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Antioxidant activity of Patanwadi breed sheep milk over cow milk—A preliminary study

TANMAY HAZRA[1]✉, SINDHAV ROHIT G[1], MITUL BUMBADIYA[1], RADHIKA GOVANI[1] and VIMAL M RAMANI[1]

Kamdhenu University, Amreli, Gujarat 365 601 India

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Clinical studies suggested that oxidative stress usually increases due to an imbalance between the production of reactive oxygen species (ROS) and anti-oxidant mediated defense systems in the human body (Alaa *et al.* 2020). Studies have also suggested that oxidative stress can be the cause of a wide spectrum of diseases like atherosclerosis, diabetes, cardiovascular disease, or other age-related diseases (Grazyna *et al*. 2017, Kalyan *et al*. 2021). Antioxidants can prevent oxidation through a variety of mechanisms, and as a result, they can prevent oxidation mediated variety of diseases (Santos-Sanchez *et al.* 2019, Alaa *et al*. 2020). The medical fraternity recommended that a diet enriched with natural antioxidants is able to prevent the formation of free radicals; therefore, anti-oxidant enriched foods are extremely helpful for the prevention of oxidative stress in the body (Kalyan *et al.* 2021). Milk is nearly a complete food, besides contributing the essential nutrients; it is a unique source of different bio-active components. essentially required for strong immune function in body. Milk is well furnished with anti-oxidant components both lipophilic (carotenoids, phospholipids, coenzyme Q10, etc.) and hydrophilic (caseins, whey proteins, vitamins, minerals, etc.) (Skrzypczak *et al*. 2019, Alaa *et al*. 2020) in nature. Studies suggested that various antioxidant mechanisms like free radical scavenging activities, pro-oxidant metal ion scavenging, etc. are different mechanisms for various antioxidant components of milk or dairy products (Grazyna *et al*. 2017). Therefore, milk or dairy products are very important sources of dietary antioxidants; hence, consumption of milk and dairy products can be beneficial to enhance the oxidative defense in the human body (Chen *et al*. 2003).

Among the different dairying species of milk produced in the world, bovine milk contributes the major portion (85%); followed by buffalo (11%), goat (2.3%), sheep (1.4%) and camel (0.2%) (Hazra *et al*. 2019). Although it has been observed that a wide population, throughout the world, is prone to cow milk protein allergy. Therefore, consumer's preferences are shifting towards small ruminants like goat or sheep milk. Sheep have been milked for thousands of years and were milked long before the first cow was milked (Ospanov and Toxanbayeva 2020). The calorific value of sheep milk was reported to be relatively higher than cow milk due to the presence of a higher concentration of proteins and lipids (Devi *et al*. 2018). Sheep milk proteins are an excellent source of bioactive peptides with the anti-oxidative, antimicrobial, antihypertensive, immunomodulatory and antithrombotic roles (Mahapatra *et al.* 2019). The world's commercial dairy sheep industry is concentrated in Europe and in the countries on or near the Mediterranean Sea (Shinde and Naqvi 2015).

In India, sheep make a valuable contribution to the livelihood of the economically weaker sections of the society. The dairy sheep industry is very small in India. A major portion of sheep milk is consumed as fluid milk. In India, very few un-organized dairies are processing sheep milk dairy products like ghee, desi butter or ice cream, etc. Till date very few studies exploited the compositional as well as functional attributes of indigenous breed of sheep milk (Gadekar *et al.* 2019). Hence, Mahapatra *et al.* (2019) reported that sheep milk has plenty of health benefits, but it is an untapped area by the Indian researchers. Patanwadi is the major sheep breed found in the Saurashtra region of Gujarat. However, the compositional as well as antioxidant properties of this Patanwadi breed sheep milk were never explored. Park *et al*. (2007) reported that wide variation of antioxidant activities exhibited in different species of milk. Hence, the present study evaluated and compared the antioxidant potential of Patanwadi breed sheep milk over cow milk.

