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Physico-chemical and microbiological qualities of locally produced raw goat milk

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Abstract

Raw goat milk is recognized as one kind of nutritious food owed to its originality and medicinal values. This study aimed to evaluate the physico-chemical and microbiological qualities of locally produced raw goat milk prior any processing steps during storage. Milk samples passed organoleptic test and C.O.B. test were mostly (88.89 %) failed in alcohol test. AOAC Official method of oven drying method, Kjeldahl method and Soxhlet method were performed in physico-chemical analysis where results obtained were partially in lined with reported literature due to subjective factors of breeds, geographical areas and feeds. The locally produced raw goat milk's compositions are high in water content and low in fat percentage. Initial total plate count, coliform count and proteolytic count tested were 3.44 log cfu/ml, 1.87 log cfu/ml and 1.97 log cfu/ml, respectively. Storage time showed significant effect on the bacterial counts ($p>0.05$) of milk samples. Shelf-life of milk samples were kept up to 12 hours under ambient temperature (3.95 log cfu/ml) had not exceeded the standard limit. The shelf-life of the milk samples were extended up to 16 days storage under refrigerated temperature of 4°C. The microbiological quality of the milk samples showed a significant bacteriological growth upon prolonged storage and high initial coliform count indicates possible poor hygienic practices at farm level.

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Introduction

The study of goat milk or its products are important and has been recognized in a number of proceedings in national and international conferences (Gruner and Chabert, 2000). Consuming milk and products could be one of the routine practices upholding in Malaysia's population. In Malaysia, the total milk production of small-scale dairy farms may not fully support the local market needs (Lye *et al*., 2013; Bamaiyi *et al*., 2014; Alyaqoubi *et al*., 2014). The local milk production may lack concern, especially for goat milk and yet it is one of the potential industries to develop. Proximate analysis helps to determine the principle constituents of milk include fat, protein, total solid, lactose and ash. Additionally, milk contains hundreds of minor constituents include milk fat, vitamins, metal ion and flavor compounds, which contribute massive impact on the nutritional technological and sensory properties of milk and dairies (Armstrong, 1995). In comparison with cow milk, goat milk has a better digestibility, buffer capacity and its particular therapeutic value in medicine and human nutrition (Haenlein, 2004). It has been clearly proven that consuming of goat milk improves the state of health and wellness of the human body, reduces the risk of developing disease especially allergies (Park and Haenlein, 2006).

Raw goat milk samples that have not been pasteurized or homogenized were used in this study. High nutrient content of raw goat milk may have a mixed microflora which is different to that found in raw cow milk, with the microbial diversity the result of multiple factors. Literatures that related on the microbiological quality of goat milk today are insufficient for the public. It is essential to check on quality control and optimize safety concern in goat milk to gain consumer confidence due to the growing interest in present (Silanikove, 2000).

In general, cow milk is subjected to strict hygiene and quality regulations controlled while microbiological quality standards for production and distribution of goat milk are seems to be more relaxed (Muehlherr *et al*., 2003). There is unlikely similarity between goat milk compositions from other milk sources, and thus the quality standards for the milk

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from small ruminant animals should be evaluated based on the individual milk source (Morgan *et al.*, 2003 and Zweifel *et al*., 2005). In the local market, a total plate count (TPC) less than 10^6 cfu/ml used as a guideline or standard by Milk Collection Centers (MCC) as a Price Incentive Program (Chye *et al*., 2004; Boniface, 2012). The milk selling price was calculated based on the bacterial count in which a high microbial load may pose economic loss to local dairy farmers.

The general total bacterial count regardless of milk types used as the main quality and safety assessment may not adequate to be a proper guideline. Researchers (Wasiksiri *et al*, 2010) from Thailand are attentive about the importance of quality aspects of raw goat milk. Raw goat milk samples were collected from 5 different farms in Thailand and the number of tests was done on these milk samples to obtain the general figures to represent standard quality of raw goat milk that produced in Thailand. Investigation on the microbiological quality like Total Plate Count (TPC), Proteolytic count and Coliforms count as quality and safety indicators was rarely found in Malaysia. *Staphylococci, Escherichia coli, Staphylococcal enterotoxin, Campylobacter, Salmonella, L. monocytogenes* are commonly detected in raw goat milk according a six years long term study in Queensland, Australia (Eglezos *et al*., 2008).

