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EFFECT OF USING LEMON CITRUS COMPARED TO RENNET COAGULATION ON PHYSIOCHEMICAL YIELD AND SENSORY PROPERTIES OF CHEESE FROM COW'S MILK AND GOAT'S MILK

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ABSTRACT: This study was conducted to assess the yield and sensory characteristics of cheese produced from different milk sources (cow and goat using lemon citrus juice as coagulant compared with the commonly used rennet enzyme. The milk of the two species was collected from experimental dairy farms of animal production college (Kuku) and college of agricultural studies (Shambat), Sudan university of science and technology. After pasteurization at 85 ºC for 30 min and cooling to 40 ºC, the collected milk samples were divided into four groups (each five liters of goats' milk and cow's milk were treated with both Rennet enzyme and citric acid (Lemmon juice), respectively. Thereafter, the samples were incubated at 38 ºC. The results revealed that Cow's and goat's milk that treated by rennet enzyme coagulated after one hour of incubation, while both milks treated by citric acid was coagulated immediately after 10 min. Significant differences (P≤0.05) on physiochemical characteristics (protein, fat, total solids or TS, acidity, ash, lactose and solids-not-fat or SNF) between goat's and cow's cheeses were obtained. It was observed that cheese manufactured from goat's milk using citric acid aggulation recorded higher protein, fat, ash, T.S, pH and TNF, percentages, while acidic cow's cheese showed high moisture, lactose, and acidity. While, enzymatic goat's cheese revealed high percentages, of protein, fat, ash, TS, Ca and acidity, and enzymatic cow's cheese revealed high moisture, lactose, SNF and Phosphorus percentage. The sensory evaluations showed significant (P≤0.05) differences on taste, flavor, smell, texture, and overall acceptability. The best values for taste, color and overall acceptability were obtained from enzymatic (rennet) goat's cheese followed by enzymatic (rennet) cow's cheese, then acidic (citric) cow's cheese and acidic (citric) goat's cheese was the last. The perfect hardness and texture were obtained from enzymatic (rennet) cow's cheese. Moreover, the best smell was recorded for the cheese produced from cows' milk by enzymatic curdling. Where, the most acceptable flavor was attained by the cheese processed from cows' milk by citric acid coagulation. The results indicated that the weight of the final products obtained from cows' and goats' milk using citric acid coagulant was higher than that obtained from the same milks using rennet enzyme coagulant. It is possible to conclude that lemon citrus juice can be utilized as an alternative coagulant in production of local Sudanese white cheese for high yield and short curdling time which performed favourably with the commonly used rennet enzyme.

Keywords: Lemon Juice, Rennet, Coagulation Cheese Processing, Cows, Goats, Milk

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INTRODUCTION

Almost one third of the world's milk production is used in cheese manufacture, where the manufacture of cheese is considered a sort of milk preservation. Moreover, cheese is highly nutritious food with many diverse flavors and textures, which can use as a snack or as a part of dish or prepackaged convenience food (Quinee, 2004). Cheese is a widely consumed product by the general population; it is highly-concentrated, rich in proteins and lipids, essential amino acids, and minerals such as calcium and phosphorus (María et al., 2017).

In order to extend the shelf-life, quality of fresh milk and provide the consumer product with good flavor and high nutritive value, the process of cheese making has been adopted widely. The first step in cheese processing is the coagulation of raw milk in which κ-caseinolytic enzymes contribute to micelle aggregation. The common coagulant used is animal rennet, which has been the traditional coagulant in cheese manufacture for several years,

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(Ahmed et al., 2009). However, coagulation determines the yield, quality and final cheese properties (Akinloye and Adewumi, 2014). The individual characteristics of each cheese variety are due to the type of milk, microbial starter culture and the manufacturing procedure used (Ahmed, 1997). Recently, utilization of coagulants of plant origin in cheese processing has been found an increased interest because of several reasons such as religious consideration, nutritional behavior (vegetarianism) and sometimes for healthy risks (some countries forbid the use of genetically modified calf rennet (Roseiro et al., 2003). It has been reported that there are several commercial plant proteases (enzymes), such as bromelain and papain, used as coagulant in cheese manufacturing (Murata et al., 1987). In some European countries (Spain and Portugal) *Cynara sp*. is used as a plant coagulant in manufacturing cheeses from sheep and goats milk (Tejada et al., 2008). In Nigeria and many parts of West Africa, the traditional cheese makers use Sodom apple, (Calotropis procera) extract for clotting milk (Aworth and Muller, 1987). Extracts from pineapple, Banincasa cerifera and solanum toruum were use in Cheddar cheese making.

