RESEARCH ARTICLE

Factors affecting milk yield, milk composition and physico-chemical parameters of ghee in Murrah buffaloes of Punjab region

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Abstract: The objective of this study was to find out the effects of season, stage of lactation, production status and parity on milk yield, milk composition and physico-chemical parameters of ghee in sixty Murrah buffaloes. Milk samples were collected from individual animal during morning in both summer (May - August) and winter (November - February) season. The results indicated that milk yield was significantly influenced by the stage of lactation, production status, and parity. The average fat percentage was significantly affected by season, stage of lactation, production status and parity. However, the protein percentage was affected by the production status and stage of lactation. The Reichert-meissl (RM) value of buffalo ghee was significantly influenced by the season, production status and lactation stage, however, Polenske value (PV) by the season, production status and parity. An effect of season, lactation stage, production status and parity on butyro-refractometer (BR) reading was significant.

Keywords: Murrah buffalo, Milk yield, Milk composition, Physico-chemical parameters of ghee

Introduction

Buffaloes are considered as the major dairy animal of India belongs to the genus *Bubalus bubalis*. The current buffalo population in India as per latest 19th livestock census is 108.7 million which accounts for 21.23% of the total livestock population. Currently there are 51.05 million milch buffaloes which contribute 53% (67.68 million tonnes) of the total milk produced in the country (19th Livestock Census, 2012). Punjab had around 4.75% buffalo population of the country and bestowed with high milk producing breeds such as Murrah and Nili Ravi (19th Livestock Census, 2012). Murrah is one of the superior breeds of Indian buffaloes with a population of 20.49 million, which constitutes about 19.45% of the total buffalo population and is known for high productivity in the country (Chitra et al. 2018). Besides milk production, buffaloes contribute significantly towards meat production, draft power, dung for manure and fuel. To enhance productivity of a dairy animal, it is necessary to develop an understanding of the factors affecting its milk production and composition. The variation in milk production is a regular phenomenon in all milking animals; broadly the factors which are responsible for such variations can be divided into genetic (breeds, stage of lactation) and non-genetic (season, amount and quality of feed, parity, period of calving etc) factors (Bernabucci et al. 2002).

The physico-chemical quality of ghee (clarified butterfat) is usually assessed by analyzing certain characteristics such as Reichert-Meissl (RM) value, Polenske value, Butyro-refractometer (BR) reading, Iodine value and Saponification value. These analytical characteristics are mostly the reflections of the fatty acid composition of the milk lipid. For instance, RM value is substantially a measure of the lower chain volatile water soluble fatty acids i.e butyric acid ($C_{4:0}$) contributes about 3/4th and caproic acid ($C_{6:0}$) about 1/4th to this value whereas Polenske value is a measure of lower chain volatile water insoluble fatty acids i.e caprylic acid ($C_{8:0}$) contributes about 1/4th and capric acid ($C_{10:0}$) about 3/4th to this value. Butyro-refractometer (BR) reading, which measures the index of refraction between air and the liquid fat and varies with the nature of the fat, is usually determined at 40°C. However, the composition of milk as well as fatty acid composition is largely affected by various factors i.e., lactation stage, lactation number, breed, season and environmental factors

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(Yadav et al. 2013). The available information on various factors influencing the composition of milk and physicochemical properties of milk fat/ ghee is scanty in Indian buffaloes. Thus the present investigation was undertaken with the objective to study the effect of various factors affecting the milk yield, milk composition and chemical parameters of ghee prepared from Murrah buffalo's milk.

Materials and Methods

Sample collection and analysis

Sixty Murrah buffaloes from the Livestock farm of Guru Angad Dev Veterinary and Animal Sciences University (GADVASU, Punjab, India) were kept under farm management. Diet of buffaloes was met through green fodder (*ad lib.*) and concentrate according to milk production. All the experimental animals were offered identical ration to meet production/maintenance requirement to negate the feeding effect on milk production. The animals had free access to water throughout the day. Samples of feed sample offered in two different seasons (May – August and November - February) were collected and ground to pass through 1 mm sieve and then analyzed for fat, protein, acid detergent fibre (ADF) and neutral detergent fibre (NDF) content (Robertson and Van Soest, 1981).

