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Original Article

Addition of transglutaminase enzyme in camel milk yoghurt to increase its sensorial aspects

Adição de enzima transglutaminase em iogurte de leite de camela para aumento de aspectos sensoriais

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Abstract

Camel milk was obtained from A-block UVAS Ravi Campus Pattoki. After pasteurization at 72 °C (15 sec) it was cooled to 42 °C, then glutathione treated transglutaminase enzyme was added with the concentration of 0.5 g/300 mL, 1 g/300 mL, 1.5 g/300 mL, 2 g/300 mL while control sample with the addition of 1.5 g/300 mL gelatin. Then inoculation of milk was done with standard cultures of Yoghurt *Lactobacillus delbrueckii subsp bulgaricus* and *Streptococcus thermophilus* at the rate of 2% for 3-4 hours at 42 °C. Samples were stored at 4 °C and were analyzed on 1st day and 28th day of storage. In our findings, there was slight increase in sensorial properties of all the samples. It was also observed that syneresis was reduced with the increase of enzyme quantity.

Keywords: camel milk, transglutaminase enzyme, yoghurt, sensory evaluation.

Resumo

O leite de camelo foi obtido do bloco B Uvas Ravi Campus Pattoki. Após a pasteurização a 72 °C (15 s), foi resfriado a 42 °C, posteriomente, à enzima transglutaminase tratada com glutationa foi adicionada com a concentração de 0,5 g/300 mL, 1 g/300 mL, 1,5 g/300 mL, 2 g/300 mL enquanto a amostra controle com a adição de 1,5 g/300 mL de gelatina. Em seguida, a inoculação do leite foi feita com culturas padrão de Iogurte *Lactobacillus delbrueckii subsp bulgaricus* e *Streptococcus thermophilus* com taxa de 2% durante 3-4 horas a 42 °C. As amostras foram armazenadas a 4 °C e analisadas no 1º dia e no 28º dia de armazenamento. Em nossos achados, houve leve aumento nas propriedades sensoriais de todas as amostras. Observou-se também que a sinérese foi reduzida com o aumento da quantidade da enzima.

Palavras-chave: leite de camelo, enzima transglutaminase, avaliação sensorial.

1. Introduction

Pakistan is a country endowed with diversified animal wealth, most of which is owned by nomadic sector scattering all over the country. There are about 18 million camels worldwide. In arid and semi-arid areas camels are the support system of millions of people for their energy needs. In dry and arid land areas, the camel has the capability to produce extra milk for an elongated time (an environment of lack of pasture, very high temperature and drought conditions cannot harm them) than in other domestic livestock species. Camel milk contains 8.9 to 14.3% solid-non-fat (SNF), 86.3 to 88.5% water, 2.5 to 4.5% protein, 0.35 to 0.90% ash, 2.9 to 5.8% lactose, and 2.9 to 3.5% fat.

Camel milk has greater total cholesterol and lower fatty acids containing less saturated fats, similar protein content, and lower lactose content, in comparison of cow's milk. In milk of camel greater contents of manganese, iron, zinc, phosphorus, sodium, potassium, vitamin C and ash are present as compared to cow's milk. Agrawal et al. (2007) also stated that Camel milk enhanced long-term glycemic control and decreased insulin dose in patients with type-1 diabetes. In African regions, a local fermented camel milk goods i.e. 'garris', is utilized for the treatment of leishmaniasis and the protozoal disease of the belly. Camel milk is more nutritious and has many therapeutic

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values than milk of any other animal. Studies showed that water contents of camel milk are increased in summer drought and milk is a good source of vitamin C and protein (Inayat and Farooq, 2005).

Worldwide from the total camel milk production, more than half the amount of milk is utilized for the formation of fermented dairy products, like Roub, yogurt, Gariss, Jibna-Beida and Mish. Meanwhile camel milk is deliberated one of the core constituents of the human diet in various parts of the world. It comprises of all vital nutrients as cow milk. Commonly, the consumption of camel milk is in sour or fresh form in Pakistan (Inayat et al., 2007). Camel milk has great biological values because of its greater contents of anti-microbial aspects such as lactoferrin, immunoglobulins and lysozyme. Fermented products like soft unripened cheese can be made from camel milk, but the difficulty in making cheese most probably refers to the technique which is being used (Inayat et al., 2003).

