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Gross compositional analysis of milk from Vechur and Kasargod Dwarf breeds of cattle

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Abstract
Milk is a well-balanced food in the human diet and its premium nutritional quality is related to its composition. Vechur and Kasargod Dwarf cattle are indigenous to the state of Kerala, India. Vechur cattle is registered as an indigenous breed as per National Bureau of Animal Genetics Resources (NBAGR) whereas Kasargod Dwarf is labelled as 'non-descript'. Limited information is available on the compositional characteristics of milk from these cattle. In the present study, pooled milk samples of Vechur and Kasargod Dwarf cattle were collected for analysis of gross composition.

Keywords: Vechur, Kasargod dwarf, gross-composition

1. Introduction
Milk is defined as the normal mammary secretion, excluding colostrum, obtained by the complete milking of one or more healthy animals, which contains a prescribed percent of fat and SNF. The chemical composition of milk is dynamic in nature and can be influenced by several factors such as animal species and genetics, environmental conditions, lactation stage, and animal nutritional status (Kalac and Samkova, 2010) [4]. Many researchers have evaluated the breed-wise differences in bovine milk composition, though indigenous breeds in the southern part of India are not much studied. The fat, protein, lactose, ash, and total solids content constitutes the gross composition of milk. According to Kapadiya *et al.* (2016) [5], cow milk is composed of 4.88% fat, 8.54% SNF, 13.79% TS, 3.49% protein, 4.76% lactose and 0.76% ash. The various genetic and non-genetic factors affecting bovine milk composition have been assessed by several workers (Nickerson, 1995; Krovvidi *et al.*, 2013) [10, 6]. Breed, feed, season, geography, and herd health are the most important aspects in the processing of milk and milk products. The variation in gross composition of milk can be related to animal breeds, season/weather conditions during milking, locality, stages of lactation, age and size of cow, environmental and dietary composition (Smit *et al.*, 2000) [13]. Among these, cattle breed is known to have a striking effect on the characteristics of milk produced.

2. Materials and Methods
2.1 Collection of milk samples
Fresh, pooled milk samples from Vechur and Kasargod Dwarf breeds of cattle were collected from Livestock farm, Kerala Veterinary and Animal Sciences University, Mannuthy. Raw milk samples were collected at the time of milking in clean containers. The samples were immediately transported to the laboratory and stored under refrigeration condition till taken for various analyses. Three trials of chemical analyses were conducted every month during the period of study from December 2020 to November 2021

2.2 Compositional analysis
2.2.1 Total Solids: The total solid content of the samples was determined using the procedure described in Handbook of Food Analysis, Part XI, Dairy Products, SP: 18 (Part XI)-1981, Bureau of Indian Standards. A known quantity of milk was dried on a boiling water bath. Subsequently, sample was dried in a hot air oven at 102±2 °C and from the weight of the residue, the total solids content in milk is determined.

2.2.2 Fat: Fat content was determined by Gerber method. Milk sample (10.75 ml) was mixed with Gerber sulfuric acid (10 ml) and amyl alcohol (1 ml) in butyrometer and closed with rubber cork. The contents were mixed and placed in a water bath at 65 °C.

Sample was centrifuged in Gerber centrifuge machine for 5 min at 1000 rpm. The fat content was determined by adjusing the stopper until the lower end of the fat column is on the zero point.

2.2.3 Solids Not Fat: SNF content in milk was estimated by Zeal lactometer using the following formula.

$$SNF\% = CLR/4 + 0.2F + 0.50$$

Where,
CLR is the corrected lactometer reading (at 29 °C), F is the fat percent

2.2.4 Protein: Total protein content of the milk was determined by Kjeldhal method. The percent of true protein was estimated by multiplying the nitrogen content by 6.38. The sample (0.2-1 g) was digested using Kjeldhal digester in the presence of catalyst mixture (2 g) where sulfuric acid (15-20 ml) was used as an oxidizing agent. Then 5 ml portion from the diluted sample was distilled with NaOH (40%), using Kjeldhal distillation unit, where steam was distilled over 4% boric acid (20 ml) containing an indicator for 3 minutes. The ammonia trapped in boric acid was determined by titrating with 0.1N HCl. The nitrogen percent was calculated using following formula

$$N\% = \frac{14\ (\text{blank volume} - \text{titre volume}) \times \text{normality of acid}}{\text{Weight of the sample} \times 1000} \times 100$$