Plant extracts are characterized by high ability to hydrolyse the κ-casein, leading to curd formation, and they are also containing main enzymes responsible for β-casein hydrolysis (Roseiro et al., 2003). It is well known that lemon (*citrus limon)* juice is used in cheese manufacturing for fast coagulation property and high availability with a low price. For this purpose; the present work was initiated to evaluate the effect of lemon (citrus limon) juice as coagulant on physicochemical properties of cheese processed from milk of Sudanese cows and sheep compared with rennet enzyme.

MATERIALS AND METHODS

Milk test (basic properties of milk)

High quality fresh cow and goat milk was obtained from the experimental farm of animal production department, Agricultural college animal production, University of Sudan for science and technology; north Bahari, Khartoum then transferred to the laboratory of the same department where the whole experiment was conducted. The major investigated tests in milks of cows and sheep were fat (determined by Gerber method according Bradly et al., 1992), protein, (determined by Kjeldahl method according to AOAC, 1990) total solids (determined according to the modified method of AOAC 1990), solids not fat (was determined from the following equation:

SNF (%) = %TS - % fat

Also, ash content was determined according to (AOAC, 1990) in addition to the pH (using digital pH meter) and Titeratable acidity, (determined according to AOAC, 1990).

Coagulants

Rennet. Rennet (HR.HANSEN coagulant stick 50ltr, of milk), was obtained from veterinary pharmacy.

Preparation of lemon juice. Lemon fruits were purchased from local market; the fruits were cut and then squeezed into a clean bowl. The extracted content was sieved, measured with measuring cylinder and thoroughly mixed with equal volume of fresh clean water.

Cheese processing

Twenty liters of raw cows and goats milk were used to manufacture cheese from milk of the above species using both rennet and lemon juice as coagulants to compare physiochemical properties of cheese produced from the two milks. Milk quantity was divided into four patches each 5 liters, (each patch was coagulated by both rennet and lemon juice). The following illustration shows cheese processing steps:

Raw milk was pasteurized at (85 C for 30 minutes)
↳ **Cooling down the milk in iced water to 45 C**
↳ **Addition of coagulants and stirring** [Rennet (0.1dissolved in water) and lemon juice (in a separated experiment)]
↳ **Adding salts** [Addition of Na Cl (180 gm (w/w) and 0.5% of calcium chloride (w/w)].
↳ **Clotting (occurred in 30 minutes) and Whey drainage**
↳ **Weigh fresh cheese to determine yield**

Determination of cheese yield

The final product of the white cheese from the species milks was weighted after three days of manufacturing and the yield of every replicate recorded by using sensitive balance. The final weight was determined by the following equation:

$$\% \text{ of cheese yield} = \frac{\text{Grams of cheese produced} \times 100}{\text{Grams of milk used}}$$

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Sensory evaluation

Ten postgraduate students were randomly selected to evaluate cheese sensory characteristics that include taste, color, flavor, texture and overall acceptability.

Statistical analysis

The data were analyzed using (ANOVA) completely randomized design. Mean separation was done using Duncan's multiple range test.