Storage of locally produced raw goat milk after milking is one of the important factors in ensuring their quality and safety. Fresh milk drawn from a healthy ruminant usually contains a low micro activity particularly with bacterial load of less than 10^3 cfu/ml (Chatterjee *et al*., 2006; Lingathurai *et al.*, 2009), but the load may increase up to 100 fold or more once it is stored for sometime at ambient temperature (Lingathurai *et al*., 2009). Chilling of raw goat milk is a common practice to keep the milk fresh and prevent the growth of non-psychrotrophic bacteria. Yet, prolonged storage for 10 to 16 days of raw goat milk samples under cold temperature may not safe. According to Yamazi *et al.* (2013), the total loads of mesophilic bacteria, coliforms bacteria, *Escherichia coli* and psychrotrophic bacteria of milk stored for 48 hours or longer were relatively higher than the storage for 24 hours or less. Thus, the storage study at different temperatures and durations should be accounted for public notifications due to food safety concerned. The present study aimed to characterize the microbiological quality changes of locally produced raw goat milk samples stored at different temperatures and prolonged durations to provide an apparent indication to the public.

Materials and Methods

Sample collection

Samples of fresh goat milk were collected from a small-scale farm named Sungai Buloh Farm located at Lot 115-F, Kg Melayu Sg Buloh, 47000 Sg Buloh, Selangor which less than 1 hour distance from the laboratory of University of Putra Malaysia. Milk samples were collected in the early morning to ensure the freshness just after the milking was done. All raw milk samples were obtained under aseptic conditions from healthy goat, to avoid any contamination which can influence the analysis (Suguna *et al.*, 2012). Samples were collected in a sterilized Scotch bottle and then kept in an ice cooler box and delivered to laboratory to perform analysis at less than 4°C during 1 hour transport. Upon arrival, milk samples were divided into 3 groups to perform different tests, including platform test, proximate analysis and microbiological analysis. Samples were tested and analyzed immediately upon arrival at the laboratory, while some of the samples stored in a freezer at -20°C until required. A total of 4 batches sampling was carried out from Sungai Buloh Farm from time to time along a year of research study.

Platform tests

Samples of raw goat milk were made into 3 aliquots (25 ml each) in sterilized 50 ml Scotch bottle. Each platform test, including organoleptic test, C.O.B test, and alcohol test was being repeated 3 times (Wasiksiri *et al*., 2010). In the organoleptic test, texture, color and smell of the milk samples were being observed. Firstly, milk sample was smell for any off or sour aroma, then followed by visually observing on the appearance of the milk to check whether there is coagulation or no coagulation. The milk samples were also tested for temperature by using a thermometer in which milk sample should not be warmer than 4°C. In the C.O.B test, 10 ml of milk sample was boiled in a test tube in a water bath for 5 minutes. If there is clotting, coagulation or precipitation, the milk sample has failed the test and therefore should be rejected. Lastly, samples were performed with the alcohol test. The alcohol test depends on the instability of the proteins if the levels of acid are increased and acted upon by the alcohol. The measured 25 ml of milk sample was mixed with an equal amount of diluted 68% ethanol (Merck) solution in a small bottle or test tube. A good quality of milk sample shall have no clotting, precipitation and small clumps. All platform tests were done in four respective batches of raw goat milk samples which obtained at several different times and each

test was repeated at least 3 times with analysis on each batch of milk samples.

Physico-chemical analysis

In the proximate analysis of raw goat milk samples, moisture content was determined using the oven drying method (AOAC 934.01, 2005), ash by furnace drying (AOAC 942.05, 1990), crude protein (N=6.25) using Kjeldahl method (AOAC 984.13,1990).and crude fat using Soxhlet method (AOAC 920.39, 2000). Proximate carbohydrate content in the milk sample was determined by difference. It was calculated by deducting the total amount of moisture, ash, protein and fat with 100%. Steps in the few analysis were repeated until a constant reading was obtained.

Assessment of pH and titratable acidity of the goat milk samples were determined according to the methods (No. 947.05) given in AOAC (2000). The pH of the milk sample was determined using an electronic pH meter (Mettler Toledo 8603, Zurich Switzerland). Milk samples should range from pH 6.5-6.7 and sample which out of the pH range considered acid milk and being rejected. The total acidity content was expressed as percentage of lactic acid. All physico-chemical tests was done on four respective batches of raw goat milk samples which obtained at several different times and each test according AOAC (2005), AOAC (2000) and AOAC (1990) shall be repeated at least 3 times with analysis on each batch of milk samples.

