



ISSN (E): 2277- 7695
ISSN (P): 2349-8242
NAAS Rating: 5.03
TPI 2019; 8(10): 150-152
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www.thepharmajournal.com
Received: 22-08-2019
Accepted: 24-09-2019

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Estimation of total antioxidant capacity of milk of Malabari goat and vechur cattle during different lactation

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Abstract

The present study was conducted to evaluate the changes in the antioxidant level of milk in different phases of lactation in Malabari goat and Vechur cattle, the native breeds of Kerala. Milk samples from 6 animals each of Malabari goats and Vechur cattles were collected separately at early, mid and late lactation period. Total antioxidant capacity was estimated by using ferric reducing antioxidant power. The study revealed that, irrespective of species the antioxidant capacity of milk at early lactation was higher than that at mid and late lactation. Throughout the lactation period the antioxidant capacity of Vechur milk was higher than that of goat milk ($p < 0.01$). The study further indicates that, irrespective of the species milk during the early lactation contained significantly higher content of antioxidants than the other two stages of lactation. High antioxidant capacity of milk of Vechur cattle is suggestive of high medicinal value which may be due to abundant presence of bioactive peptides.

Keywords: Antioxidant capacity, ferric reducing antioxidant power, Malabari goat, vechur cattle

Introduction

Fresh milk is an important food source of antioxidants [1]. Antioxidant compounds have the ability to scavenge free radicals and help to relieve stress [2]. Dairy products contribute significantly to the total intake of antioxidants. Milk contains essential nutrients necessary for normal growth and also rich in various physiologically functional components including proteins, vitamins (such as vitamins E and C), flavonoids, and carotenoids all of which contribute significantly to its antioxidant property. In milk, proteins (especially caseins) are the most important radical scavenger compounds [3]. Antioxidants contribute to health promotion and disease prevention [4]. Antioxidants gives protection against free radicals, but a sufficient concentration of antioxidants is necessary to balance the disruption caused by these radicals. Hence, milk with a high antioxidant capacity neutralize free radicals and highly protects consumers from exposure to oxidative stress. The oxidative stress is responsible for many fatal diseases such as carcinoma on different tissues and organs, cardiovascular diseases [5]. Free radicals induce oxidative damage to proteins, DNA and lipids in the organism especially on the cellular wall therefore causing cellular death [6] and it also affects the ability of the biological system to detoxify these reactive byproducts or to repair the damage occurred by their action.

The antioxidants help to neutralize the reactive components and thus prevent the formation of free radicals and inhibit lipid peroxidation, reducing the severity of oxidative stress. The dairy animals possess variable levels of antioxidant across different lactation period [7]. Compared to *Bos taurus*, Vechur milk and goat milk have better medicinal properties [8]. However no studies were conducted to compare the antioxidant property of Vechur milk with goat milk. Antioxidant status being a factor that contributes to medicinal property, this study was conducted to assess and compare the total antioxidant status of milk from Malabari goat and Vechur cattle the native breeds of Kerala at early, mid and late lactation period. The present study will also help to generate value of antioxidant level in these medicinally important breeds in different lactation phases which is not available.

Materials and methods

In the present study, total antioxidant capacity (TAC) of milk samples of Malabari goat and Vechur cattle were evaluated across early (5-15 days), mid (90-120 days) and late (>150 days) stages of lactation. A total of 36 milk samples from Malabari goats (n=6) and Vechur cows

(n=6) were used for the study. All of the milk samples were obtained early in the morning and stored at 4 °C till processing.