2.2.5 Lactose: Lactose content was estimated by Lane Eynon's method as described in Handbook of Food Analysis, Part XI, Dairy Products, SP: 18 (Part XI)-1981, Bureau of Indian Standards. Twenty five ml of milk sample was diluted with distilled water to about 125 ml and treated with 3.75 ml 10% glacial acetic acid. The contents were boiled, mixed and then cooled. The volume was made upto 250 ml using distilled water. The mixture was filtered through Whatman filter No.42. The filtrate so obtained was titrated against 10 ml mixture of Fehlings reagent (Five ml each of Fehlings A and Fehlings B solution) with constant heating to boiling temperature for 1 min. The end point of the titration can be detected using methylene blue as an indicator, which on reduction, changes colour of the solution from blue to colourless. At this stage complete precipitation of Cu2O occurs in the form of brick-red precipitation. The Fehlings solution was standardized using 0.5% standard lactose solution and the lactose content is calculated.
Ash: Ash content was determined by Gravimetric method, as described in the Handbook of Food Analysis, Part XI, Dairy Products, SP: 18 (Part XI)-1981, Bureau of Indian Standards. Five ml of milk, weighed in a silica crucible, was initially evaporated in a boiling water bath. After removal of water, the crucibles were placed in a muffle furnace and dry ashed at 550 °C till formation of carbon free white residue.

2.3 Statistical Analysis
The results obtained were expressed as mean ± standard deviation of three replicate analysis. The chemical properties were analyzed every month and the mean tested for significance using One-way ANOVA with Duncan test at 5% level of significance. Statistical testing was done using SPSS (Statistical Packages for Software Solutions) software, Version 23 designed by IBM Company, USA

3. Results
The gross composition of milk from Vechur and Kasargod dwarf breeds of cattle was estimated and the results are explained below.

3.1 Total Solids: The total solid content in the milk samples of Vechur and Kasargod dwarf breeds of cattle was estimated using gravimetric method and the results are tabulated below (Table 1). Total solid content of Vechur cow milk ranged from 13.72 to 16.62% with an average of 15.09%. The highest total solids percent was observed in the month of June (16.62%) and minimum value was observed in December (13.72%). The total solid concentration increased from 13.72% in the month of December to 16.62% in June, then reduced gradually till November. There is statistically significant difference in the total solids content of Vechur cow milk with the period of study. Total solid content of Kasargod Dwarf milk ranged from 13.73 to 15.03% with an average of 14.46%. The highest total solids percent was observed in the month of October (16.62%) and minimum value was observed in June (13.72%). Season had no significant effect on total solids content of Kasargod Dwarf cow milk.

Table 1: Total solid content (%) in Vechur and Kasargod Dwarf cow milk

Period	Vechur Cow	Kasargod Dwarf Cow
Dec 2020	13.72 ± 0.33^{a}	14.27 ± 0.47^{a}
Jan 2021	14.34 ± 0.24^{abc}	13.90 ± 0.59^{a}
Feb 2021	14.18 ± 1.23^{ab}	14.17 ± 1.35^{a}
Mar 2021	15.15 ± 0.17^{bcd}	14.78 ± 0.72^{a}
Apr 2021	15.23 ± 0.42^{bcde}	14.98 ± 0.57^{a}
May 2021	15.52 ± 0.34^{bcde}	14.36 ± 0.01^{a}
Jun 2021	16.62 ± 0.34^{e}	13.73 ± 0.40^{a}
Jul 2021	15.89 ± 0.41^{de}	14.48 ± 0.52^{a}
Aug 2021	15.64 ± 0.49^{cde}	14.96 ± 0.35^{a}
Sept 2021	15.00 ± 0.50^{abcd}	14.18 ± 1.27^{a}
Oct 2021	14.97 ± 0.73^{abcd}	15.03 ± 0.78^{a}
Nov 2021	14.80 ± 1.04^{abcd}	14.68 ± 0.39^{a}
Mean ± SD	15.09±0.95	14.46±0.76
CV%	5.02	2.85

Figures are mean ± standard deviation of triplicate analyses. Means with different superscripts within a column differ significantly ($p<0.05$)