The milk samples of cows (25) and Patanwadi sheep (25) were collected from the local village of Amreli. Thereafter each sample was transferred at 2–4°C to the laboratory of College of Dairy Science, Amreli.

Proximate composition of milk samples were analyzed using MilkoScan™ Mars instrument (Foss analytics, Hilleroed, Denmark). The acidity of milk was estimated by titration method using 0.1 N NaOH solution. The titratable

Present address: [1]College of Dairy Science, Kamdhenu University, Amreli, Gujarat. ✉Corresponding author email: tanmayhazra08@gmail.com

acidity was expressed as per cent lactic acid. The degree of lightness of milk samples were measured by Hunter colorimeter (color flex EZ colorimeter, Germany) and it was represented as L* value (represents lightness). The instrument was standardized with standard reference with scale 1 to 100 (higher score represents the higher intensity of lightness). The antioxidant properties of milk were evaluated by DPPH radical scavenging activity (Alaa *et al.* 2020), ABTS radical scavenging activity (Skrzypczak *et al.* 2019) and the metal (Fe^{2+}) chelating activity (Alaa *et al.* 2020). The results were expressed as (%) DPPH radical inhibition activity, (%) ABTS radical inhibition activity and (%) of metal ion (Fe^{2+}) chelating activity, respectively.

Data were presented as mean value±standard error. Statistical comparisons between all the samples were analyzed by Duncans Post-hoc or t-test at 95% confidence level ($p<0.05$) for testing the significance. Graphs were prepared using MS excel (2010).

The composition of sheep and cow milk is given in Table 1. The titratable acidity of sheep and cow milk were 0.20±0.01 and 0.13±0.00 (% lactic acid), respectively. The milk fat content of both the cow and sheep milk were 4.22±0.15 and 5.74±0.05%, respectively. Park *et al.* (2007) reported that due to the presence of higher fat content in sheep milk; this milk is richer source of energy (5932 kJ/kg) than cow milk. It was observed that sheep milk had a significantly ($p<0.05$) higher content of total solids (TS), lactose and protein than that cow milk (Table 1). This result is inconsistent with the results of Alaa *et al*. (2020) who reported that sheep milk had a higher concentration of fat, lactose and protein than cow milk. Our observation was also in line with few other researchers who earlier reported that sheep milk had a higher concentration of total solids (TS), fat, lactose and protein than that of cow milk (Park *et al.* 2007, Devi *et al.* 2018). The color of milk is usually white, however, due to presence of high concentration of beta carotene in cow milk, the lightness intensity of cow milk is little bit lesser than other species milk. It was observed that sheep milk had significantly ($p<0.05$) higher lightness than cow milk (Table 1). This can be ascribed to that the presence of higher concentration of beta carotene suppresses the intensity of lightness in cow milk.

Milk contains antioxidants that are both hydrophilic and lipophilic in nature and they have different mode of action; therefore, a single antioxidant assay is unable to evaluate antioxidant capability of milk solely (Alaa *et al.* 2020). Hence, in this present work, the antioxidant capacity of both cow and sheep milk were evaluated by DPPH, ABTS radical scavenging assay and metal chelating assay and the result is represented in Fig. 1. Sheep milk exhibited DPPH and ABTS radical inhibition (20.72±0.61 and 50.97±0.69%, respectively). The corresponding values for cow milk were 12.91±0.43 and 36.15±1.32%, respectively. It was clear that the DPPH and ABTS radical inhibition assay activity of sheep milk was significantly ($p<0.05$) higher than cow milk. The superior anti-oxidation capacity of sheep milk in comparison to cow milk can be a collective effect of

Table 1. Compositional and lightness variations between cow and sheep milk

Constituent	Cow milk	Sheep milk
Fat (%)	4.22±0.15[a]	5.74±0.05[b]
Protein (%)	3.43±0.05[a]	4.63±0.06[b]
Lactose (%)	4.30±0.06[a]	4.55±0.05[b]
Total Solids (TS) (%)	12.97±0.20[a]	15.49±0.09[b]
Acidity (% LA)	0.13±0.00[a]	0.20±0.01[b]
L value	81.61±0.29[a]	84.73±0.02[b]