Microbiological analysis of storage study

The volume of 25 ml raw goat milk was made into 171 aliquots in total. A number of 63 aliquots were stored at ambient temperature of 27±1°C and 108 aliquots were stored at chilled temperature of 4±1°C respectively. All the aliquots were later checked for mesophilic total plate count, mesophilic proteolytic count, mesophilic coliform count and psychrotrophic total plate count after particular storage duration correspondingly. Twenty five milliliters of milk sample (each aliquot) were dispensed into a sterile stomacher bag (Bagmixer 400, Model L, Interscience, France) containing 225 ml of 1 % peptone water (ISO, 2001). The mixture was then homogenized with stomacher for 90 seconds. ISO 6887-1 (1999) was referred to the common rules of the preparation of the initial suspension and decimal dilutions. Subsequent serial decimal dilutions of milk were prepared in sterile 1 % peptone water. Each aliquot of the dilution with a volume of 100 µl was pipette and spread plated in triplicate onto agar plates.

The total plate count of mesophilic bacteria was firstly enumerated by the plate count technique by using plate count agar (Merk) medium stored at 37±1°C for 48 hours. Coliform count was determined by enumerated on Eosin Methylene Blue (EMB) agar (Merck) and incubation at 37°C for 48 hours. EMB agar contains methylene blue, which able to inhibit the growth of gram-positive bacteria (Atlas and Bertha, 1997). Proteolytic count was determined on skim milk plate count agar (Merck), stored at 37±1°C and observed after 48 h. The numbers of bacterial colonies on the plate were counted and expressed in log colony forming unit (CFU) per milliliter. In skimming milk agar plates, only bacterial colonies that are surrounded by a clear halo were considered as proteolytic bacteria.

Similar to the mesophilic bacteria counting techniques, psychrotrophic bacteria of the total plate count were carried out by using the same plate count agar (Merck) medium. Samples were checked on their bacterial count in every interval of 2 days where storage up to 16 days. All inoculated plates were then incubated at a lower temperature of 4±1°C for 10 days. The numbers of bacterial colonies were also counted and expressed in log cfu/ml.

Statistical analysis

All microbiological plate counting techniques were done in four respective batches of raw goat milk samples which obtained at several different times and each test was repeated at least 3 times with analysis on each batch of milk samples. The significant difference was determined by using analysis of variance (ANOVA) and followed by Duncan's multiple range tests (DMRT) for means comparison. All the statistical analysis was performed by using IBM SPSS Statistics (Statistical Product and Service Solutions) Version 16.0 for windows (SPSS, 2008) and MINITAB 14. Values of $P<0.05$ was used to indicate the significant deviation.

Results and Discussion

Platform tests

The observations from platform tests were recorded are presented in Table 1. In the very firstly organoleptic test, the obtained raw goat milk samples are showing 100% normal with white milky color from the aspect of visualization, good natural smell and no sediment contamination. In the C.O.B test, high percentage of 77.78% of total samples was detected normal by not showing any precipitation after boiling in the water bath. Abnormal milk samples or sour milk developed acid (>0.2% acidity) and showed coagulation due to heat treatment which

Table 1. Platform tests observations in raw goat milk samples

Platform tests	Observations
Organoleptic Test	All samples were tested normal with white colour, good natural smell, and no sediment contained.
Clot on boiling Test (C.O.B)	77.78% samples were detected normal by not showing any precipitation.
Alcohol Test	11.11% samples were detected normal by not showing any precipitation.

Note: Raw goat milk was collected from the Sungai Buloh Farm, Selangor and checked on three major platform tests with observations. The percentage of normal samples was counted based on a total of 4 batches samples in a triplicate form.

is the result of dissociation of calcium caseinate salt (Agrinfo, 2011). In the dairy industry, such abnormal milk samples failed in C.O.B test means cannot stand the heat treatment in milk processing and thus being rejected.