3.2 Fat: Fat is the most valuable among the milk components, and plays a crucial role in the pricing of milk. Moreover, it is attributed with the flavour, mouthfeel and textural characteristics of milk and milk products. Fat content of Vechur cow milk ranged from 3.87 to 5.30% with an average of 4.45%. The fat content was shown to differ significantly with the month ($p<0.05$). The highest fat percent was observed in the month of June (5.3%) and minimum value was observed in November (3.87%). The fat percent is observed to follow an increasing trend from December to June attaining a peak and then decreasing constantly. Fat content of Kasargod Dwarf cow milk ranged from 3.08 to 4.68% with an average of 3.92%. The fat content showed significant difference with the month ($p<0.05$). The highest fat percent was noticed in the month of March (4.68%) and minimum value was observed in June (3.08%). The fat

percent is not observed to follow any constant pattern of increase or decrease.

Table 2: Fat content (%) in Vechur and Kasargod Dwarf cow milk

Period	Vechur Cow	Kasargod Dwarf Cow
Dec 2020	4.00±0.00^{ab}	4.33±0.85^{bcd}
Jan 2021	4.00±0.14^{ab}	4.03±0.51^{abcd}
Feb 2021	4.18±0.17^{ab}	3.73±0.53^{abcd}
Mar 2021	4.53±0.25^{abc}	4.68±0.62^{d}
Apr 2021	4.50±0.20^{abc}	3.45±0.21^{ab}
May 2021	4.84±0.70^{bc}	3.20±0.21^{a}
Jun 2021	5.30±0.17^{c}	3.08±0.30^{a}
Jul 2021	5.20±0.71^{c}	3.70±0.10^{abc}
Aug 2021	4.67±0.25^{abc}	3.83±0.23^{abcd}
Sept 2021	4.03±0.58^{ab}	3.83±0.72^{abcd}
Oct 2021	4.28±0.68^{ab}	4.53±0.59^{cd}
Nov 2021	3.875±0.63^{a}	4.67±0.15^{cd}
Mean ± SD	4.45±0.59	3.92±0.68
CV%	10.29	13.41

Figures are mean ± standard deviation of triplicate analyses. Means with different superscripts within a column differ significantly (p<0.05).

3.3 Solids not Fat: The SNF content in the Vechur cow milk ranged between 8.48-9.56% with an average value of 9.05%. The highest SNF content (9.56%) was observed in the month of September and lowest (8.48%) in April. There is statistically significant difference in the SNF content of Vechur cow milk with varying months of analysis. The SNF content in the Kasargod Dwarf cow milk ranged between 8.75- 9.34% with an average value of 9%. The highest SNF content (9.34%) was observed in the month of August and lowest (8.75%) in March. Season had statistically no significant effect on the SNF content of Kasargod Dwarf milk.

Table 3: SNF content (%) in Vechur and Kasargod Dwarf cow milk

Period	Vechur Cow	Kasargod Dwarf Cow
Dec 2020	8.93±0.18 ^{abc}	8.87±0.17 ^{ab}
Jan 2021	8.96±0.10 ^{abc}	9.01±0.45 ^{ab}
Feb 2021	9.02±0.05 ^{bcd}	9.08±0.19 ^{ab}
Mar 2021	9.20±0.04 ^{cd}	8.75±0.21 ^{a}
Apr 2021	8.48±0.62^{a}	9.13±0.13 ^{ab}
May 2021	8.66±±0.10 ^{ab}	8.76±0.22 ^{a}
Jun 2021	8.64±0.09 ^{ab}	9.12±0.20 ^{ab}
Jul 2021	9.07±0.37 ^{bcd}	8.98±0.23 ^{ab}
Aug 2021	9.52±0.12 ^{d}	9.34±0.14 ^{b}
Sept 2021	9.56±0.01 ^{d}	9.18±0.14 ^{ab}
Oct 2021	9.29±0.23 ^{cd}	8.91±0.64 ^{ab}
Nov 2021	9.28±0.31 ^{cd}	8.93±0.03 ^{ab}
Mean ± SD	9.05± 0.39	9.00± 0.28
CV%	3.60	1.87

Figures are mean ± standard deviation of triplicate analyses. Means bearing at least one common superscript do not differ significantly (p<0.05).

