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Comparison of Microbiological Quality Between Organic and Conventional Goat Milk: A Study Case in Bogor, Indonesia

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Abstract | The aim of this study was to compare microbiological quality between organic and conventional goat milk in Bogor District, West Java Province, Indonesia. Between March to August 2018, a total of 36 bulk tank milk samples were collected from 3 locations of organic and conventional goat farms. The milk samples were determined for bacteria population (total plate count, *Staphylococcus aureus*, *Enterobacteriaceae* and coliform) and parasite population (*Toxoplasma gondii*, *Entamoeba* sp. and *Balantidium* sp.). The determination was according to US FDA-BAM (United States Food and Drug Administration–Bacteriological Analytical Manual) for Aerobic Plate Count analysis. Data were analyzed by Mann Whitney test. The presence of parasite was analyzed descriptively. Results showed that the populations of total bacteria, *S. aureus*, *Enterobacteriaceae* and coliform present in organic goat milk were 5.58, 3.51, 4.32 and 3.69 log cfu/ml, respectively. On the other hand, total bacteria, *S. aureus*, *Enterobacteriaceae* and coliform populations present in conventional goat milk were 5.02, 2.89, 4.12, and 2.46 log cfu/ml, respectively. Population of *S. aureus*, *Enterobacteriaceae*, and coliform in organic goat milk exceeded the maximum limit of the Indonesian National Standard, whereas for conventional goat milk only *S. aureus* and *Enterobacteriaceae* exceeded the maximum limit. *T. gondii* was not found either in organic or conventional goat milk, but other types of protozoa, i.e. *Entamoeba* sp. and *Balantidium* sp. were found. In conclusion, microbiological quality of organic and conventional goat milk in Bogor was relatively similar and appropriate within the Indonesian National Standard.

Keywords | Organic milk, Goat, Bacteria, Protozoa, Microbiology

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INTRODUCTION

Organic milk is defined as milk that free of hazardous materials, originating from livestock based on environmental sustainability and animal welfare. It is produced in organic farms that apply minimum standards to ensure livestock well-being (Sundrum, 2001; Schwendel et al., 2015). Presently the demand for organic milk is increasing (Liu et al., 2013; Malissiova et al., 2015) since it is believed to promote health better as compared to conventional milk (Malissiova et al., 2015). Organic milk products are free from antibiotics, hormones, synthetic chemicals, and ge-

netic modification; and they should be beneficial for human health (Schwendel et al., 2015). Despite its benefit, risk of parasitic contamination in dairy products from organic farms is higher than those from conventional farms (Sundrum, 2001) due to the typical grazing system. In a previous study, organic milk from cows was contaminated with a number of microbes such as coliform bacteria, *S. aureus*, *Enterobacter aerogenes* and *Proteus vulgaris* (Iqbal et al., 2016). In addition, protozoa species such as *Toxoplasma gondii* as also found in raw goatmilk, and it had been studied in the United States, Brazil, Greece and Indonesia (Dubey et al., 2014; Da Silva et al., 2015; Sadek et al., 2015;

Saridewi et al., 2015). *T. gondii* can cause toxoplasmosis, an intracellular obligate cosmopolitan zoonosis, when infects humans and warm-blooded animals. Prevention can be done to avoid contamination of *T. gondii* in goat's milk by improving sanitation management. *T. gondii* in goat's milk can be transmitted to humans if it was raw consumed or used without pasteurization (Dubey et al., 2014; Da Silva et al., 2015).

In Indonesia, dairy goats have not received much attention as compared to dairy cows (Taufik et al., 2011). Meanwhile, the tradition in drinking raw goat milk is still common for many consumers, which is believed that it can enhance health (Claeys et al., 2013) beside better taste and nutrition (Taufik et al., 2011). However, raw milk might contain microbial pathogens such as *S. aureus*, *Escherichia coli*, *Listeria*, *Campylobacter* and *Salmonella* (Hill et al., 2012; Kalmus et al., 2015), and zoonotic pathogens such as *T. gondii* (Dehkordi et al., 2013). To date, comparison of microbiological quality (including the pathogens) between organic and conventional goat milk has never been previously investigated in Indonesia. Therefore the aim of this research was to compare the microbiological quality, i.e. total plate count (TPC), *S. aureus*, *Enterobacteriaceae*, coliform and parasites between organic and conventional goat milk in Bogor District, West Java, Indonesia.

