as the fermentation process taking place. However, the final pH value at h-24 of yoghurt produced using *Lactobacillus acidophilus, Lactobacillus bulgaricus*, and *Streptococcus thermophilus* were not significantly different (5.30; 5.29; and 5.34 respectively).

Similar to the goat milk-based yoghurt, the initial pH value (h-0) of cow milk yoghurts produced using *Lactobacillus acidophilus* was 6.56. The value was not significantly different compared to yoghurts produced using *Lactobacillus bulgaricus* and *Streptococcus thermophilus* (6.43 and 6.41). Compared to *Streptococcus thermophilus*, the pH value at h-0 of yoghurt produced using *Lactobacillus acidophilus* was significantly different, yet insignificant compared to *Lactobacillus bulgaricus* yoghurt until storage time at h-0. During h-18 to h-24 observation, the pH value decreased yet insignificantly different. The final pH value at h-24 were recorded as follow *Lactobacillus acidophilus* (5.57), *Lactobacillus bulgaricus* (5.68), and *Streptococcus thermophilus* (5.66).

The decreasing pH value from h-0 to h-24 indicated lactic acid production from all starters. The pH values is negatively correlated with the total titrated acid. The declining pH value of yoghurt after 24 hours storing demonstrated the accumulation of acid contents. The result is similar with Fatmatwati *et al*. (2013) who reported that the pH value at day-0 of goat milk-based yoghurt using *Lactobacillus bulgaricus* and *Streptococcus thermophilus* was 6.99. The acidity level decreased to 5.94 at day-2. Meanwhile, cow milk-based yoghurt using the same starter combination had 6.99 and 5.02 of pH values at day-0 and day-2 respectively.

The pH value observed on this study was higher than study carried out by Rahmawati and Suntornsuk (2016), who reported 4.39 of pH value of cow and goat milk-based yoghurt. The lower pH value was also reported by Hidayatat *et al*. (2013), combination of *S. thermophilus, L. Bulgaricus,* and *L. acidophilus* produced cow milk-based yoghurt with 4.8 of pH value.

The differences between studies are presumed due to the number of lactic acid bacteria used on each study. The starter culture combination may produce more lactic acid bacteria, thus lowering the pH value. A combination of *S. thermophilus* and *L. bulgaricus* is a mutualism relationship in which each type of bacteria provide essential component for each other to increase the lactic acid production. The nutrient availability and storing temperature may also affect the ability of LAB in producing lactic acid.

Total titrated acid

The analysis of variance carried out on this study demonstrated that starter types resulted in different total titrated acid at incubation period for

Table 4. pH value of goat and cow milk-based yoghurt

Incubation (h-)	Starter types					
	Streptococcus salivarius subsp *thermophilus* RRAM-01		*Lactobacillus delbruecki* subsp *bulgaricus* RRAM-01		*Lactobacillus acidophilus* IIA-2B4	
	YSK	YSS	YSK	YSS	YSK	YSS
0	6.40±0.04^{b}	6.42±0.10^{b}	6.38±0.04^{b}	6.43±0.01^{ab}	6.40±0.01^{b}	6.56±0.10^{a}
1	6.38±0.05^{b}	6.41±0.12^{b}	6.37±0.06^{b}	6.43±0.01^{b}	6.37±0.03^{b}	6.54±0.11^{a}
2	6.37±0.02^{b}	6.41±0.08^{b}	6.36±0.03^{b}	6.41±0.01^{b}	6.36±0.02^{b}	6.53±0.07^{a}
3	6.37±0.04^{b}	6.40±0.08^{b}	6.36±0.03^{b}	6.40±0.02^{b}	6.35±0.02^{b}	6.51±0.08^{a}
4	6.35±0.03^{b}	6.38±0.07^{ab}	6.35±0.04^{b}	6.39±0.01^{ab}	6.32±0.00^{b}	6.46±0.06^{a}
5	6.32±0.04^{b}	6.39±0.04^{ab}	6.32±0.03^{b}	6.35±0.0^{b}	6.31±0.00^{b}	6.44±0.07^{a}
18	5.71±0.14^{bc}	5.93±0.16^{ab}	5.68±0.13^{c}	5.96±0.03^{a}	5.67±0.19^{c}	5.77±0.20^{abc}
20	5.68±0.13^{ab}	5.85±0.18^{a}	5.58±0.16^{b}	5.88±0.02^{a}	5.64±0.2^{ab}	5.75±0.26^{ab}
22	5.52±0.16^{b}	5.84±0.22^{a}	5.51±0.12^{b}	5.81±0.01^{b}	5.50±0.19^{b}	5.66±0.23^{ab}
24	5.34±0.16^{b}	5.66±0.18^{a}	5.29±0.12^{b}	5.68±0.03^{a}	5.30±0.27^{b}	5.57±0.22^{ab}

Different superscripts indicate significant difference ($P<0.05$) among groups. YSK: goat milk-based yoghurt. YSS: cow milk-based yoghurt.