High percentages up to 88.89% of total samples were detected abnormalities and failed in alcohol test. Most of the milk samples showed at least little precipitation or small clumps formed when tested with 68 % concentration ethanol (Merck). This result is shown a similar outcome as the raw goat milk study made by Wasiksiri *et al*. (2010) in lower southern Thailand area. Hundred percentages of their samples failed in alcohol test as well. Alcohol test is mainly based on the instability of the proteins when the concentration of acid and rennet raised and thus acted upon by the alcohol. According to Horne and Parker (1982), different proportions of the individual caseins resulting goat milk have low ethanol stability. The study was also done found that fresh goat milk, precipitated when equal amount of 44 % ethanol added in while fresh cow's milk will only formed precipitation as 70 % ethanol mixed (Guo *et al*., 1998). Besides, the study also suggested that low ethanol stability characteristic of goat milk, possibly due to ratio of sodium to potassium (Na/K). A lower ratio of Na/K found in goat milk (0.189 mg/100) compared to cow's milk (0.333 mg/100g) has contributed to this outcome (Anjaneyulu *et al*., 1985; Darnton-Hill *et al*., 1987; Chandan *et al*., 1992).

A previous study from Lou and Gou (1991) found that adding in sodium to increase the salt concentrates in goat milk able to improve their ethanol stability of casein micelle. Consequently, the result from alcohol test may not represent the freshness and microbiological quality of goat milk due to their low ethanol stability. Platform tests are simple and quick to know the freshness and the quality of the milk samples prior to any detailed analysis. It helps us to reject odd milk samples rapidly, but may not all apply to raw goat milk and thus further quality analysis is obligatory.

Physico-chemical analysis

A step further on physico-chemical analysis, the mean values with standard deviations of proximate physical characteristics and chemical compositions of raw goat milk sample were presented in Table 2. A comparison was made on each value of properties with the values which recently reported by Soliman (2005), Imran *et al.* (2008) and Mayer and Fiechter (2012). The raw goat milk samples were mainly structured of water and the remaining are the total milk solids (6.43%) which are the sum of fat, protein, carbohydrates and minerals. The water content in the milk samples was relatively high if compared to the reference range of 80% to 90%. In contrast, the remained contents total solids of ash, protein, fat and carbohydrates are much lower than the range of reference. The main composition of water works out as a medium for a solution and colloidal suspensions for the other components present in milk (Imran *et al*., 2008). The pH of the milk samples found in the current study (6.56 ± 0.32) falls in the reference range and in better agreement with the findings from previous investigations (6.59 ± 0.04) (Mayer and Fiechter, 2012). There is no significant difference in the pH category of the milk samples between the current study and the reference study.

Both the total crude protein (2.31%) and total fat content (2.54%) in raw goat milk samples were also found significantly lower than those previously reference findings. The primary constituent of the milk protein is casein which contributes approximately 75% of the total milk proteins is having high nutritional value (Hassan, 2005). The other about 25 % of whey proteins consist of lactoalbumin, lactoglobulin, serum albumin, immunoglobulin, lactoferrin and lysozyme (Greppi *et al*., 2008). Total fat contents in goat milk are predominated by smaller fat globules where 90% of the fat particles in goat milk were less than 5.21 µm in comparison to cow milk fat particles, 90% of which less than 6.42 µm (Tziboula-Clarke, 2003). This related property of smaller fat globules with broader surface area was giving the benefits of easy

Table 2. Proximate compositions and physico-chemical characteristics of raw goat milk

Analysis	Proximate compositions of raw goat milk		
	Current study	Range from other references*	Reference**
Moisture (%)	93.57±0.01[a]	87.38±0.06[b]	87.03±0.15[SE]
Protein (%)	2.31±0.13[a]	2.95±0.99[b]	3.56±0.03[SE]
Fat (%)	2.54±0.17[a]	3.74±0.39[b]	4.14±0.05[SE]
Ash (%)	0.45±0.00[a]	0.81±0.03[b]	0.82±0.00[SE]
Carbohydrates (%)	1.13±0.28[a]	4.44±0.02[b]	4.45
pH	6.56±0.32[a]	6.59±0.04[a]	-
Titratable Acidity (%)	0.07±0.00[a]	1.35±0.38[b]	-
Total Solids (%)	6.43±0.01[a]	12.62±0.06[b]	-

(*) Source from Mayer and Fiechter, 2012, Imran et al., 2008 and Soliman, 2005.
(**) Source from U.S. Department of Agriculture, Agricultural (USDA) National Nutrient Database for Standard Reference, Release 26. (2013).
([SE])Values are mean ± standard error.
Values are mean ± standard deviation.
Different alphabet superscripts are significantly different ($P<0.05$).

to digest and quicker lipase activity (Chandan *et al*., 1992). Total lipids in goat milk found to have higher physical characteristics compared to cow's milk, but may vary among different reports (Anifantakis, 1986; Park, 2006). Nevertheless, total protein contents and total fat contents in goat milk were found to be different according the breed, feed and seasonal effects. Fernandez *et al.* (2008) stated that the total amount of fat content and protein content was higher at the beginning and lower towards the end of lactation when milk volume decreased. According to the studies in some seasonal areas, milk yield is high in summer while the fat and protein contents are low and during winter, the milk yield may low, but the fat and protein concentrations are higher (Haenlein and Wendorff, 2006; Mioč *et al*., 2008; Pal *et al*., 2011).