RESULTS AND DISCUSSION

Physiochemical properties of goat milk compared to Cows, milk

Table 1 shows the physiochemical composition of the two species milk, the results revealed significant (P≤0.05) differences in majority of physiochemical properties of goats and cows' milk. However, minerals (Calcium and phosphorus) in the two milks (goats and cows) secured insignificant (P≥0.05) different. It was observed that protein and SNF in goat milk (3.500±0.329, 9.200 ±0.521) were higher than that found in cow milk (3.300±0.329, 9.08±0.521). The average of Protein content of goats' milk in this study was similar to that estimated by Wilson (1984) 3.4%. The average of Lactose in the present study was 4.85 % is similar to that found by Elamin and Wilcox (1992) 4.2%. Moreover, (Byoumi, 1990) estimated it as (5.5%), the difference may be due to the stage of lactation, as documented that, lactose % increased gradually at the commence of parturition until it reached 5.58% at the tenth day. Generally lactose content of goat milk varies greatly depending on feeding and watering conditions (Yagil and Etzion, 1980 and Yagil, 1994). Total solids and ash values in the presents study both were higher than those obtained by Aisha (2009) 11.3% and (Rehab, 2013) 0.73% respectively. Calcium and phosphorous concentrations in this study were lower than those found by Aisha (2009) and Gorban and Izzeldin (1997), the difference might be attributed to seasonality. The acidity in the present study was higher than that reported by Aisha (2009) 0.18%, and similar to that reported by Karim and Gooklani (1987) the difference may be due to breed factor. Furthermore, PH value in this study is similar to that reported by Mahmud (2009) and higher than that obtained by Rehab (2013) differences may be due to breeds and also, may be due to the analytical procedure applied, or to lactation stage.

Table 1 - Physiochemical analysis of cows and goats' milk.

Parameters	Treatments (Mean ± S.E)		Level of significant
	Cow Milk	Goat Milk	
Moisture	87.720^{a}±0.670	87.250^{a}±0.670	***
Fat	3.20±0.115	3.550±0.115	***
Protein	3.300^{a}±0.329	3.500^{b}±0.329	***
Ash	0.790±0.306	0.840±0.306	*
Lactose	4.990^{a}±0.163	4.86^{a}±0.163	*
TS	12.280^{a}±0.618	12.75^{a}±0.618	***
SNF	9.08^{a}±0.521	9.20^{b}±0.521	***
pH	6.660 ^{a}±0.231	6.35^{a}±0.231	NS
Acidity	0.180^{a}±0.029	0.170 ^{a}±0.029	***
Calcium	117.000±7.627	121.500±7.627	NS
Phosphorus	85.000±4.082	88.000±4.082	NS

TS= total solids, SNF= solids-not-fat. In this table and the successive one; mean in the same column not followed by the same common letter(s) differ significantly at (P < 0.05).

Physiochemical characteristics of cheese of cows and goats' milk

The data pertinent to physiochemical characteristics of goat's and cow's milk cheese is illustrated in Figure 1 The statistical analysis revealed significant (P≤0.05) differences among cheese physiochemical characteristics for the two species. It was noticed that acidic cheese prepared from goat's milk had higher contents of protein, fat, ash, TS, TNF and phosphorus. While acidic cheese from Cow's milk; showed high moisture, lactose, Ca^{++} and acidity. Moreover, enzymatic goats' milk cheese secured high percentage, of protein, fat, ash, TS, Ca and acidity. Whereas, cow's milk cheese which coagulated by enzyme revealed high percentage, of moisture, lactose, ph., TNF and Phosphorus. The average moisture of goat's enzymatic cheese in the present work (61.33 %) is similar to that demonstrated by Allahgabo (1986) he also found 61.33% for the moisture of cheese processed from goats' milk. The average protein (14.33%) obtained in this study is higher than that found by Adorkour (1992) 12.36% but,

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similar to that reported by Abdel Razig (1996). The fat percent in this study was also significantly (P≤0.05) higher (15.67%) than that found by Allahgabo (1986), who reported fat content of 12.65%. The total solid calculated in the present work is 36.67% however, was slightly lower than that reported by Ibrahim (1999) 36.75 %, and also, lower than that found by Mortada et al. (2013) 33.77%.