Table 5. Total titrated acid of goat and cow milk-based yoghurt during h-0 to h-24 of storage time

Incubation (h-)	Starter types					
	Streptococcus salivarius subsp *thermophilus* RRAM-01		*Lactobacillus delbruecki* subsp *bulgaricus* RRAM-01		*Lactobacillus acidophilus* IIA-2B4	
	YSK	YSS	YSK	YSS	YSK	YSS
0	0.24±0.01^{a}	0.23±0.01^{ab}	0.20±0.02^{b}	0.21±0.01^{b}	0.23±0.00^{ab}	0.23±0.00^{ab}
1	0.24±0.02	0.23±0.00	0.23±0.03	0.23±0.00	0.23±0.00	0.23±0.00
2	0.25±0.00	0.23±0.01	0.23±0.01	0.24±0.00	0.24±0.00	0.25±0.00
3	0.26±0.01^{ab}	0.24±0.01^{b}	0.24±0.01^{b}	0.26±0.00^{ab}	0.24±0.00^{b}	0.27±0.00^{a}
4	0.27±0.00^{a}	0.26±0.00^{ab}	0.25±0.00^{b}	0.26±0.00^{ab}	0.27±0.00^{a}	0.27±0.00^{a}
5	0.26±0.01	0.27±0.02	0.25±0.01	0.26±0.01	0.27±0.00	0.28±0.01
18	0.46±0.00^{a}	0.38±0.00^{c}	0.46±0.00^{a}	0.38±0.01^{c}	0.43±0.00^{b}	0.39±0.00^{c}
20	0.46±0.03^{a}	0.42±0.00^{b}	0.48±0.00^{a}	0.40±0.00^{b}	0.48±0.01^{a}	0.40±0.00^{b}
22	0.53±0.01^{a}	0.47±0.01^{b}	0.49±0.01^{b}	0.42±0.00^{c}	0.48±0.01^{b}	0.42±0.00^{c}
24	0.64±0.04^{a}	0.48±0.01^{c}	0.61±0.02^{ab}	0.48±0.03^{c}	0.56±0.02^{b}	0.48±0.00^{c}

Different superscripts indicate significant difference ($P<0.05$) among groups. YSK: goat milk-based yoghurt. YSS: cow milk-based yoghurt.

0, 3, 4, 18, 20, 22, and 24 hours (Table 5). Total acid of goat milk-based yoghurt using *Streptococcus salivarius* subsp *thermophilus* RRAM-01 (0.24) was significantly higher than *Lactobacillus delbruecki subsp bulgaricus* RRAM-01 yoghurt (0.20), yet insignificantly different compared to *Lactobacillus acidophilus* IIA-2B4 yoghurt (0.23). Starter types used on cow milk-based yoghurt did not alter the total acid at h-0 of incubation. The total acid on either goat or cow milk-based yoghurt increased along with the length of incubation period.

Total acid of goat milk-based yoghurt at h-24 of incubation was 0.56 to 0.64%, higher than cow milk-based yoghurt (0.48%). In general, the total acid produced on this study was lower than BSN standard (2009), in which acidity of yoghurt ranges from 0.5 to 2%. Harjiyanti *et al*. (2013) reported yoghurt added with mango flavor was 0.75 to 0.79%. The lower total titrated acid may be a result form the use of single starter culture. Sunarlim *et al*. (2007) stated that viability of single bacteria is lower than double or triple starters due to no mutualism symbiosis present among bacteria.

Streptococcus thermophilus produce acid faster than *Lactobacillus*. Hence, the total titrated acid of goat milk-based yoghurt produced using *Streptococcus salivarius* subsp *thermophilus* RRAM-01 (0.64%) was higher than *Lactobacillus acidophilus* IIA-2B4 yoghurt (0.56%). The higher total titrated acid on goat milk yoghurt at h-24 than cow milk-based yoghurt may be caused by the higher lactose content on goat milk. Ekawati (2013), reported that the average lactose content of Etawa (Peranakan Etawa) goat milk is 0.05733 g/ml, while the average lactose content of cow-milk is 0.02367 g/ml. Budiarsana and Sutama (2001) also reported higher lactose content on goat milk (5.21%) than cow milk (4.7%) (Sinuhaji, 2006).

Conclusions

The total count of LAB of goat and cow milk-based yoghurt at h-0 were 10^{9} log CFU ml^{-1} and reached 10^{11}-10^{12} log CFU ml^{-1} at h-24. Yoghurt produced on this study can be categorized as probiotic yoghurt as have met the minimum standard by Indonesian National Standard (10^{7} log CFU ml^{-1}). pH value of goat and cow milk-based yoghurt were 5.29-5.34 and 5.57-5.68 respectively. The total titrated acid of goat milk-based yoghurt was higher (0.56-0.64%) than cow milk-based (0.48%).

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