Ash content obtained from the raw goat milk sample was not really in line with the values that are reported from previous studies. It was slightly lower than the reference values. Ash is the remained inorganic residue after heating while moisture and organic matter were being removed. Ash content represents the total amount of minerals present within the milk samples and major constituents in ash content comprised of oxide and chloride of mineral elements (Imran *et al.*, 2008). Ash content measurement is important to represent the quality, microbiological stability and nutrition in a particular food product.

The proximate carbohydrate measurement (1.13%) in the current study was obtained by difference from the total amount of water, ash, protein and fat with 100% and it was showing significant difference from the reference values of 4.45%. Lactose, one of the most important carbohydrate or milk sugar composed of galactose and glucose in milk. Synthesizing of lactose from glucose in the mammary gland required active contribution of the milk protein of α-lactalbumin (Ebner and Schanbacher, 1974). An average composition of lactose present in goat' milk reported by Park *et al.* (2007) compiled from previous study is 4.1% and it showed approximately range 0.2 to 0.5% lower than cow's milk (Haenlein and Caccese, 1984; Chandan *et al*., 1992). Other than the lactose component, other carbohydrates that found in goat' milk is glycopeptides, oligosaccharides, glycoproteins and nucleotide sugars in small amounts (Larson and Smith, 1974). It is always helpful to those who suffer from lactose-intolerance when consuming cow's milk since lower lactose content in goat milk.

The value for the titratable acidity of raw goat milk samples obtained from current study was only 0.07% lactic acid and shown significant differences with reported values of 1.35% from an earlier study (Imran *et al*., 2008). There is no lactic acid in fresh milk virtually and the acidity of milk samples is due to carbon dioxide, phosphates, citrates, caseins and whey proteins. The acidity of milk could be varied due to factors of lactation period where it was frequently

Table 3. Total aerobic, coliform and proteolytic bacteria loads (log cfu/ml) storage for 24 hours at ambient temperature of 27±1°C

Hours	Aerobic bacteria	Coliform bacteria	Proteolytic bacteria
0	3.44±0.8^{a}	1.87±1.53^{a}	1.97±1.63^{a}
4	3.71±0.7^{a}	1.88±1.56^{a}	2.84±0.8^{ab}
8	3.80±0.8^{a}	2.46±1.60^{ab}	3.88±0.66^{bc}
12	3.95±0.9^{a}	3.24±1.04^{ab}	4.20±0.85^{c}
16	5.20±0.6^{b}	4.20±1.33^{cde}	5.05±0.76^{cd}
20	5.94±0.5^{b}	4.97±1.50^{de}	6.16±0.81^{de}
24	6.83±0.7^{c}	5.55±1.69^{e}	6.73±1.08^{e}

Bacteria loads of raw goat milk samples was checked in every interval 4 hours. The numbers represent mean ± sd of three replications. Values followed by different smaller letters within a column are significantly different in Duncan test (P<0.05).

having lower acidity towards the end of the lactation stage (John, 1996). In addition, high moisture content in the milk samples may also cause a fact of lesser precipitation of tri-calcium phosphate and resulting in low titratable acidity value obtained.

The results recorded from the current study were partially in line with the values of reported literature. The results revealed that locally produced milk composition may not constant and subjected to extensive variation within breeds with breeds, age, parity, season, geographic areas and duration of lactation. According to Park (2006) and Raynal-Ljutovac *et al*. (2008), the breed is another important factor which strongly affecting the composition of goat milk. The compositions of goat milk could be varied among breeds within a species or yet between each goat within a breed. The compositions might even differ from the first milk and the last milk drops during milking depending on feeding and water. Literatures according to Li and Li (2011), the milk fat, protein, whole milk solids and grease milk solids showed regular changes in Hebei and Xuzhou in China throughout a year of study. Besides, the reported reference values were most likely compiled from studies of different areas and are plainly differing to a certain level of results attained in the current study.

