RESEARCH ARTICLE

Association of AM-PM Milk Records with Daily Milk Traits in Gir Cows

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Abstract

Present study evaluated association of morning (AM) and afternoon (PM) milk records with 24 h daily milk traits (milk yields, composition and somatic cells) in Gir cows (n=50). Milk yield, composition and somatic cell count (SCC) were recorded at fortnightly interval. The milk yield did not differ at AM and PM milking sessions, while fat corrected milk yield was significantly (p=0.021) higher at PM than AM milking session. The AM milk had significantly lower fat content (p=0.034), but higher protein (p=0.032), lactose (p=0.011), ash (p=0.004) and solid-not-fat (p=0.009) content than PM milk. Milk total solids content and SCC of AM and PM milk did not differ significantly. All the AM-PM milk traits showed positive and significant (p<0.001) correlation with daily milk traits. Linear regression equations for prediction of daily milk traits from AM-PM milk records revealed that values of coefficient of determination (R^2) ranged from 0.51 to 0.77 and from 0.48 to 0.86 at AM and PM milking sessions, respectively. Among different milk traits, PM milk yield was more accurate (86%) to predict daily yield. Taken together it is concluded that daily milk traits of Gir cows particularly milk yield can be predicted from afternoon yield with higher accuracy.

Keywords: AM-PM milking, Gir cows, Milk composition, Milk yield, SCC.

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Introduction

Recording of milk traits like yield, composition and somatic cell counts (SCC) etc. is most important for identification of better performing animals and practical management of dairy herds. Accurate recording of milk yields and compositions is also essential for genetic evaluation of breeding sires under progeny testing programmes (Berry *et al.,* 2006; Forsback *et al.,* 2010; McParland *et al.,* 2019). In organized dairy herds the milk yield is recorded at each milking and 24-h total yield is calculated by summing up the yields during 24 h, but the milk composition is estimated at monthly interval either sampling the milk at morning (AM) or afternoon (PM) milking sessions. Under field conditions for progeny testing programmes milk recording is done once a month and on the same day milk composition is estimated from morning milk samples (Anonymous, 2012). The recording of dairy cows' daily yield and estimation of milk composition is a laborious task not only under field condition due to lack of infrastructure facilities, skilled man powers, logistic facilities etc. but also in well established organised dairy farms during busy milking sessions. Therefore, to reduce the cost of recording of the milk traits scientists have suggested once recording of milk traits either at AM or PM milking sessions followed by calculation of 24-h daily yield and composition by using adjustment factors (Quist *et al.,* 2008; McParland *et al.,* 2019). The evaluation of association of milk yield and composition either in AM or PM milk with 24-h daily yield and composition will give a clear picture of reliability of existing recording system for evaluation of breeding bull in progeny testing programmes in different breeds.

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The milk yield, composition and SCC of dairy bovines show diurnal variations which may affect milk traits at AM-PM milking sessions. Although milk yield, compositions and somatic cells show diurnal variation, the seasonal factors and nutritional status of the animals also play crucial role on such variation (Alhussien and Dang, 2017; Garantjang *et al.,* 2020). It is also essential to identify the milk record (AM or PM) that is more accurate to predict the daily yield, composition and SCC in dairy cows. That is why the present study was planned to see the effect of milking time (AM-PM) on milk yield, 4% fat corrected milk yield (FCM), milk compositions and SCC in Gir cows. Further, the association of AM-PM milk records

with 24-h daily milk traits was also evaluated to identify the milk trait that is more accurate to predict the daily milk yield, composition and SCC.