Water binding and elevation in increased viscosity are their mode of action in yoghurt. One of the utmost characteristics is texture which describes the value of yoghurt and upsets mouth-feel, appearance and overall adequacy. These variations might be because of differences in milk configuration, as well as changes in processing, storage conditions and incubation. Dairy ingredients and thickeners have been broadly added to the milk base to prevent these flaws, to provide an adequately firm texture and to lessen syneresis (Phillips and Williams, 2009). Among all Transglutaminase showed much more effective results in the textural properties of the yoghurt.

Starch utilized in yoghurt to enhance viscosity, increase mouth-feel, and avoid syneresis but it also causes ropiness to the yoghurt while Transglutaminase enzyme assimilates water and swell to several times their original size, causing amplified viscosity of the solution. Starch is one of the utmost commonly utilized thickening materials in the production of yoghurt because it is easy to use and very cost-effective as compared to hydrocolloids (Farnsworth et al., 2006). A hydrocolloid ingredient may act as a stabilizing agent and an emulsifying agent or both (Phillips & Williams, 2009).

Transglutaminase act as a stabilizer that deliberates long-term consistency on a suspension, perhaps by a mechanism including adsorption, but not essentially so. Hydrocolloids (such as carrageenan, xanthan, and carboxymethylcellulose) action of stability in oil-in-water (O/W) emulsions, are usually credited to the thickening, gelation of the aqueous phase (Ellis et al., 1985).

Transglutaminase levels of <1% is normally added to fermented dairy products. Though, health advantageous effects of Transglutaminase are attained with higher levels (3±5%). Enhancing gut efficiency and within food design may effect in cooperating the organoleptic properties of the product. However, because of the low viscosity, Transglutaminase have a greater potential to be easily assimilated into different foods (Bonisch et al., 2007a).

Milk of camel is also utilized for the cure of many diseases in various regions in addition with the use as food. Between the composition of cow and camel milk the main difference could lead performing differently during treating so it could affect the ultimate excellence of camel's milk dairy products. Traditional yogurt is mainly a fermented cow's milk product. Preparation of yogurt from camel milk is almost absent in Pakistan. Therefore, in this study we would like to assess the sensory quality before and after the storage of camel milk yogurt with the addition of transglutaminase enzyme in different percentages.

2. Materials and Methods

2.1. Sample collection

The Camel milk was collected under hygienic conditions from A block, the University of Veterinary and Animal Sciences (UVAS), Ravi Campus, Pattoki and then transported to the laboratory, department of Dairy Technology A block, University of Veterinary and Animal Sciences (UVAS), Pattoki for further processing. Glutathione treated transglutaminase enzyme samples (2 packs of 100 grams each from Ajinomoto Foods Europe SAS, France) were procured from the market. The experimental plan was laid out under Completely Randomized Design (CRD) (Table 1).

2.2. Processing of yoghurt

For the yoghurt production, raw and fresh buffalo and camel milk was used. Pasteurization was done to kill the pathogenic bacteria (for 15 sec at 72 °C) and then it was cooled to 42°C. After this milk was mixed with Glutathione treated Transglutaminase enzyme (T_1, T_2, T_3, and T_4). 1.5 g/300 mL Gelatin was added to manufacture the control sample at 42°C. Then inoculation of milk was done with standard cultures of yogurt, *Lactobacillus delbrueckii*, subsp *bulgaricus* and *Streptococcus thermophiles*, at the rate of 2% and incubation of culture was done for 3-4 hr at 42°C and then yoghurt was stored at 4 °C for 28 days

Table 1. Treatments at different level of TGase enzyme.