Goat milk is different from cow milk, but having better digestibility, alkalinity, buffering capacity or even particular remedial values in medicine and human diet (Haenlein and Caccese, 1984; Park and Chukwu, 1989; Park, 1994). Physico-chemical characteristics of the locally produced goat milk may not identical in different batches of sampling. Studies of the locally produced goat milk are limited and references are highly depending on outsources especially from European standard, Thailand, Indonesia, New Zealand and China. It was not confident if depend on the outsource information solely as the compositions of goat milk may different since it could be easily affected by locations, weather and breeds. An update information about the physico-chemical characteristics of locally produced raw goat milk gained from the current study. The economic values of goat milk products are rising and thus further information about the nutritional composition of these products is essential.

Microbiological analysis of storage study

Storage study of raw goat milk samples for 24 hours was done at ambient temperature 27±1°C to check the bacteria concentration as shown in Table 3. The total plate count (aerobic bacteria), coliform count (coliform bacteria) and proteolytic count (proteolytic bacteria) were 3.44 log cfu/ml, 1.87 log cfu/ml and 1.97 log cfu/ml respectively at zero hours. Generally, all three categories of bacteria concentration showed a trend of increasing when storage time getting longer. The p-value for variable of storage time shows smaller than 0.05 (P-value<0.05) has concluded that it has a significant effect on the total plate count, coliform count and proteolytic count.

An initial bacteria count represents the level of contamination, freshness and quality of the milk samples from farms. Normally, high bacteria loads mean the milk samples were actually contaminated during early milking period. Variance of storage temperature, cleanliness of storage equipment, milk handling and health of udder could be the most potent factors responsible for the contamination. Total aerobic bacteria concentration recorded once upon arrival in the laboratory was 3.44 log cfu/ml. A reference according the Malaysians Food Act 1983 and Food Regulations 1985, the total aerobic bacteria

Table 4. Amount of aerobic, coliform, proteolytic and psychrotrophic bacteria (log cfu/ml) obtained from raw goat milk which storage for 16 days at cold temperature of 4±1°C

Days	Aerobic* bacteria	Coliform* bacteria	Proteolytic* bacteria	Psychrotrophic** bacteria
0	3.01±0.67^{a}	1.87±1.53^{a}	1.97±1.63^{a}	3.03±0.19^{a}
2	2.92±0.54^{a}	1.76±1.43^{a}	2.79±0.66^{ab}	3.08±0.25^{a}
4	2.99±0.66^{a}	2.73±1.78^{a}	2.97±0.62^{bc}	4.77±0.06^{b}
6	3.57±0.41^{ab}	2.78±0.40^{a}	3.76±0.45^{cd}	4.81±0.13^{b}
8	4.09±0.44^{b}	3.18±0.62^{a}	4.27±0.53^{de}	5.84±0.09^{c}
10	5.21±0.84^{c}	4.59±1.17^{b}	4.85±0.45^{e}	5.86 ±0.08^{c}
12	5.59±0.75^{cd}	4.78±1.08^{b}	5.90±0.57^{f}	6.77±0.09^{d}
14	6.08±0.69^{d}	5.16±0.74^{b}	6.77±0.48^{g}	6.95±0.05^{d}
16	7.74±0.73^{e}	5.63±0.56^{b}	7.77±0.57^{h}	7.97±0.11^{e}

Bacteria loads of raw goat milk samples were checked in every interval 2 days.
(*) Bacteria count obtained after plates were incubated at 37°C after 24 hours.
(**) Psychrotrophic count obtained after plates were incubated at 7°C after 10 days.
The numbers represent mean ± sd of three replications. Values followed by different letters within a column are significantly different in Duncan test ($P<0.05$).

concentration in milk, which safe for consumed should not exceed 5.0 log cfu/ml (Food Act 1983 and Food Regulations 1985 – Act 281, 2005).

A recent study was done by Suguna *et al.* (2012) in Penang island, Malaysia showed the total plate count range from 4.2 to 4.5 log cfu/ml. The present study of total plate count has no significant difference from zero hour to 12 hours storage and they only exceed the microbiological standard limit (5.0 log cfu/ml) when storage up to 16 hours (5.20 log cfu/ml). Bacterial count might rise up to 100 fold or even higher when stored at ambient temperature for prolonged duration (Chye *et al*., 2004). Present study results revealed that the good quality raw goat milk was actually could be stored up to 12 hours at ambient temperature 27±1°C and the total aerobic bacteria concentration is within the range of microbiological standard. The coliform count is another important indication of safety evaluation and thus the safety of milk samples should not be judged from total plate count solely.