Determination of total antioxidant activity

The ferric reducing antioxidant power assay was used as a direct method for measuring the total antioxidant capacity. At low pH, ferric 2,4,6-tripyridyl-s-triazine [Fe (III)-TPTZ] complex gets reduced to ferrous 2,4,6-tripyridyl-s-triazine [Fe (II)-TPTZ] complex, which has an intense blue color, can be monitored by measuring the change in absorption at 593 nm. Working FRAP reagent was prepared by using 300 mM acetate buffer, pH 3.6 (3.1 g sodium acetate trihydrate, plus 16 mL glacial acid); 10 mM TPTZ (2,4,6-tris (2-pyridyl)-s-triazine), in 40 mM HCl; and 20 mM FeCl₃•6H₂O in the ratio of 10:1:1. Milk sample (100 µL) at room temperature was mixed with 3mL of FRAP reagent and kept in dark for 10 minutes. Then the sample was centrifuged at 8000 rpm for 5 minutes supernatant solution was taken. The absorbance of the supernatant solution was monitored at wavelength 593 nm with the spectrophotometer. Aqueous solutions of FeSO₄•7H₂O (100–3000 µM) was used as standard [9]. The data is shown as FRAP values (µM/mL Fe (II)).

Statistical analysis

Data were expressed as means ± standard error. For statistical comparisons between periods of lactation, the results were subjected to one way ANOVA using Statistical Package for the Social Sciences (SPSS). Significant differences ($p < 0.01$) between Malabari goat and Vechur cattle at each stage of lactation were analyzed by Independent t-test.

Results and discussion

In the FRAP method, the ability of sample to reduce ferric ion is used as a criteria for the measurement of total antioxidant capacity of milk. In both Vechur cattle and Malabari goat significantly higher level of total antioxidant capacity in the milk of early lactation when compared to mid and late lactation was noticed (Fig.1 and 2). However, there was no significant difference in the mean total antioxidant capacity values of mid and late lactation in both goat and Vechur cattle. Vechur milk showed a significantly higher total antioxidant capacity value compared to that of goat across

different stages of lactation ($p < 0.01$) (Fig.3).

The results of our study were similar to published data regarding total antioxidant capacity levels of bovine milk [9] and human milk [10] where they reported colostrum had higher total antioxidant capacity when compared to mature and transitional milk. The higher level of total antioxidant capacity in milk of early lactation might be to protect the young one which depends on milk as the sole source of nutrients. Young ones were potentially vulnerable to oxidative stress due to the inefficiency of their antioxidant defense system or increase in free radical production. Higher antioxidant level in early lactation was considered to be crucial to guard newborn's health against oxidative stress as they would go through many challenges to the oxygen-rich environment compared to the low-oxygen intrauterine environment [11]. In this study, total antioxidant capacity of milk during the course of lactation (early to late) showed a significant decrease which might be a natural result of decline in antioxidant storage of the adult animals.

Throughout lactation Vechur milk total antioxidant capacity was higher than total antioxidant capacity value of goat milk. While comparing the result with that of [9], Vechur milk has got higher total antioxidant capacity value than that of cross bred cattle and buffalo, suggestive of its better medicinal value. The structural and functional features of lactoferrin gene of Vechur cattle was studied by [12]. The gene was expressed for the production of recombinant bovine lactoferrin protein. Lactoferrin was assumed to have an important role in antioxidant capacity by preventing the oxidation of fatty acids. Lactoferrin was found to have antimicrobial, antiviral, antitumor, immunodefence and anti-inflammatory properties [13].

Increased total antioxidant activity of milk was due to the milk protein, which is an abundant source of bioactive peptides [14]. Vitamins in milk also contribute significantly to milk total antioxidant capacity. Vitamin E and carotenoids were located in the membranes of fat globules of milk where they prevent auto oxidation of milk fat. Vitamin C was an important antioxidant which was an electron donor in the conversion of tocopheroxyl radical back to antioxidant active vitamin E [15]. Many components of milk themselves had certain antioxidant capacity.