3.4 Protein: Protein content of Vechur cow milk ranged from 3.18 to 4.09% with a mean of 3.63%. The highest protein concentration was observed in the month of March (4.09%) and minimum value was observed in November (3.18%). Protein content of Kasargod Dwarf cow milk ranged from 3.49 to 4.25% with an average of 3.87. The peak protein percent was noticed in the month of September (4.25%) and minimum value was observed in April (3.49%). The protein percent was not observed to follow any constant pattern of increase or decrease.

Table 4: Protein content (%) in Vechur and Kasargod Dwarf cow milk

Period	Vechur Cow	Kasargod Dwarf Cow
Dec 2020	3.39±0.10^{ab}	3.53±0.02 ^{a}
Jan 2021	3.54±0.04 ^{abc}	3.68±0.03 ^{ab}
Feb 2021	3.84±0.62 ^{bcd}	4.01±0.21 ^{de}
Mar 2021	4.09±0.34 ^{d}	3.98±0.06 ^{cde}
Apr 2021	3.28±0.01 ^{a}	3.49±0.08 ^{a}
May 2021	3.42±0.13 ^{ab}	3.81±0.15 ^{bcd}
Jun 2021	3.91±0.02 ^{bcd}	3.63±0.02 ^{ab}
Jul 2021	4.02±0.02 ^{cd}	3.77±0.03 ^{bc}
Aug 2021	3.62±0.16 ^{abcd}	4.11±0.17 ^{ef}
Sept 2021	4.06±0.11 ^{cd}	4.25±0.04 ^{f}
Oct 2021	3.2±0.03 ^{a}	4.06±0.03 ^{ef}
Nov 2021	3.18±0.01 ^{a}	4.14±0.01 ^{ef}
Mean ± SD	3.63±0.37	3.87±0.26
CV%	9.01	6.33

Figures are mean ± standard deviation of triplicate analyses. Means bearing at least one common superscript do not differ significantly (p<0.05).

3.5 Lactose: The lactose content was analysed using Lane Eynon method. The mean values are tabulated in Table 5. Lactose content of Vechur cow milk ranged from 4.72 to 5.25% with an average of 5.04%. The lactose content did not show any significant difference with the month. The highest lactose percent was observed in the month of January (5.25%) and minimum value was observed in July (4.72%). Lactose content of Kasargod Dwarf cow milk ranged from 4.87 to 5.33% with an average of 5.05%. The lactose content was observed to differ significantly with the month (p<0.05). The highest lactose percent was noticed in the month of December (5.33%) and minimum value was observed in March (4.87%). The lactose concentration gradually decreased from a peak value in the month of December to the minimum value in March, and further increased till November.

Table 5: Lactose content (%) in Vechur and Kasargod Dwarf cow milk

Period	Vechur cow	Kasargod Dwarf cow
Dec 2020	5.18±0.16 ^{b}	5.33±0.16 ^{c}
Jan 2021	5.25±0.22 ^{b}	5.13±0.22 ^{bc}
Feb 2021	4.95±0.02^{ab}	4.92±0.08^{ab}
Mar 2021	5.00±0.20^{ab}	4.87±0.09 ^{a}
Apr 2021	5.05±0.10^{b}	4.96±0.04 ^{ab}
May 2021	5.11±0.04 ^{b}	4.95±0.03 ^{ab}
Jun 2021	4.94±0.06^{ab}	4.95±0.13 ^{ab}
Jul 2021	4.72±0.01^{a}	5.07±0.00 ^{ab}
Aug 2021	5.06±0.02 ^{b}	5.13±0.05 ^{bc}
Sept 2021	5.09±0.02 ^{b}	5.09±0.04 ^{abc}
Oct 2021	5.06±0.01 ^{b}	5.09±0.03 ^{abc}
Nov 2021	5.12±0.31 ^{b}	5.13±0.01 ^{bc}
Mean ± SD	5.04±0.16	5.05±0.16
CV%	2.52	2.42

Figures are mean ± standard deviation of triplicate analyses. Means bearing at least one common superscript do not differ significantly (p<0.05).