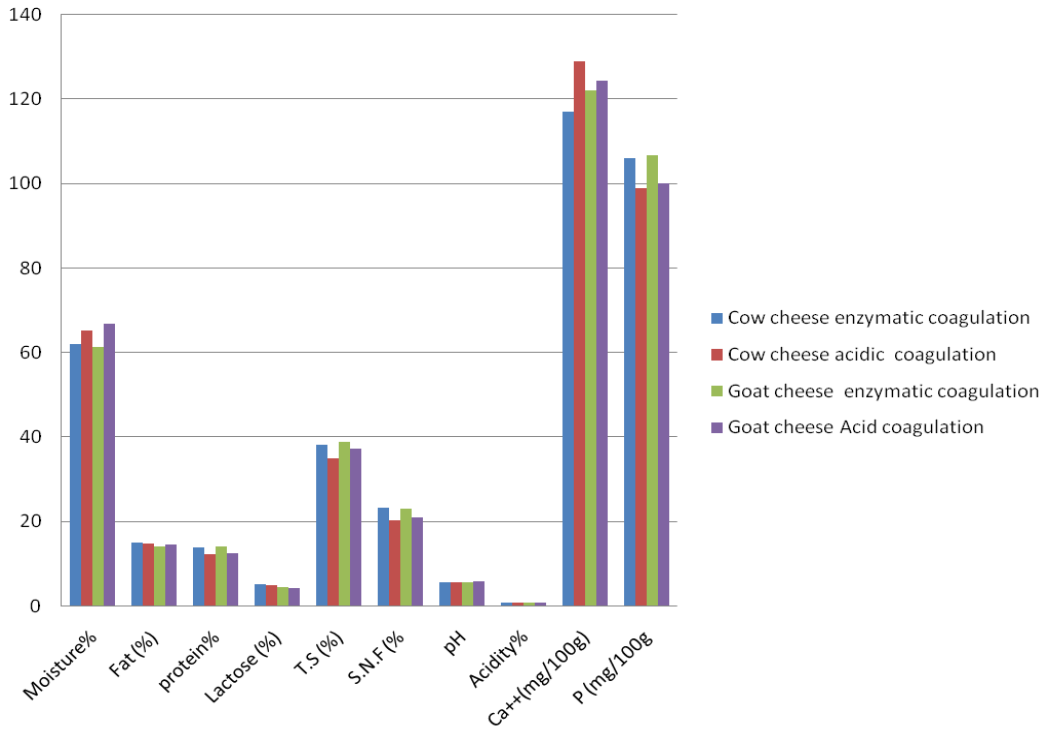

Figure 1 – Physiochemical characteristics of chees of cows and goats' milk

Effect of type of milk and coagulation agents on sensory evaluation of Cheese

Table 2 demonstrates the sensory evaluation of cheese processed from cows and goat's milk using rennet and lemon juice as coagulants. The statistical analysis revealed significant (P≤0.05) effect on all cheese sensory characteristics (taste, flavor, smell, texture, color and overall acceptability). The evaluation of panelists disclosed that best value for the taste, color and overall acceptability were obtained from enzymatic (rennet) goat's cheese followed by enzymatic (rennet) cow's cheese, then acidic (citric) cow's cheese and the last one was acidic (citric) goat's cheese treatment. Whereas, the best preferences for hardness and texture were obtained respectively, from enzymatic (rennet) cow's cheese, acidic cow's cheese, enzymatic (rennet) goat's cheese and acidic (citric) goat's cheese. However, the best values of smell were attained from enzymatic (rennet) cow's cheese, acidic (citric) cow's cheese, acidic (citric) goat's cheese and enzymatic (rennet) goat's cheese respectively, while the best flavor was recorded by the cheese manufactured from acidic (citric) cow's cheese, then enzymatic (rennet) goat's cheese followed by enzymatic (rennet) cow's cheese and acidic (citric) goat's cheese respectively.

Table 2 - Effect of coagulants on Sensory Evaluation of manufactured cheese from cow and goats' milk

Parameters	Treatments (Mean±SE)				Grant mean	Level of significant
	cow's cheese (Enzymatic)	cow's cheese (Acidic)	Goats' cheese (Enzymatic)	Goats' cheese (Acidic)		
Smell	7.70±0.70	7.30±0.75	6.50 ±0.79	6.90±0.64	7.10±0.35	***
Color	7.90±0.59	7.60±0.65	8.80±0.57	7.20±0.49	7.88±0.29	**
Flavor	6.90±0.66	7.40±0.67	7.20±0.68	6.50±0.83	7.00±0.35	**
Taste	7.10±0.67	7.50±0.64	7.80±0.63	6.30±0.58	7.18±0.32	***
Hardness	8.10±0.71	8.50±0.45	7.50±0.62	5.60±0.60	7.43±0.34	**
Texture	8.20±0.51	7.90±0.59	7.90±0.60	5.80±0.55	7.45±0.31	**
Over all acceptability	8.20±0.44	8.00±0.56	8.10±0.46	6.40±0.54	7.68±0.27	**