Materials and Methods

The present experiment was carried out at Cattle Breeding Farm, Junagadh (India) on 50 healthy lactating Gir cows for a period of 2 months during summer season (April-May, 2022). Junagadh is located at 21° 31′ north latitudes, 70° 36′ east longitudes and 60 meters above sea level. The climate of the study area is dry to sub-humid with average annual rain fall of about 625-750 mm. The mean minimum and maximum temperature during the study period (April-May, 2022) was 24.50±0.23 and 40.22±0.26 °C, respectively; while the mean minimum and maximum relative humidity was 27.79±1.35 and 74.72±1.86%, respectively. The experimental cows (1-4 parities) were selected based on good body condition and similar production performance. The cows were maintained under loose housing system with pucca floor. The cows were offered roughage (@ 10 kg green/day and *ad libitum* quantity of dry fodder) and measured quantity of concentrate mixtures (Amul dan, cotton seed cake and ground maize) to fulfill the nutrient requirements as per ICAR (2013). The cows were hand milked twice a day, *i.e.* morning (4.00 AM) and afternoon (4.00 PM) in the milking parlour at 12 h interval and yields were recorded by using a dial type weighing balance. Animals were maintained under uniform managerial practices. A total of 400 milk samples (200 samples each at AM and PM milking sessions) were collected from 50 Gir cows at fortnightly intervals. The milk samples, immediately after collection were transported to the laboratory for estimation of milk compositions and somatic cells. The milk composition (fat, solid not fat, protein, lactose, ash and total solid) was estimated by using the "Lactoscan MCC combo" machine (Milkotronic Ltd., Bulgaria). The milk somatic cell counts (SCC) was estimated by using 4 channel disposable lactochip and reading was noted using the "Lactoscan MCC combo" machine. The daily (24-h) milk yield was calculated by adding the AM and PM yields; while, the daily milk compositions and SCC was calculated by taking average of the AM and PM records. The 4% fat corrected milk yield (FCM) was calculated using standard formula: FCM (kg) = ([0.4×milk yield (kg)] + [15×fat yield (kg)]).

The collected data sets were analysed by using descriptive statistic for mean and standard error (SE). The extreme values (outlier) beyond mean ± 3SD were excluded prior to analysis. The milk yield and compositions at different milking sessions (AM and PM) were compared by paired 't' test. The relationship between AM or PM milk traits (milk yields, composition and SCC) with daily milk traits were carried out by 'Pearson correlation'. Linear regression analysis was performed to predict daily yields, composition and SCC using either AM or PM records. The results were considered statistically significant at 'p≤0.05'. The statistical analyses were carried out using statistical package for the social sciences (SPSS) version 16.

Results and Discussion

AM-PM Milk Yields, Milk Composition and SCC

Milk Yields: The mean value of milk yield and 4% FCM yield at morning (AM) and afternoon (PM) milking sessions are depicted in Table 1. The mean milk yield did not differ between AM and PM milking sessions (p>0.05), which agreed with previous studies conducted on Holstein Friesian cows (Lee and Min, 2013) and Holstein Friesian crossbred cows (Senbeta *et al.,* 2021). However, the 4% FCM was significantly (p=0.021) higher at PM than AM milking sessions. Contrary to this study, several authors reported significantly (p<0.05) higher milk yield at AM than PM milking sessions in different dairy breeds such as Swedish Red (Lakic *et al.,* 2009; Forsback *et al.,* 2010), Holstein Friesian cows (Quist *et al.,* 2008; Bondan *et al.,* 2019; Garantjang *et al.,* 2020; Izzadeen *et al.,* 2021) and crossbred cows (George *et al.,* 2021). The higher yield at AM may be due to the longer milking intervals. The cows also get proper rest at night due to less human activities, increases blood flow to udder which may result in higher yield at AM

Table 1: AM-PM milk yields, composition and SCC in Gir cows

Parameters	Morning (AM) (n=200)	Afternoon (PM) (n=200)	Daily (or 24-h)	'p'-value
Milk yield (kg)	3.67±0.08	3.79±0.11	7.46±0.18	0.178
FCM (kg)	3.90^{a}±0.11	4.17^{b}±0.13	8.07±0.21	0.021
Fat (%)	4.35^{a}±0.07	4.57^{b}±0.07	4.46±0.05	0.019
Protein (%)	3.21^{a}±0.01	3.16^{b}±0.01	3.18±0.01	0.032
Lactose (%)	4.69^{a}±0.01	4.62^{b}±0.02	4.66±0.01	0.009
Ash (%)	0.697^{a}±0.002	0.688^{b}±0.001	0.693±0.001	0.002
SNF (%)	8.59^{a}±0.02	8.48^{b}±0.03	8.53±0.02	0.005
Total solids (%)	12.95±0.08	13.05±0.09	13.00±0.07	0.342
SCC ($\times 10^3$ cells/mL)	187.18±9.90	188.26±10.30	187.72±7.95	0.930

FCM: Fat corrected milk yield (4%); Means with different superscript (a, b) differs significantly in a row.

milking session. However, in this study equal interval was practiced for milking, which might be attributed to the non-significant effect of milking time on milk yield. The traits like milk yield and fat content are used for calculation of FCM, and both of these traits were higher at PM than AM milking session. This might have resulted significantly higher value of FCM in this study at PM than AM milking session.