3.6 Ash: Ash content of Vechur cow milk ranged from 0.61 to 0.87% with an average of 0.78%. The highest ash percent was noticed in the month of September (0.87%) and minimum value was observed in January (0.61%). The ash concentration was significantly lower in the initial months of

study (December and January), which then increased and remained almost constant till August. The ash content was observed to differ significantly with varying period of study. Ash content of Kasargod Dwarf cow milk ranged from 0.70 to 0.85% with an average of 0.77%. There was statistically significant difference in the ash content of milk from Kasargod Dwarf during the period of study. The highest ash percent was noticed in the month of November (0.85%) and minimum value was observed in July (0.70%).

Table 6: Ash content (%) in Vechur and Kasargod Dwarf cow milk

Period	Vechur cow	Kasargod Dwarf cow
Dec 2020	0.62±0.01^{a}	0.73±0.01 ^{ab}
Jan 2021	0.61±0.10 ^{a}	0.74±0.05 ^{abc}
Feb 2021	0.80±0.01 ^{b}	0.79±0.04 ^{abcd}
Mar 2021	0.78±0.01 ^{b}	0.75±0.04 ^{abc}
Apr 2021	0.78±0.01 ^{b}	0.73±0.03 ^{ab}
May 2021	0.80±0.01 ^{b}	0.78±0.01 ^{abcd}
Jun 2021	0.83±0.02 ^{b}	0.81±0.10 ^{bcd}
Jul 2021	0.79±0.03 ^{b}	0.70±0.03 ^{a}
Aug 2021	0.80±0.01 ^{b}	0.73±0.06 ^{ab}
Sept 2021	0.88±0.01 ^{b}	0.82±0.01 ^{bcd}
Oct 2021	0.84±0.06 ^{b}	0.83±0.02 ^{cd}
Nov 2021	0.83±0.09 ^{b}	0.85±0.05 ^{d}
Mean ± SD	0.78±0.08	0.77±0.06
CV%	10.23	5.98

Figures are mean ± standard deviation of triplicate analyses. Means bearing at least one common superscript do not differ significantly (p<0.05).

4. Discussion

Bindya and Gayathri (2015) [1] compared the gross composition of milk from Vechur, Sahiwal, Kankrej and crossbred HF cattle and reported a TS content 12.69+/-0.71 for Vechur cow milk. Thirupathi and Iype (1998) [14] found the average percent of total solids in morning and evening milk from Vechur cow to be 14.79+/-0.13 and 15.53+/-0.12, respectively. The results by Mahmood and Usman (2010) [8] illustrated that the concentration of TS in cow milk, collected from various regions of Gujarat and Pakistan was 11.23-14.26% with an average of 12.94+/- 0.97%. Imran *et al.*, (2008) [3] reported mean TS content as 13.5% in cow milk collected from different areas of Pakistan. The results obtained for TS content in Vechur and Kasargod Dwarf milks is similar to the findings of Thirupathi and Iype (1998) [14], whereas slightly higher than the values reported by Bindya and Gayathri (1995) [1]; Imran *et al.* (2008) [3]; Naicy (2009) [9]; and Mahmood and Usman (2010) [8]. The TS content of milk from both the cattle varieties were found to be higher than those available in the literature for other indigeneous and crossbred cattle in southern part of India.

According to Thirupathy and Iype (1998) [14], the average percent of milk fat in Vechur milk samples was 5.95+/-0.12 for morning milk and 6.62+/-0.13 for evening milk. Bindya and Gayathri (2015) [1] reported that Vechur milk had a mean fat% of 3.61+/-0.27. The results from the study conducted by Mahmood and Usman (2010) [8] illustrated that the range of fat content in the cow milk collected from Gujarat and Pakistan was 3.44-4.96% with a mean value of 4±0.43%. As per the study conducted by Naicy (2009) [9], the average fat percent of milk from crossbred cattle, maintained at University Livestock Farm, Kerala Agricultural University, Mannuthy and Cattle Breeding Farm, Thumburmuzhi, was 3.738±0.0788%. The data obtained from present study for mean fat percent of milk from the cattle was lower than that noted by Thirupathy and Iype (1998) [14], but slightly higher than that observed by Bindya and Gayathri (2015) [1]. Throughout the study, Vechur milk was found to have slightly higher fat percent than Kasargod Dwarf milk. Seasonal difference was observed in the fat content of Vechur and Kasargod Dwarf milk, which is in accordance with the studies conducted by various workers on the effect of season in milk composition. Observation of a higher fat percent in the Vechur milk during the rainy season, is similar to the findings of Patwardhen *et al.* (1986) [11]. This may be accounted to the availability of higher quantities of roughage in the rainy season. On contrary, Kasargod Dwarf milk exhibited lower fat concentration in the rainy season. This may be attributed to the combined effect of factors like stage of lactation, individuality, health and age of animal.