MATERIALS AND METHODS

STUDY AREA AND MILK SAMPLING

Thirty six individual milk samples were collected from six dairy goat farms all over Bogor District, Indonesia, in March to August 2018. Milk samples were collected from two types of goat farms, i.e., organic and conventional goat farms. We used a composite samples from Etawa and Sapera goats. Samples were collected once a month for 6 months. Every 500 mL of bulk was collected in sterile glass bottles and transferred to laboratory under cool condition (5°C). All samples were analyzed immediately at Protozoology Laboratory and Veterinary Public Health, Faculty of Veterinary Medicine, Bogor Agricultural University.

PROCEDURES

The tests carried out in this research were according to Kornacki dan Johnson (2001). This research consisted of some methods like conventional culture of TPC, whereas for the total *S. aureus* count, *Enterobacteriaceae* count, and coliforms count. The number of bacteria was calculated with count method by pouring. Some agars had been used in this study such as Plate Count Agar (PCA) (CM325 Oxoid), Vogel Johnson Agar (VJA) (CM641 Oxoid), MacConkey Agar (MCA) and Violet Red Bile Agar (VRBA) (CM485 Oxoid) (AOAC 2005). The presence of parasites was done by detection of tachyzoites and trophozoites.

The method used microscopic test with Giemsa and Lugol stain. To observe the tachyzoites and trophozoites a prepartate of samples examined thoroughly with a microscope using an oil immersion objective lens (100×) (Sadek et al., 2015; Al-Habsi et al., 2017).

STATISTICAL ANALYSIS

The data obtained were analyzed by Mann Whitney test. Kolmogorov-Smirnov and Shapiro Wilk tests were applied to perceive normality data. The presence of parasite was analyzed descriptively.

RESULTS

There was no difference in bacteria population (TPC, *S. aureus*, *Enterobacteriaceae* and coliform) between organic and conventional goat milk (Table 1). The total bacteria population in both organic and conventional goat milk was still below the Indonesian National Standard threshold and EU Council Directive 92/46/EEC. However, *Enterobacteriaceae* count in both organic and conventional goat milk was above the SNI threshold (3.00 log cfu/ml), i.e. 4.32 and 4.12 log cfu/ml, respectively. The number of *S. aureus* and coliform in organic goat milk were higher than conventional goat milk namely 3.51 and 3.69 log cfu/ml, respectively, and they exceed both SNI and EEC maximum standard. The TPC and *Enterobacteriaceae* were positively correlated with coliform count in organic goat milk ($P < 0.05$) (Table 2). Similarly, the count of TPC in conventional goat milk showed a positive correlation with coliform count (Table 3). *T. gondii* was not detected, however, *Entamoeba* sp. and *Balantidium* sp. were found in both organic goat milk and conventional goat milk (Table 4; Figure 1).

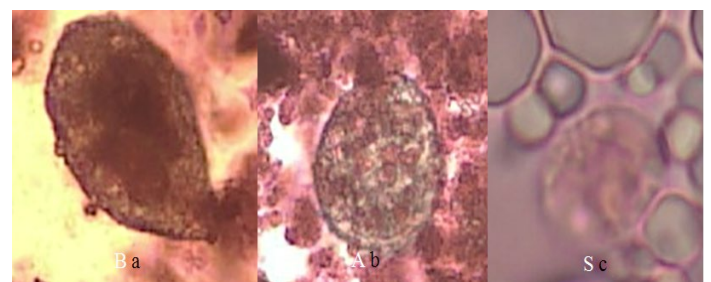


Figure 1: Form of protozoan trophozoites in goat's milk with Lugol's staining (40x magnification) (a) *Balantidium* sp. in organic goat milk; (b) *Balantidium* sp. in conventional goat milk; (c) *Entamoeba* sp. in conventional goat milk.