Means with different superscript letters are significantly different (≤0.05) from each other. [a–b]Means with different superscript are significantly different ($p<0.05$) from each other in row.

different antioxidants present in milk. Scientific studies support that the concentration of protein, and other minor components directly affect the antioxidant properties of milk (Chauveau-Duriot *et al*. 2010, Alaa *et al.* 2020). In a similar study, Ertan *et al.* (2017) reported that milk lipid and lipid-soluble bio-active components directly affect the anti-oxidant properties of milk. Higher radical (DPPH and ABTS) inhibition activity of sheep milk can be ascribed to the protein and fat content that is higher in sheep milk than in cow milk (Table 1).

Iron is one of the principal pro-oxidant that accelerates oxidation reactions; therefore, iron chelation is one of the major mechanisms of anti-oxidation. Metal (Fe^{2+}) chelating activity of sheep milk was significantly ($p<0.05$) higher than cow milk (Fig. 1). Simos *et al*. (2011) reported that wide compositional variation directly affects the exhibited antioxidant activities of different species of milk (Simos *et al*. 2011). The three anti-oxidant assays exhibited in the present study showed (Fig. 1) that sheep milk had higher antioxidant activity over cow milk. Our study was in line with the earlier observation of Alaa *et al*. (2020), who

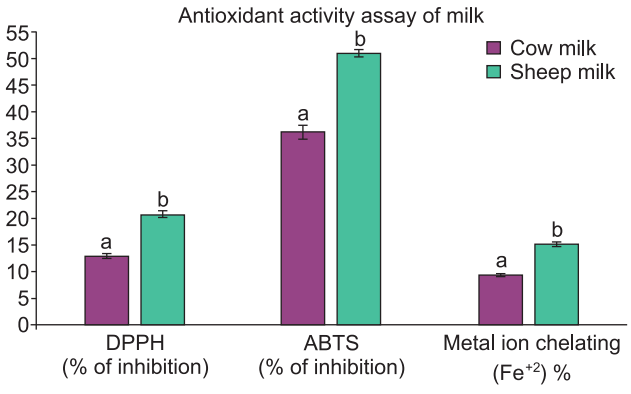

Fig. 1. Different antioxidant activity assay of sheep and cow milk. Means with different superscript letters are significantly different (≤0.05) from each other. [a–b]Means with different superscript are significantly different ($p<0.05$) from two different species milk. ABTS, 2,2-azinobis (3-ethylbenzothiazoline-6-sulfonic acid); DPPH, 2,2-diphenyl-1-picrylhydrazyl.

reported that sheep milk exhibited higher antioxidant activity than cow milk.

From this present scientific study, it can be concluded that sheep milk is a better candidate for dietary fat and protein over cow milk. Moreover, antioxidant activities in sheep milk established that this milk is a rich source of dietary antioxidants; therefore, it could be extremely helpful in decreasing oxidative stress-mediated diseases. As a result, there is an urgent demand for a couple of scientific investigations to develop Indian breed sheep milk-derived unique functional dairy products.

Present study confirmed that Patanwadi sheep milk had a higher concentration of fat and protein than cow milk. Higher antioxidant activity of Patanwadi sheep milk over cow milk, valorize unique features of this Indian breed's sheep milk quality. Moreover, these findings accelerate the Indian sheep breed dairying commercially; which strengthens up financial condition of the economically weaker section of our country. However, scientific researchers are required to develop process parameters for the production of sheep milk products commercially in Indian condition.