The buffaloes were grouped according to production status (high yielders, >15 kg/day; medium yielders, 8-15 kg/day; low yielders, <8 kg/day), stage of lactation (early - up to 100 days of postpartum; mid – 101 to 200 days; late – more than 201 days of postpartum) and parity (1st to 6th lactation). Milk samples were collected from individual animal during morning in both summer (May - August) and winter (November - February) season. Milk yield was recorded after complete milking. Contents of fat, solid-not-fat (SNF) and protein in milk samples were analyzed by a MilkoScreen (Indifoss Analytical Pvt Ltd, Ahmedabad, India). Cream was separated from the each of the individual milk samples by centrifugal method in the cream separator. Cream samples were then converted in to ghee by direct cream method as described by De (2005). Ghee samples were stored in refrigerator (4°C) till for further analysis. Ghee samples were analyzed for Reichert Meissl (RM) and Polenske value as per the standard procedure described in IS: 3508 (ISI, 1966). Butyro refractometer (BR) reading of ghee was

measured using digital butyro refractometer (Atago Co Ltd, Tokyo, Japan).

Statistical analysis

The data was analyzed in a factorial design (Snedecor and Cochran, 1994) by using the software package SPSS version 16 (SPSS, 1996) and differences in mean was assessed by using Tukey's b test.

Results and Discussion

The chemical composition of feed offered to buffaloes in different season is represented in the Table 1. The buffaloes were offered 18.5 and 12.8 kg of dry matter in summer and winter, respectively. The roughage to concentrate ratio was 65:35 and 57:43 per cent in summer and winter, respectively. The roughage portion constituted available green fodder (48%) and silage (17%). The crude protein and fat content were higher in the winter fodder, whereas NDF and ADF content were higher in the summer fodder.

Effect of season on milk yield, milk composition and physico-chemical parameters of ghee

The seasonal differences can attributed to the differences in quality and quantity of available fodder and climatological variation in different seasons which have direct impact on production traits. The data in Table 2 revealed that milk yield of lactating buffalo was not affected by the season. However, fat percent was observed to be significantly higher ($P<0.05$) during winter than summer season. Pawar et al. (2012) observed that milk fat percentage of buffaloes calved in winter season (November – February) was significantly ($P\leq 0.05$) higher than that of the animals calved in summer (March – June) season, due to environmental effects. The content of protein, SNF and daily yield of fat and protein were not affected by the season. Sikka et al. (2004) observed that season of calving did not show any significant difference in protein content of buffalo milk.

The RM and Polenske values of buffalo ghee were observed to be higher ($P<0.05$) during summer than winter season. Mor et al. (2018) also reported that RM and Polenske values of pure buffalo ghee clarified at 110°C vary significantly with season. BR reading of buffalo ghee was found to be lower ($P<0.05$) in summer

Table 1. Composition (% dry matter) of the feed offered to buffaloes in different seasons

Parameter	Season	
	Summer	Winter
Dry matter, kg /day	18.5	12.8
Roughage : Concentrate ratio, %	65:35	57:43
Crude Protein, %	13.0	18.1
Fat, %	2.4	3.13
NDF, %	54.12	44.5
ADF, %	34.01	31.2

Results are expressed as mean values, n=5

compared to winter season. For buffalo milk fat, the lowest BR reading (40.40) was observed in January but highest (41.20) was observed in the July (Kumar et al. 2017). Mor et al. reported that BR reading of buffalo ghee clarified at 110°C was highest in the month of February - March (42.22 ± 0.004) and lowest in month of August -September (41.29 ± 0.004). From the results it can be concluded that effect of season was visible in BR reading, RM and Polenske value of ghee. This may be due to the change in feeding as well as some seasonal variations like heat and humidity, wherein behavior of animals with respect to feed intake and digestion was affected.

Effect of production status on milk yield, milk composition and physico-chemical parameters of ghee

The daily milk yield varied from 4.43 (low yielders) to 13.31 kg (high yielders). The fat and protein content of milk followed the reverse trend and was observed to be highest in milk of low producing animals (Table 3). However, SNF content was not affected by production status of animals. Khan et al. (2011) also reported that high yielders (7.36%) had the lowest fat contents,

followed by moderate (7.46%) and low yielders (7.58%). However, SNF content did not show any significant difference among the groups. The daily yield of fat and protein was observed to be higher ($P < 0.05$) from animals yielding higher milk. Higher fat and protein yield were observed in high milk producing buffaloes than medium and low producing buffaloes.

RM value of buffalo ghee varied from 30.09 (low yielders) to 35.25 (high yielders) (Table 3). RM value was found to be higher ($P < 0.05$) in high yielders than medium and low yielders. The polenske value of ghee was observed to be highest ($P < 0.05$) in medium yielders and lowest ($P < 0.05$) in low yielders. The BR reading increased with decrease in milk yield of animals and values were found to be higher in low yielders than high and medium yielders.