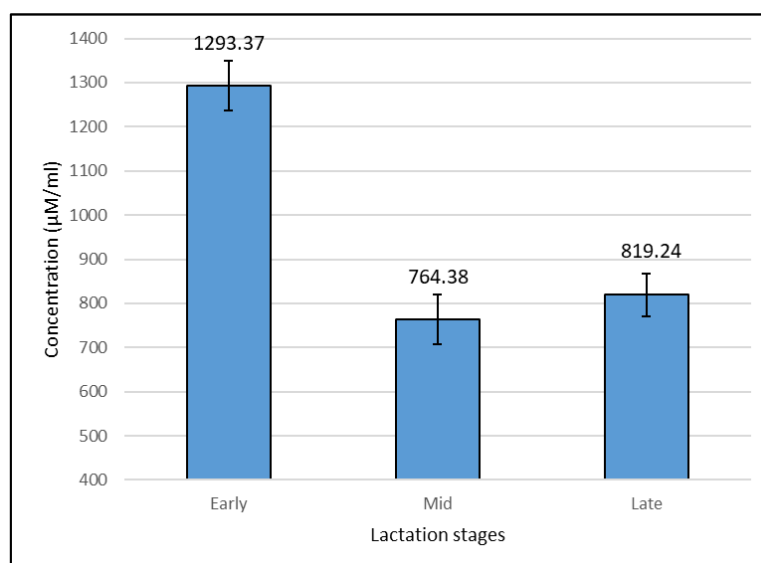


Fig 1: Total anti-oxidant capacity of milk of Malabari goat at different stages of lactation

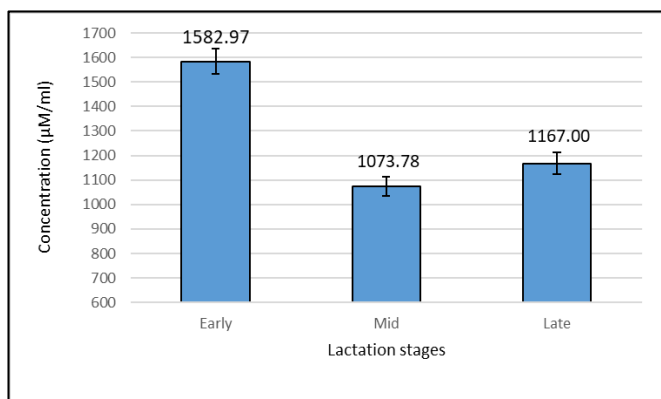


Fig 2: Total anti-oxidant capacity of milk of Vechur cattle at different lactation

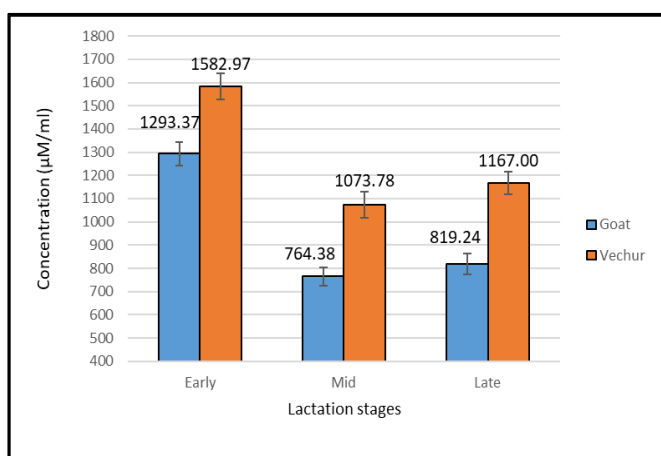


Fig 3: Comparison of total anti-oxidant capacity of milk of Malabari goat and Vechur cattle at different lactation

Conclusion

In this study, total antioxidant capacity of milk samples of Malabari goat and Vechur cattle throughout lactation were evaluated. It was demonstrated that the antioxidant levels varied during different stages of lactation and was found to be higher in early lactation stage when compared to later lactation stages. Also milk of Vechur cattle had high total antioxidant capacity compared to goat milk suggestive of high medicinal value of Vechur cattle milk.

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