Coliform count checked from the raw goat milk sample was initially recorded as 1.87 log cfu/ml at zero hour (Table 3). The coliform bacteria concentration was showing a gradually increased and there is no significantly different when storage time prolonged up to 16 hours (4.20 log cfu/ml) and continuously. Malaysian Food Act (1983) and Food Regulations Act (1985) revealed that coliform count should not exceed 1.7 log cfu/ml in milk samples (Food Act 1983; Food Regulations 1985 – Act 281, 2005). Samples of raw goat milk collected from farm showed contaminated and higher than the limit of coliform bacteria concentration in microbiological standard. The coliform bacteria load was getting higher when prolonged storage period. The high coliform count may indicate a problem contamination at farm level from infected udder, unsanitary milking practices or unclean container. Coliform bacteria could be ubiquitous in feces, manure and soil allowing easy dispersal of pathogens throughout the farm (Son *et al*., 2009; Lingathurai and Vellathurai, 2010). Burgess *et al.* (1994) also stated that the amount and diversity of microorganism present in milk mostly depending on microbial quality, the milking conditions, temperature and duration of storage.

Proteolytic bacteria count in raw goat milk samples shown in Table 3 was initially recorded as 1.97 log cfu/ml and regularly increased up to 6.73 log cfu/ml when storage until 24 hours long under ambient temperature. Proteolytic bacteria produced the proteolytic enzymes such as protease and lipase (Sorhaug and Stepaniak, 1997) resulting the coagulation, bitter flavor, rancid, putrid and yeasty flavor in milk (Cousin and Marth, 1976; Matta and Puni, 1999; Burdova *et al*, 2002). The data and information related to the limitation of proteolytic count for goat milk quality are unsatisfied for reference. In fact, the proteolytic concentration could be one of the helpful indicators to represent the quality and condition of milk samples since milk is categorized in protein rich food aspects.

According to the standard reference method, involving incubation of culture plates at 4°C to 7°C

for 10 days, is recommended for the determination of psychrotrophic colony counts for investigational purposes (Thomas, 1969). Three types of bacterial counts were checked on the raw goat milk samples which stored at 4°C in every interval of 2 days up to 16 days was presented in Table 4. There is no siginificant difference of microbiological counts from the reference standard of 5 log cfu/ml when stored up to 10 days in a preliminary observation. Thus, storage time was then extended up to 16 days for observation. The total amount of aerobic, proteolytic and coliform bacteria grew on the plates were recorded after incubation of 37°C for 24 hours. The total aerobic bacteria concentration was initially recorded as 3.01 log cfu/ml and it only showed significantly different after 6 days of storage. The aerobic bacteria concentration was showing no difference after 4 days of storage. It has only exceeded the reference standard (5 log cfu/ml) after stored for 10 days. Obviously, the growth of aerobic bacteria was much slower when milk samples were under cold temperature (4°C) storage. Storage temperature presents as one of the dominant factor to the bacteria count in raw milk. The duplication of bacteria could take more than 24 hours if the milk samples were stored under cold temperature of 4°C. Griffiths *et al.* (1987) found that the initial count and storage temperature are the major factors affecting the sample storage time until the bacterial count reached 7 log cfu/ml when they studied about the effect of cold temperature storage on raw milk samples.

The initial count of coliform bacteria was high when compared to reference standard of 1.7 log cfu/ml. The duplication of coliform bacteria was slow and the bacteria concentration will only show significant difference after 10 days where the raw milk samples stored under cold temperature. They took 7 times longer time to duplicate under cold temperature if compared to the records of the storage under ambient temperature. Coliform bacteria may grow slower under cold temperature, but numbers of them such as *E. coli* and *Klebsiella* spp. are well temperature tolerated and reboot fast with a favorable temperature and environment. Thus, these coliform groups throw in a particular risk on the spoilage and contamination of the raw goat milk sample though storage under cold temperature.