Milk Fat: There was significant (p=0.019) effect of AM-PM milking sessions on milk fat content (Table 1). The milk fat content was higher at PM than AM milking session (4.57 vs. 4.35%), which was in consonance with the previous studies conducted on Swedish Red cows (Lakic *et al.,* 2009) and Holstein Friesian cows (Quist *et al.,* 2008; Rahman *et al.,* 2016; Bondan *et al.,* 2019; Garantjang *et al.,* 2020). In a similar line, George *et al.* (2021) and Senbeta *et al.* (2021) also reported higher milk fat content in crossbred cows at PM milking session compared to AM milking session. The milk fat content shows inverse relationship with yield which might be attributed to diurnal variation. In this study, though the milk yield was similar at both milking sessions and milking was carried out at equal intervals, the milk fat content was higher at PM milking. The milk fat content also changes during milking process; last drawn milk has higher fat content than the first drawn milk. Incomplete milking increases residual milk volume and decreases milk fat content; while in next milking session the fat content increases (Ravikala *et al.,* 2014). The exercise of animals during day time and high ambient temperature may also affect the milk fat content. These factors may influence the fat content of milk at AM and PM milking sessions.

Milk Protein: The mean milk protein content was 3.21 and 3.16%, respectively, at AM and PM milking (Table 1). The result showed a significantly (p=0.032) higher value of milk protein at AM than PM milk. The present result agreed with Rahman *et al.* (2016), who observed significantly (p<0.01) higher protein content in AM than PM milk in Holstein Friesian cows. Contrary to this study, Senbeta *et al.* (2021) and Izzadeen *et al.* (2021) observed non-significant effect of milking time on milk protein content. There is less day-to-day variation of protein content of milk and it does not change during milking process (Ravikala *et al.,* 2014), which might be attributed to non-significant effect of milking sessions on this trait. Further, several authors reported significantly (p<0.05) higher milk protein content at PM milking session as compared to AM session in Holstein Friesian cows (Quist *et al.,* 2008; Bondan *et al.,* 2019; Garantjang *et al.,* 2020) and Swedish Red cows (Lakic *et al.,* 2009), which contradict to the present findings. The variation of milk protein content at different milking sessions as observed by different studies might be attributed to milk yield due to dilution effect as milk yield and protein yield are negatively correlated (Lakic *et al.,* 2009; Morton *et al.,* 2016).

Milk Lactose: Milking sessions (AM-PM) had significant (p=0.009) effect on milk lactose percentage (Table 1). The AM milking session had higher milk lactose content (4.69%) than PM milking session (4.62%), which was in agreement with the study conducted on Holstein Friesian cows (Rahman *et al.,* 2016). Contrary to this study, Bondan *et al.* (2019) and Izzadeen *et al.* (2021) noted non-significant effect of time of milking on milk lactose content in Holstein Friesian cows. On the other hand, in Swedish Red cows, Lakic *et al.* (2009) observed higher milk lactose content at AM than the PM milking session. In general lactose is least variable (≤1% day-to-day variation) among different milk components due to its osmotic regulatory mechanism (Ravikala *et al.,* 2014). However, during intra-mammary infection lactose shows drastic alteration owing to change in membrane permeability and utilization by microbes (Lakic *et al.,* 2009; Patbandha *et al.,* 2016). In this study, the milk SCC, an indicator of udder infections did not differ at AM and PM milking sessions. Thus the reason of unexpected variation of milk lactose at different milking session is unclear. Further studies are encouraged to find the expected physiological reasons for the diurnal variation of milk lactose in dairy cows during higher ambient temperature.