Effect of stage of lactation on milk yield, milk composition and physico-chemical parameters of ghee

Average milk yield was 10.45 kg/day during early lactation, decreased up to 6.5 kg/day during late lactation (Table 4). With

Table 2. Effect of season on milk yield, milk composition and physico-chemical parameters of ghee

Parameter	Season [#]		
	Summer	Winter	PSE
Milk yield kg/day	8.57	8.42	0.21
Fat, %	7.84 ^a	8.73 ^b	0.27
Protein, %	4.55	4.65	0.076
SNF, %	11.22	10.85	0.16
Fat yield, kg/day	0.65	0.70	0.026
Protein yield, kg/day	0.38	0.38	0.01
RM value	34.51 ^b	31.96 ^a	0.42
Polenske value	1.18 ^b	0.85 ^a	0.023
BR reading	40.03 ^a	41.12 ^b	0.11

Mean values with different superscripts in a row differ significantly ($P < 0.05$), $n=60$. [#]Irrespective of lactation number, stage of lactation and production status. PSE – Pooled standard error

Table 3. Effect of production status on milk yield, milk composition and physico-chemical parameters of ghee

Parameter	Production status [#]			PSE
	High yielders	Medium yielders	Low yielders	
Milk yield kg/day	13.31 ^c	7.74 ^b	4.43 ^a	0.26
Fat, %	7.54 ^a	8.39 ^b	8.93 ^c	0.34
Protein, %	4.38 ^a	4.53 ^a	4.80 ^b	0.10
SNF, %	11.09	11.02	11.00	0.20
Fat yield, kg/day	0.99 ^c	0.64 ^b	0.38 ^a	0.033
Protein yield, kg/day	0.58 ^c	0.35 ^b	0.21 ^a	0.013
RM value	35.25 ^c	32.66 ^b	30.09 ^a	0.53
Polenske value	0.92 ^b	1.03 ^c	0.82 ^a	0.029
BR reading	40.08 ^a	40.59 ^b	41.05 ^c	0.014

Mean values with different superscripts in a row differ significantly ($P < 0.05$). [#]Irrespective of lactation number, stage of lactation and season. PSE – Pooled standard error

the advancement of lactation stage, daily milk yield decreased significantly. Results of the present study is in agreement with study of Yadav et al. (2013) who reported that effect of lactation stage on milk yield of Murrah buffaloes was significant which varied from 9.41 (1-12 weeks) to 4.36 kg/day (> 48 weeks). The fat percentage of buffalo milk increased with advancement of stage of lactation and values were higher ($P<0.05$) during late than mid and early lactation. Sethi et al. (1994) reported that lactation stage significantly affected the fat content of buffalo milk. Milk fat content increased with a concomitant decrease in milk yield during advance lactation (Yadav et al. 2013). The increase in total lipid contents may be due to the activity of fatty acid synthesizing enzymes particularly acetyl CoA carboxylase which is a regulatory enzyme in the fatty acid synthesis might have slightly increased in late lactation than early and mid lactation (Sharma et al. 2000). Protein percentage also increased with progress of lactation stage, however, significant difference was not observed between early and mid lactation. Similar result was also reported by Sharma et al. (2000) for fat and protein content of buffalo milk. The results (Table 4) depicted that SNF content, yield of fat and protein remain almost unchanged throughout the lactation period. These

results are in good agreement with those reported by Sharma et al. (2000).

The RM value of ghee decreased with the advancement of lactation stage and values were higher ($P<0.05$) during early than mid and late lactation (Table 4). However, Sharma et al. (2000) reported that short chain fatty acids ($C_{4:0}$ and $C_{6:0}$) of Murrah buffalo milk was higher during mid than early and late lactation. Polenske values were lower in early than mid and late lactation, however, significant difference was not observed. Our results are in good agreement with study of Sharma et al. (2000) who reported that short chain fatty acids ($C_{8:0}$ and $C_{10:0}$) content were lower in early than mid and late lactation. BR reading of ghee increased with advancement of lactation period and values were higher ($P<0.01$) in late than early and mid lactation. Sharma et al. (2000) also observed that total unsaturated fatty acids of buffalo's milk fat were higher during late than early and mid lactations.