The results obtained for SNF content in the present study is in close agreement with the study conducted by Bindya and Gayathri (2015) [1], who reported a mean SNF% of 9.09±0.07 in 95 samples of morning milk from Vechur cows of early second lactation. Thirupathy and Iype (1998) [14] observed that the least square mean for SNF content in milk samples collected from Vechur cows for 1 to 44 weeks was 8.89±0.13%. The SNF content in the Vechur and Kasargod Dwarf milk observed in the present study is slightly higher than that reported by Thirupathy and Iype (1998) [14]. Naicy (2009) [9] compared the milk composition traits of ten sire families of crossbred cattle of Kerala and noted a mean SNF percent of 8.456±0.0053, which is lower than that observed in the present study. This could be attributed to better management approaches and the herd's genetic potential.

The Kasargod Dwarf milk exhibited a higher mean protein content compared to Vechur cow milk and this observation is reflected in various other physico-chemical properties analysed, as well. The mean protein content of Vechur and Kasargod Dwarf milk are higher than the values reported by Bindya and Gayathri (2015) [1] for Vechur cow milk. As suggested by Schmidt *et al.* (1996) [12], the higher titrable acidity observed in the milk may be an indication of its increased protein content.

Bindya and Gayathri (2015) [1] compared the chemical composition of milk from indigeneous and crossbred cattle under subtropical climates and reported an average lactose content of 4.99±0.04% in Vechur cow milk. Lactose was not observed to undergo seasonal fluctuation. Hashmi (1975) [2] found that indigenous cow milk had a higher lactose concentration. The present data on lactose content in Vechur and Kasargod Dwarf milk is in close agreement with the above- mentioned findings. Although the effect of season is seen to have a contradictory result, the range and mean lactose content of Kasargod Dwarf milk was quite similar to that of Vechur milk.

As such, there is no data available for ash content of Vechur and Kasargod Dwarf milk. The available literature mostly pertains to the ash content of raw milk obtained from different regions in and around India. Mahmood and Usman (2010) [8] compared the composition of milk from different species in Gujarat, Pakistan and found an average ash percent of 0.6% for cow milk. Imran *et al.* (2008) [3] reported average ash content as 0.64% in cow milk collected from various areas of Pakistan. Lingathurai *et al.* (2009) [7] studied the chemical composition of cow milk obtained from different locations in Madurai and observed an average ash content of 0.80%.

Hashmi (1975) [2] reported ash content of 0.81% for the milk from desi cow Deoni. The ash content of milk from Malnad Gidda cow was observed to be 0.71% by Yuvaraja *et al.* (2015) [15]. The data pertaining to the ash content of milk samples in the present study were in line with those reported for cow milk by Hashmi (1975) [2], Lingathurai *et al.* (2009) [7] and Yuvaraja *et al.* (2015) [15], whereas it is higher than that observed by Imran *et al.* (2008) [3] and Mahmood and Usman (2010) [8].

5. Conclusion

The role of native cattle varieties in the dairy sector is well evident from the Livestock census reports (2019), which unveils that indigeneous/non-descript cattles accounts for 73.8% of total cattle population in India. The milk from indigeneous cattle breeds is claimed to possess nutritional and therapeutic properties and this has urged the dairy stakeholders and Government organizations to conserve and develop native cattle breeds, that were almost at the verge of extinction. The present study attempts to estimate the gross chemical composition of Vechur and Kasargod Dwarf breeds of cattle that are native to the state of Kerala, India. The gross compositional analysis of milk revealed that Vechur milk had higher concentration of total solids, fat and ash compared Kasargod Dwarf milk, but protein content was more in the latter. Food safety authorities are in constant process of amending the standards and the results obtained in the compositional parameters of milk from native cattle can provide a scientific database for building standards.

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7. References

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