Milk Ash: The effect of milking time on milk ash content was significant (p=0.002). The AM milk had higher ash content as compared to PM milk (0.697 vs. 0.688%). Contrary to this study, Rahman *et al.* (2016) observed non-significant effect of milking session on milk ash content in Holstein Friesian cows. Further, Garantjang *et al.* (2020) reported variation of results in 2 different experiments depending on types of feed offered to the cows, but the overall milk ash contents at AM and PM milking sessions were similar. The cows fed with complete feed and elephant grass produced milk of higher ash content at PM milking session, while those fed with complete feed, elephant grass and fermented corn cobs produced higher milk ash content at AM milking session (Garantjang *et al.,* 2020). The results reflect that feed of the cows markedly affects the milk ash content at different milking sessions; however, the hidden physiology of diurnal variation is not clear. The higher ambient temperature (about 40^{o}C) during afternoon may affect the mammary gland physiology and thereby affect the milk ash content. The cows also get more rest during night due to less husbandry activities as compared to day, which may affect the milk ash content.

Milk Solid Not Fat (SNF): The milk solid not fat (SNF) content at AM milking session (8.59%) was significantly (p=0.005) higher than the PM milking session (8.48%) (Table 1). In a similar line, Rahman *et al.* (2016) also observed significantly (p<0.05) higher milk SNF at AM than PM milk (8.60 vs. 8.22%). Contrary to this study, several authors reported non-significant effect of AM-PM milking sessions on milk SNF content in Holstein Friesian (Izzadeen *et al.,* 2021; Senbeta *et al.,* 2021) and crossbred cows (George *et al.,* 2021). The higher milk SNF content observed at AM milking session in this study was obvious as the milk components like protein, lactose and ash contents were higher at AM milking session as compared to PM milking session. Further, the breed of cattle used for the studies might have resulted disparity in diurnal variation of milk SNF content.

Milk Total Solid: Milk total solid content of PM milk (13.05%) was numerically higher than the AM milk (12.95%), but didn't differ significantly (p>0.05). However, Rahman *et al.* (2016) in Holstein Friesian cows and George *et al.* (2021) in crossbred cows found significantly higher milk total solid at PM than AM milking session. The milk total solid content is not affected by single milk trait, rather influenced by milk fat, protein, lactose, ash etc. In this study, the fat content was higher during PM milking session, while protein, lactose, ash contents were higher at AM milking session. This might have resulted non-significant effect of AM-PM milking sessions on milk total solid content.

Milk SCC: The AM-PM milking time didn't affect milk SCC in Gir cows (Table 1). The mean milk SCC values at AM and PM milking session were 187.18×10^3 cells/mL and 188.26×10^3 cells/mL, respectively. In a similar line, Quist *et al.* (2008) did not observe any significant variation of milk SCC in Canadian dairy herds at AM-PM milk. Moreover the diurnal variation of milk SCC they observed was inconsistent during five days experimental period. In consonance to the present results, Alhussien and Dang (2017) reported non-significant effect of milking session on milk SCC in Karan Fries crossbred cows during winter (December-January) and thermoneutral (March-April) seasons. Similarly, Bondan *et al.* (2019) also observed non-significant effect of milking session on milk SCC in Holstein Friesian cows. Contrary to our results, Lakic *et al.* (2009) reported lower milk SCC at AM milking session than PM milking session in Swedish Red cows. Alhussien and Dang (2017) observed lower milk SCC at AM than PM during summer season (May-June) in Karan Fries crossbred cows. Lower milk SCC observed at AM than PM milking session by previous authors might be attributed to dilution effect due to higher milk yield at AM milking session. The environmental stresses during summer due to higher temperature-humidity index enhance mammary infection by increasing milk SCC (Alhussien and Dang, 2017). Although, the present study was conducted during extreme summer, the environmental stress has little effect on milk SCC in Gir cows. This non-significant variation of milk SCC in AM-PM milk during extreme summer may be due to better heat tolerant capacity of Gir cows.