Sr. No.	Treatment	Different %age of GT treated TGase	Replicates
1	T_0 (Control)	Yogurt with (1.5 g/300 mL) with Gelatin	3 (A,B,C)
2	T_1	Yogurt with (0.5 g/300 mL) of GT+TGase	3 (A,B,C)
3	T_2	Yogurt with (1.0 g/300 mL) of GT+TGase	3 (A,B,C)
4	T_3	Yogurt with (1.5 g/300 mL) of GT+TGase	3 (A,B,C)
5	T_4	Yogurt with (2.0 g/300 mL) of GT+TGase	3 (A,B,C)

(Aprodu et al., 2011). The physico-chemical examination of both controlled and GT+TGase treated yoghurt were carried out by the standard methods as described by AOAC (2000).

2.3. Sensory assessment

The sensory assessment was carried out using color, acidity, appearance, taste, aroma, flavor, and complete adequacy (Figure 1) on a hedonic scale of nine points (Meilgaard et al., 2007). Prepared yogurt samples were used for sensory assessment by a panel of ten jury members. The panel included faculty members and postgraduate students of the department of Dairy Technology, University of Veterinary and Animal Sciences (UVAS). The sensory assessment was conducted within 12 h of yogurt preparation (Table 2). All the assessments were done at room temperature on the same day in the Department of Dairy Technology University of Veterinary and Animal Sciences (UVAS), Ravi Campus, Pattoki. The obtained data were converted to numerical values using a metric scale.

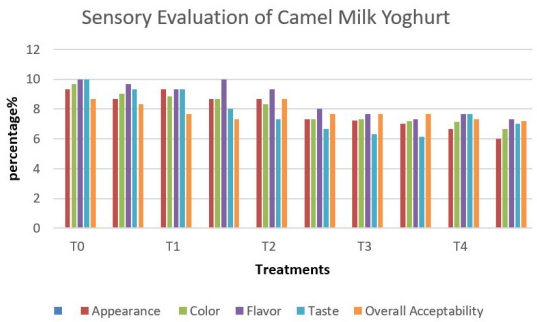

Figure 1. Sensory evaluation of Camel Milk Yoghurt prepared with Glutathione treated Transglutaminase enzyme.

2.4. Statistical analysis

Statistical data analysis was performed using SAS 9.1 Statistical Software. All data were checked for normal distribution using the Shapiro-Wilk's test and homogeneity of variances (Levene test). Moreover, all data were presented as the mean ± standard deviation (SD) of each group. Statistical significance of differences was determined by two-way analysis of variance (ANOVA). Post hoc Duncan multiple-range tests were applied to compare the means among treatments when there was a significant difference. Differences were considered significant at $P < 0.05$.

3. Results

The results of various physicochemical and sensory assessments of yogurt by the addition of Transglutaminase enzyme treatments are given below.

3.1. Sensory evaluation of glutathione treated transglutaminase enzyme based yoghurt

Yoghurt was evaluated for 28 days. An assessment was done by use of 9-point hedonic scale sensory assessment grade card which was assessed by panelists for score recording (Meilgaard et al., 2007). All the results showed significant differences among all the treatments (T0, T1, T2, T3, T4) including color, Flavor, appearance and overall acceptability while taste have slight effect of bitterness as we increase the concentration of the enzyme. Kumar and Mishra (2004). Taste decreased from 9.75-6.89% in T1, T2, T3, and T4.

4. Discussion

Yoghurt from camel milk was prepared and transglutaminase enzyme was added into it for the

Table 2. Sensory Evaluation of Camel Milk Yoghurt prepared with Glutathione treated Transglutaminase enzyme.