Within the proteolytic count category, the bacteria concentration was showing consistently increasing trend in comparison with the total plate count and coliform count when raw goat milk samples stored under cold temperature. The proteolytic bacteria concentration showed significantly different after the samples were stored for 2 days. Numbers of the proteolytic bacteria are psychrotrophic and one of the common examples is *Pseudomonas* spp. where account for more than 50% of the psychrotrophs (Chambers, 2002). These psychrotrophic bacteria able to grow below 7°C even though their optimum temperature ranges from 20°C to 30°C (McPhee and Griffiths 2011). The other bacteria counts may low and not showing any significant different after few days stored in the beginning exclusive of proteolytic count. It is clear to show that proteolytic count could be one of the helpful indicators for the shelf life and spoilage level of the raw goat milk samples. According to the study of McPhee and Griffiths (2011), products with longer storage time of more than 10 days comprises other types of microorganisms and the numbers of Pseudomonas spp. reached up to 68% of the population.

Obviously, cold temperature storage under 4°C could be effectively extended the shelf-life of raw goat milk samples. In my study, the duplication of the bacteria may take at least 6 times longer hours if storage under cold temperature compared to those stored at ambient temperature. Lafarge *et al.* (2004) did storage study of raw milk samples for 24 hours at 4°C found that there was a noticeable difference in the bacteria population composition during the storage period. Species that were not detectable in the beginning appeared and some existing species absent in the population after the cold storage. They also found that the amount of psychrotrophic bacteria increases significantly in 24 hours of cold storage.

Psychrotrophic bacteria account one of the major spoilage bacteria group present in raw goat milk samples or other dairies. Most of the psychrotrophic microorganisms belong to the genus *Pseudomonas* spp. and the most common species that isolated from milk is *Pseudomonas fluorescens*, who dominates the bacterial community at the time of spoilage (McPhee and Griffiths, 2011). Psychrotrophic bacteria produce the heat resistant enzymes, including proteolytic and lipolytic enzymes at low temperatures, which able to hydrolyze milk fat and protein structures leading to the forming of off-flavors (Ercolini *et al.*, 2009). According to the results obtained from the study, psychrotrophic count was getting higher and significantly different in every interval 2 days of storage under cold temperature.

Cold storage of the milk samples can mask the contamination caused by unhygienic conditions on a farm (Chambers, 2002). The quality of milk may not change and does not suffer during this cold storage time, but the growth potential of bacteria has increased significantly. As reviewed in the current study, the amount of total plate count may

low and still under limit even though storage up to 10 days under cold temperature. In contrast, the psychrotrophic count is closely approaching the standard limit of 5.0 log cfu/ml in the beginning of 4 days cold storage. Occurrence of psychrotrophs in farm during milk collection highlights the tendency of these bacteria to grow and reproduce once storage under low temperature. The current practices for collection and cold storage have improved the quality of milk and dairy products, but they have also led to a selection of psychrotrophic bacteria (McPhee and Griffiths, 2011). Thus, cold storage may not 100% guaranteed the quality of the raw goat milk as the psychrotrophic count could be one of the important factor accounts for the milk spoilage.

Conclusions

Goat milk shows its variable changes in physico-chemical composition, and microbiological quality depending on genetic factors, environmental conditions and goat farming practices. Raw goat milk samples analyzed showed high water content (93.57%) and low in other proximate composition, including protein content (2.31%), fat content (2.54%), ash content (0.45%) and carbohydrate content (1.13%) if compared with the reported literatures previously. Raw goat milk samples collected from the local Sungai Buloh Farm mostly failed in alcohol test (88.89%), generally passed in organoleptic test and clot on boiling test. There is a lack of current data on raw goat milk properties in Malaysia and thus more study is necessary in order to obtain the most updating data as a reference. An updated knowledge of the composition and nutritive value of the locally produced goat milk is of considerable importance for the dairy industry, food analysis and manufacture high quality local dairy products.

The total load of mesophilic aerobic bacteria, proteolytic bacteria and coliform bacteria in raw goat milk samples increased gradually after prolonged storage up to 24 hours at ambient temperature. The recorded values of TPC (5.20 log cfu/ml) exceeded the standard limit of Food Act (1983) and Food Regulations 1985 – Act 281 (2005) after storage of 16 hours. The shelf-life of raw goat milk samples are extended when stored under cold temperature of 4°C, up to 16 days. The occurrence of the psychrotrophic bacteria in local raw goat milk samples highlights the tendency of the bacteria to grow and multiply when stored at low temperature. High initial coliform count (1.87 log cfu/ml) exceeded the limit standard was not advisable for raw milk consumption or raw goat milk products proceedings due to food contamination concerned. The finding of this study provided updated information on locally raw goat milk in perspective of compositions and microbiological characteristics during storage. More detailed study about raw goat milk from different locations locally is encouraged to suggest a significant standard for reference purpose.

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