Association between Part Time and Daily Milk Parameters

The results (Table 2) indicated that milk yield and 4% FCM at both milking sessions (AM-PM) had strong, positive and significant (p<0.001) correlation with daily yield (r = 0.879 and r = 0.927) and daily FCM (r = 0.848 and r = 0.912, respectively). Similarly, AM-PM milk components like fat, protein, lactose, ash, SNF, total solids and SCC showed strong, positive and significant (p<0.001) correlations with same daily milk parameters (Table 2). The results reflect that, except milk ash content, all parameters had higher correlation coefficient value at PM milking session than the AM milking session. The results were inconsonance with previous studies (Berry *et al.,* 2006; Chladek *et al.,* 2011; McParland *et al.,* 2019), who reported strong, positive and significant association of part time milk parameters with daily same parameters in dairy cows. The strong and positive association between part and daily milk traits could be helpful to predict the 24-h daily milk yield and milk composition in Gir cows.

Table 2: Correlation coefficient of AM-PM records with daily milk traits

Parameters	'r'-value	
	Morning (AM)	Afternoon (PM)
Milk yield	0.879***	0.927***
FCM	0.848***	0.912***
Fat	0.754***	0.786***
Protein	0.769***	0.816***
Lactose	0.717***	0.720***
Ash	0.761***	0.697***
SNF	0.722***	0.764***
Total solids	0.749***	0.802***
SCC	0.777***	0.796***

*** indicates p<0.001

Prediction of Daily Milk Traits from AM-PM Records

The linear regression equations for prediction of daily milk yield, compositions and SCC using part yields, compositions and SCC at AM-PM milking sessions are presented in Table 3. The coefficient of determination (R^2) values ranged from 0.51 to 0.77 at AM milking and from 0.48 to 0.86 at PM milking sessions. The results revealed that among different milk traits at both milking sessions the R^2 value for PM milk yield was higher (86%). In consonance to the present study, Chladek *et al.* (2011) reported higher values of coefficient of determination (R^2 = 0.89-0.95) for prediction of milk yield from part yields (AM-PM yields). However, comparatively higher value of coefficients of determination (R^2 = 0.957 and 0.937 for AM and PM yields, respectively) was observed by Jovanovac *et al.* (2005) for prediction of daily yield from AM-PM part yields. Berry *et al.* (2006) reported that daily SCC can be predicted from either AM or PM milk SCC alone (R^2 = 0.91 and 0.88, respectively) in dairy cows with higher accuracy. Further, Chladek *et al.* (2011) reported comparatively higher value of coefficient of determinations for prediction of fat (R^2 = 0.71-0.85), protein (R^2 = 0.93-0.96), lactose (R^2 = 0.89-0.95) and milk SCC (R^2 = 0.88-0.93). McParland *et al.* (2019) also observed higher value of coefficient of determination to predict daily yields and compositions from part yields and compositions. They also opined that daily yield and composition can be predicted by using a single AM part record.

Table 3: Linear regression equation of milk records to predict daily milk traits

Parameters	Morning (AM)	Afternoon (PM)
Milk yield	y = 0.83 + 1.81x (R^2=0.77)	y = 1.76 + 1.50x (R^2=0.86)
FCM	y = 1.36 + 1.72x (R^2=0.72)	y = 2.11 + 1.43x (R^2=0.83)
Fat	y = 1.86 + 0.59x (R^2=0.57)	y = 1.78 + 0.58x (R^2=0.62)
Protein	y = 1.12 + 0.64x (R^2=0.59)	y = 1.23 + 0.62x (R^2=0.67)
Lactose	y = 2.23 + 0.52x (R^2=0.51)	y = 2.27 + 0.51x (R^2=0.52)
Ash	y = 0.32 + 0.53x (R^2=0.58)	y = 0.32 + 0.53x (R^2=0.48)
SNF	y = 3.77 + 0.55x (R^2=0.52)	y = 3.884 + 0.55x (R^2=0.58)
Total Solids	y = 5.05 + 0.61x (R^2=0.56)	y = 5.26 + 0.59x (R^2=0.64)
SCC	y = 71.12 + 0.62x (R^2=0.60)	y = 72.20 + 0.61x (R^2=0.63)

y = Daily milk traits; x = AM or PM milk traits; R^2 = Coefficient of determination

Conclusions

The morning (AM) milk had lower 4% fat corrected milk yield; but higher protein, lactose, ash and solid not fat content than afternoon (PM) milk. However, there was higher milk fat content at afternoon than morning milking session. The milk traits at both AM and PM milking sessions showed strong and positive association with same daily milk traits. Among different milk traits, afternoon milk yield is more accurate to predict daily yield in Gir cows.

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