Treatments	Days	9- Point Hedonic Scale				
		Appearance	Color	Flavor	Taste	Overall Acceptability
T0	1st	8.440.67bc	8.37±0.42a	8.54±0.99B	8.43±0.27b	8.78±0.57cd
	28th	8.85±0.43c	8.01±0.73ab	8.25±0.43c	8.21±0.75ab	8.26±0.43c
T1	1st	8.26±0.71c	8.12±0.89d	9.21±0.82ab	9.75±0.34a	7.43±0.76d
	28th	9.83±0.34a	8.68±0.21c	9.74±0.91a	8.26±0.67bc	8.89±0.98c
T2	1st	9.71±0.26a	8.46±0.63bc	9.13±0.46a	7.76±0.81d	8.76±0.65d
	28th	8.59±0.54c	7.35±0.89e	9.23±0.36b	8.56±0.44e	7.35±0.23de
T3	1st	8.34±0.81bc	7.22±0.72de	7.68±0.62d	6.89±0.62de	7.12±0.56de
	28th	9.69±0.29ab	8.69±0.71bc	8.26±0.71bc	7.45±0.74d	7.56±0.34e
T4	1st	8.54±0.42c	8.32±0.57c	7.67±0.94e	7.87±0.41d	7.21±0.87f
	28th	9.23±0.84b	8.74±0.25c	9.33±0.68b	8.34±0.87c	8.34±0.61d

Values represent Weight (g) and are means ±S.E. of three replicates. Those not sharing a common alphabet within a respective column are significantly different from each other. Single factor analysis of variance at $P<0.05$. Whereas:T0 = Control (Yoghurt with 1.5 g/300 mL Gelatin), T1 = Yoghurt with (0.5 g/300 mL) of GT+TGase, T2 = Yoghurt with (1.0 g/300 mL) of GT+TGase, T3 = Yoghurt with (1.5 g/300 mL) of GT+TGase, T4 = Yoghurt with (2.0 g/300 mL) of GT+TGase.

evaluation of sensory properties. According to the previous studies, camel milk shows longer coagulation time as compared to all other types of milk (Farah and Bachmann, 1987; El Zubeir and Jabreel, 2008; Ahmed and El Zubeir, 2011). This was credited due to the modifications in the properties and size of casein particles (Farah and Atkins, 1992; Farah and Rüegg, 1989; Shuiep et al., 2013). Also, casein micelles are large in camel milk coagulum as compared to bovine milk coagulum (Farah and Rüegg, 1989). According to El Zubeir et al. (2012a), the chemical composition of yoghurt was different which was prepared from different percentage of camel milk and with two different starter cultures. Considerably low content of protein, total solids and fat have been observed in pure camel milk yoghurt as compared to all other types of milk. Camel milk yoghurt also has a more liquid texture. The addition of transglutaminase enzyme in the processing of yoghurt enhanced all these parameters, especially the content of total solids in yoghurt. Camel milk is not considered good for the processing of yoghurt as compared to other types of milk (El Zubeir et al., 2012b) due to the lower level of total solids and some major nutrients (Raynal-Ljutovac et al., 2008). In our study, the yoghurt samples show variation in the acidity during the first five hours which is similar to the observation of the El Zubeir and Ibrahium (2009). Attia et al. (2001) also described that the camel milk is less valuable for the lactic fermentation due to the low activity of the inoculated lactic starter in camel milk as compared to the bovine milk. Though, the titratable acidity is greater in all other types of milk yoghurt. This is because of the greater buffering capacity which is due to the increase protein content in milk (Li and Guo, 2006).

The significant differences between T1, T2, T3 and T4 yoghurt samples for the scores of colors is consistent with Stahl et al. (2006) study. Additionally, the color of the camel milk was liked by panelists, which can be due to the white color of fat present in camel milk. T2 and T3 sample yoghurt qualified to the lesser content of carotene in camel milk (Stahl et al., 2006). The best flavor score was gained in T1 and T2 sample yoghurts. The panelists observed that the texture of the yoghurt was not firm which was made from camel milk and this observation was similar to the findings of Hassan et al. (2007). Attia et al. (2001) also stated that the camel milk yoghurt has little dispersed small casein remains at the surface and a firm gel at the bottom of the container but don't have curd structure.

5. Conclusion

The results in the current investigation have clearly showed that addition of Transglutaminase enzyme have no adverse effect on all type of milks and its use can be beneficial for the replacement of the gelatin as gelatin usage have contradiction in Islamic countries related to its source of production. Thus, Transglutaminase enzyme will be an economic and easy available source for curdling, gel matrix strength enhancer or as a nutraceutical component of foods for human health.

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