Effect of parity on milk yield, milk composition and physico-chemical parameters of ghee

Table 4. Effect of stage of lactation on milk yield, milk composition and physico-chemical parameters of ghee

Parameter	Stage of lactation [#]			PSE
	Early lactation	Mid lactation	Late lactation	
Milk yield, kg/day	10.45 ^c	7.94 ^b	6.50 ^a	0.24
Fat, %	7.24 ^a	8.26 ^b	8.75 ^b	0.32
Protein, %	4.46 ^a	4.47 ^a	4.75 ^b	0.09
SNF, %	11.03	10.82	11.26	0.19
Fat yield, kg/day	0.66	0.69	0.68	0.031
Protein yield, kg/day	0.39	0.38	0.39	0.012
RM value	34.67 ^c	32.69 ^b	31.24 ^a	0.50
Polenske value	0.99	1.05	1.01	0.027
BR reading	40.33 ^a	40.83 ^b	41.45 ^c	0.14

Mean values with different superscripts in a row differ significantly ($P<0.05$). [#]Irrespective of season, lactation number and production status. PSE – Pooled standard error

Table 5. Effect of parity on milk yield, milk composition and physico-chemical parameters of ghee

Parameter	Parity [#]						PSE
	1	2	3	4	5	6	
Milk yield, kg/day	7.85 ^{ab}	8.08 ^{ab}	7.63 ^a	7.32 ^a	8.67 ^b	8.74 ^b	0.33
Fat, %	7.54 ^a	7.75 ^{ab}	8.85 ^b	8.60 ^{ab}	8.66 ^{ab}	8.33 ^{ab}	0.44
Protein, %	4.58	4.53	4.62	4.61	4.65	4.61	0.12
SNF, %	11.00	10.74	11.18	11.11	11.05	11.12	0.26
Fat yield, kg/day	0.59	0.66	0.69	0.68	0.70	0.71	0.42
Protein yield, kg/day	0.37	0.39	0.37	0.38	0.39	0.39	0.015
RM value	33.14	33.27	32.56	33.81	32.88	33.73	0.68
Polenske value	0.96 ^{ab}	0.88 ^a	1.02 ^b	1.02 ^b	1.05 ^{bc}	1.16 ^c	0.037
BR reading	40.86 ^b	40.80 ^b	40.54 ^b	40.63 ^b	41.12 ^b	39.50 ^a	0.18

Mean values with different superscripts in a row differ significantly ($P<0.05$). [#]Irrespective of season, stage of lactation and production status. PSE – Pooled standard error

Effect of parity on milk yield, milk composition and chemical parameters of ghee is represented in the Table 5. The daily milk yield varied from 7.32 (3rd) to 8.74 kg (6th) and it was significantly influenced by the parity but no systematic trend on daily milk yield could be observed. Study corroborates with earlier findings (Yadav et al. 2013) revealing that significant effect of parity on milk yield exists in Murrah buffaloes. Most of the workers have reported increase in milk yield up to fifth parity and decline thereafter (Lee and Kim, 2006; Yadav et al. 2013). However, Bashir et al. (2015) reported that average milk yield increased with parity, peaked in the third lactation and declined in the later parities. Bath et al. (1985) suggested 20% of the increase in milk production with advancing lactations (age) due to increased body weight and 80% increase is due to the effect of recurring pregnancy in cattle. The fat percentage of buffalo milk increased from first to third lactation and then declined in subsequent lactations. Similar result was also reported by Sodhi et al. (2008) in Murrah buffaloes. Verma et al. (2017) reported that milk fat percentage varied significantly among the parities with no consistent increase over the advancement of the parities. The content of protein and SNF and daily yield of fat and protein were not differed significantly in different parities. Similar results were reported by Sodhi et al. (2008) for the protein and lactose content in buffalo milk. Milk protein level did not vary significantly over the parities till sixth parity (Yadav et al. 2013). Sikka et al. (2004) reported that no significant difference was found in milk protein in relation to the variation in parity of buffaloes.

RM value of ghee did not differ significantly ($p>0.05$) in different parity, however it varied from 32.56 to 33.81. The Polenske value was observed to be highest ($p<0.05$) of ghee obtained from animals in 6th parity and low in 1st parity. BR reading varied from 39.5 (6th) to 41.1 (5th). There was no significant difference in BR reading was observed till 5th parity and later decreased during 6th parity.

Conclusions

From study, it was found that effect of season, lactation stage, production status and parity were visible in milk yield, milk composition and chemical parameters of buffalo ghee. Average milk yield was significantly influenced by the stage of lactation, production status, and parity. The fat percentage was significantly affected by season, stage of lactation, production status and parity. However, the protein percentage was affected by the production status and stage of lactation. The RM value of buffalo ghee was significantly influenced by the season, production status and lactation stage, however, polenske value by the season, production status and parity. An effect of season, lactation stage, production status and parity on BR reading was significant. The values obtained for different physico-chemical constants of ghee were within the limit specified by Food Safety and Standard (Food Products Standards and Food Additives)

Regulations, 2011 (FSSAI, 2006) irrespective of season, lactation stage, production status and parity.

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