DISCUSSION

The *S. aureus* grows on VJA was showed colored black colonies, convex, shiny surrounded by yellow zones (Baird and Lee, 1995). *Enterobacteriaceae* have been grown on MCA shows two different types of colonies such as red or pink

Table 1: Bacteria population (log cfu/ml) present in organic and conventional goat milk compared to the standard maximum limit

Bacteria	Organic (mean±SD)	Conventional (mean±SD)	P value	Indonesian National Standard for Fresh Milk	EU Council Directive 92/46/EEC (raw goat and sheep milk) (EC, 1992)
Total plate count	5.58±1.25	5.02±0.91	0.137	6.00	5.70
<i>S. aureus</i>	3.51±1.32	2.89±1.56	0.206	2.00	3.30
Enterobacteriaceae	4.32±1.51	4.12±1.56	0.670	3.00	-
Coliforms	3.69±2.05	2.46±1.86	0.067	3.00	2.00

Table 2: Correlation matrix of microbiological quality in organic goat milk (n =18)

Parameter	TPC	<i>S. aureus</i>	Enterobacteriaceae	Coliforms
TPC	1.000	0.381	0.441	0.589*
<i>S. aureus</i>		1.000	0.011	0.327
Enterobacteriaceae			1.000	0.494*
Coliform				1.000

Table 3: Correlation matrix of microbiological quality in conventional goat milk (n = 18)

Parameter	TPC	<i>S. aureus</i>	Enterobacteriaceae	Coliforms
TPC	1.000	0.302	0.423	0.709**
<i>S. aureus</i>		1.000	0.312	0.310
Enterobacteriaceae			1.000	0.393
Coliform				1.000

Table 4: Presence of *T. gondii*, *Entamoeba* sp. and *Balantidium* sp.(in percentage) in organic and conventional goat milk

Protozoa	Organic (n = 18)	Conventional (n=18)
<i>T. gondii</i>	0	0
<i>Entamoeba</i> sp.	0	16.7%
<i>Balantidium</i> sp.	33.3 %	11.1%

(bacteria that ferment lactose like *Escherichia coli* and *Klebsiella* sp.) and colorless or transparent (bacteria that do not ferment lactose such as *Salmonella* sp. and *Shigella* sp.) (Kornacki dan Johnson, 2001). The coliform grows on VRBA was showed all purplish red colonies, surrounded by red zones (Kornacki dan Johnson, 2001).

The presence of microbes in this study was seen from TPC, *S. aureus*, *Enterobacteriaceae*, and coliform in organic and conventional goat milk with no difference. This result similar to another study conducted by (Sundrum, 2001), which was found no difference in microbial count between organic and conventional goat milk. TPC is the best way to assess milk management and quality by calculating bacterial density in milk and estimating the amount of milliliter aerobic bacteria. TPC values can be high if contamination occurs from milking equipment, dirty udders, and livestock are suffered to subclinical or clinical mastitis (Cicconi-Ho-

gan et al., 2013). In this study we presumed that contamination occur from milk milking since it still used manual milking rather than machine milking.

55 There was no significant difference between organic and conventional goat milk in *S. aureus* count. Although higher in organic udder, the presence of *S. aureus* in bulk tank milk was associated with mastitis, age of the cage and a higher percentage of cows with 3 or fewer teats in both the organic and total herd (Cicconi-Hogan et al., 2013). *S. aureus* can originate from mastitis which can be contagious causes IMI, furthermore it will the affects milk production and somatic cell count (SCC) (Keefe, 2012). *S. aureus* can spread easily from animal to animal through a milking hand or milking device.

The total of coliform in organic goat milk was higher than conventional, although it was not significantly different. In contrast, research in the United States (Iqbal et al., 2016), the total coliform in organic cow milk was lower than conventional cow milk. The presence of coliforms, such as *E. coli* and *Klebsiella* spp., in milk indicates contamination, that can occur from dirty environments and equipment (Iqbal et al., 2016) and can come from mastitis infected livestock (Cicconi-Hogan et al., 2013). Besides coliform, another bacteria also contaminate organic cow milk in Pakistan, especially *Staphylococcus aureus*, and *Proteus vulgaris*. The count of TPC and *Enterobacteriaceae* had a correla-

tion with the amount of coliform in organic goat milk ($P < 0.05$). This result shows that the high of coliform increases the number of total plate count and *Enterobacteriaceae*. Coliform bacteria are used as microbial indicators of cleanliness in the dairy industry. In addition to coliform was also *Enterobacteriaceae* used as an indicator of cleanliness in the dairy industry (Hervert et al., 2016). Coliform bacteria consist of *Escherichia*, *Klebsiella*, *Citrobacter*, and *Enterobacter*, although more than 20 gram negative genera meet the phenotypic coliform criteria (Masiello et al., 2016). The *Enterobacteriaceae* family includes most coliform bacteria usually lack the ability to ferment lactose such as *Salmonella* and *Yersinia* (Imhoff, 2005).