SUMMARY

Milk is an excellent source of dietary antioxidants, however, the antioxidants activity of different species of milk varies significantly. Sheep milk's compositional and functional properties are one of the untapped areas for Indian researchers. This present study evaluated and compared the antioxidant potential of Patanwadi breed sheep milk over cow milk. The protein, fat, and lactose content of Patanwadi breed sheep milk was significantly higher than cow's milk. Different anti-oxidant assays confirmed that the antioxidant activity of sheep milk was superior to cow milk. The present study supports that Patanwadi breed sheep could be a potential source of dietary antioxidants. Therefore, more scientific studies are required to develop sheep milk functional dairy products, and thereof optimized process parameters for sheep milk dairy products in Indian conditions.

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REFERENCES

Alaa A E F, Mohamed A, Hany E and Ahmed E. 2020. Antioxidant properties of milk: Effect of milk species. Milk fractions and heat treatments. *International Journal of Dairy Science* **15**: 1–9.

Chauveau-Duriot B, Doreau M, Noziere P and Graulet B. 2010. Simultaneous quantification of carotenoids, retinol, and tocopherols in forages, bovine plasma, and milk: Validation of a novel UPLC method. *Analytical and Bioanalytical Chemistry* **397**: 777–90.

Chen J, Lindmark-Mansson H, Gorton L and Akesson B. 2003. Antioxidant capacity of bovine milk as assayed by spectrophotometric and amperometric methods. *International Dairy Journal* **13**: 927–35.

Devi S, Gandhi K, Sao K, Arora S and Kapila S. 2018. Sheep milk: An upcoming functional food. *Indian Dairyman* **70**: 56–62.

Ertan K, Bayana D, Gokce O, Alatossava J T, Yilmaz Y and Gursoy O. 2017. Total antioxidant capacity and phenolic content of pasteurized and UHT-treated cow milk samples marketed in Turkey. *Akademik Gýda* **15**: 103–08.

Gadekar Y P, Shinde A K, Gauri J and Bhatt S. 2019. The quality of sheep milk and its products. *Indian Journal of Dairy Science* **72**: 119–21.

Grazyna C, Hanna C, Adam and Magdalena B M. 2017. Natural antioxidants in milk and dairy products. *International Journal of Dairy Technology* **70**: 165–78.

Hazra T, Sen D C and Saha P. 2019. Horse milk: A white ambrosia. *Indian Dairyman* **71**: 68–71.

Kalyan S, Meena S, Kapila S, Yadav R and Deshwal G K. 2021. *In vitro* assessment of antioxidative potential of goat milk, casein and its hydrolysates: Comparison of goat milk with bovine and buffalo milk. doi.org/10.21203/rs.3.rs-546200/v1 (last accessed on 15 June 2021)

Mohapatra A, Shinde A K and Singh R. 2019. Sheep milk: A pertinent functional food. *Small Ruminant Research* **181**: 6–11.

Ospanov A and Toxanbayeva B. 2020. Switching to sheep's milk industry: Problems and prospective. *EurAsian Journal of BioSciences* **14**: 1263–70.

Park Y W, Juarez M, Ramos M and Haenlein G F W. 2007. Physico-chemical characteristics of goat and sheep milk. *Small Ruminant Research* **68**: 88–113.

Santos-Sanchez N F, Salas-Coronado R, Villanueva-Canongo C and Hernnndez-Carlos B. 2019. Antioxidant compounds and their antioxidant mechanism, pp. 23–50. *Antioxidants*. (Ed.) Shalaby E. Intech Open, London, UK.

Shinde A K and Naqvi S M K. 2015. Prospects of dairy sheep farming in India: An overview. *Indian Journal of Small Ruminant* **21**: 180–95.

Simos Y, Metsios A, Verginadis I, D'Alessandro A G, Loiudice P, Jirillo E and Karkabounas S. 2011. Antioxidant and anti-platelet properties of milk from goat, donkey and cow: An *in vitro*, *ex vivo* and *in vivo* study. *International Dairy Journal* **21**: 901–06.

Skrzypczak K, Gustaw W, Kononiuk A, So³owiej B and Wasko A. 2019. Estimation of the antioxidant properties of milk protein preparations hydrolyzed by *Lactobacillus helveticus* T80, T105 and B734. *Czech Journal of Food Sciences* **37**: 260–67.