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The final Cheese yield

Figure 2 shows the effect of co-aggulation type on final weight, it was found that the white cheese obtained from cow's milk that coagulated with rennet enzyme exerted lower total weight than the that obtained from the same milk using Lemmon juice (citric acid coagulant), while the cheese obtained from goat's milk using rennet enzyme coagulant was recorded lower total weight than the that obtained from the same milk using Lemmon juice (citric acid coagulant), where the values of weight of the final Sudanese cheese (Gibna Bayda) obtained for the two treatments of cow's and goat's white cheese using rennet and acid coagulant were (0.640 kg, 0.735kg, 0.720kg and 0.785kg, respectively. The findings revealed that the weight of the final products obtained from cow's and goat's milk using citric acid coagulant were higher than that attained from the same milk using rennet enzyme coagulant. This may possibly be due to the fact that goat milk has higher total solid fat and protein content than cow milk. These findings were not in line with those obtained by Akinloye and Adewumi (2014). They found that the yield of cheese produced by using lemon juice extract were significantly lower than the other 2 coagulants used in their experiments.

It is possible to conclude that *lemon citrus* juice can be utilized as an alternative coagulant in production of local Sudanese white cheese for availability, high yield and short curdling time which performed favourably with the commonly used rennet enzyme. Further researches are needed to improve cheese yield and sensory characteristics using lemon juice as coagulant.

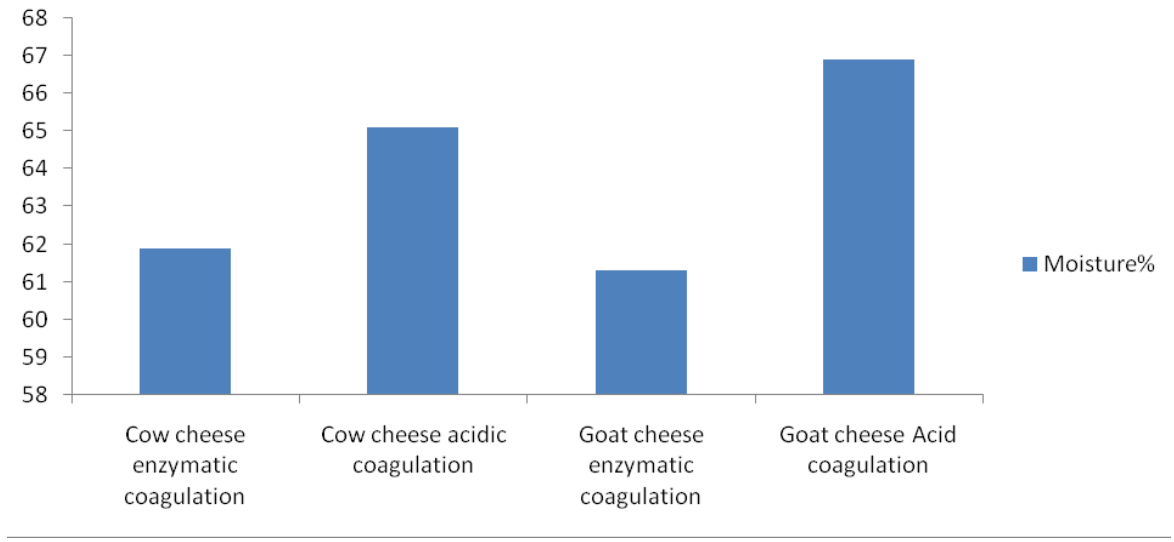

Figure 2 – Effect of co-aggulation type on final chees weight

CONCLUSION AND RECOMMENDATION

It is possible to conclude that lemon citrus juice can be utilized as an alternative coagulant in production of local Sudanese white cheese for high yield and faster curdling time which performed favourably with the commonly used rennet enzyme.

DECLARATIONS

Authors' contribution

Bhagiel BI performed data statistical analysis. Hamid EE conducted the lab work. Hamza AE contributed in writing, revising and printing manuscript, final draft.

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Competing interest

The authors declare that they have no competing interests.

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