In this study there was no *T. gondii* found in both milk samples (Table 3). *T. gondii* was an intracellular protozoa which cause of toxoplasmosis infect attack warm-blooded animals and humans. Tachyzoites *T. gondii* is found in the milk of sheep, goats, cows, and rats (Dubey et al., 2014; Da Silva et al., 2015; Sadek et al., 2015). Infection in humans was transmitted through drinking consumption of raw goat milk practice (Skinner et al., 1990) and suspected transmission through lactation can also occur in humans (Bonametti et al., 1997). Toxoplasmosis can occur through drinking unpasteurized goat's milk (Skinner et al., 1990; Saridewi et al., 2015). Excretion of tachyzoites in milk comes from naturally infected goats with *T. gondii*. Although infections occur due to drinking goat's milk, other researchers report that drinking raw milk products from other animals can also cause transmission of *T. gondii* horizontally (Kijlstra & Jongert, 2008; Dubey et al., 2014).

Although *T. gondii* was not found, we have found another gastro intestinal parasites namely *Entamoeba* sp and *Balantidium* sp. The risk of parasitic contamination in dairy products from organic farms was higher. It's due to more intensive grazing system in organic farm than conventional farm because organic farms use a more intensive grazing system compared to conventional farms (Sundrum, 2001). Gastrointestinal parasite was an unusual problem in small ruminant production systems (Rahman et al., 2017). Gastrointestinal parasites such as worms and protozoa are very common in goats and sheep [29] so it becomes an infection and can reduce milk and meat production (Murthy & Rao, 2014). Diseases caused by gastrointestinal parasites can interfere with health and harm the economy of farmers (Asif et al., 2008).

Entamoeba sp. was found in conventional goat milk as much as 16.67% (3 samples). Gastrointestinal parasites such as *Entamoeba* sp. can be found in goat's milk probably because of contamination from animal or human feces. *Entamoeba* sp. has been found in goats in Kenya (Kanyari et al., 2009), Thailand (Sangvaranond et al., 2010), Cameroon (Ntonifor et al., 2013), Brazil (Radavelli et al., 2014),

Nigeria (Adua et al., 2017) and Australia (Al-Habsi et al., 2017).

Balantidium sp. was found in organic goat milk (33.33%) and conventional goat milk (11.11%) in the Bogor District. *Balantidium* sp. cysts and trophozoites found 7.1% ($n = 14/198$) in goat intestines and stools in the Egyptian region (Elmadawy & Diab, 2017). The prevalence of *Balantidium coli* in Tanzania was 4.8% (Mhoma et al., 2011) whereas in Kenya, the prevalence was 3%, which is mostly a mixed infection with other parasites (Kanyari et al., 2009). Goat milk has the opportunity to be contaminated with *Balantidium* sp. during the milking process. Cyst *B. coli* that contaminate food and water can come from human and animal feces. Balantidiasis in humans occurs due to contact with sheep, goats and pigs (Yazar et al., 2004; Jamil et al., 2015).

CONCLUSION

The results showed no differences in TPC, *S. aureus*, *Enterobacteriaceae*, and coliform between organic and conventional goat milk. The amount of *S. aureus*, and coliform in organic goat milk exceed the Indonesian National Standard. This is due to the possibility of organic maintenance practices allowing for high amounts of microbes in livestock and in the enclosure environment. *T. gondii* is not found in organic and conventional goat milk, but another parasite was found, namely *Entamoeba* sp. trophozoites in conventional goat milk and *Balantidium* sp. in organic and conventional goat milk. The presence of *Entamoeba* sp. and *Balantidium* sp. in milk is probably due to fecal contamination.

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CONFLICT OF INTEREST

All authors declare that there is no conflict of interest.

AUTHORS CONTRIBUTIONS

VW performed sample collection, data analysis and wrote the manuscript draft. MBW, TP and AJ designed the study, supervised the experiment and revised the manuscript. All the authors have read and approved the final manuscript

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