



The livestock sector in Pakistan occupies a unique position in the national agenda of economic development. The current study was conducted to evaluate a heat stress mitigation strategy through different feeding times on the performance i.e. milk yield, milk composition and physiochemical properties of milk in dairy cattle during the summer season. Twenty Holstein Friesian cows (HF) were randomly divided into four groups (G1. 20/80, G2. 50/50, G3. 0/100, G4. 100/0 feeding at day/night %). Milk yield, milk composition, physiochemical properties and malondialdehyde (MDA) in blood serum were determined. In the first week morning time milk yield was significantly highest in group G1 followed by G2, G3 and G4, respectively. During the 2nd to 8th weeks in the morning time it was highest in G2 followed by groups G1, G3 and G4 followed. It was highest in G1 followed by G4, G3 and G2 in the evening time during 1 to 8th weeks. In 5th week milk yield was more in G4, G3, G2 and G1 in the evening time. In all weeks (1 to 8) pH of milk was higher in groups G1, G2 and G4 followed by G3. In weeks 1 and 2, protein and lactose contents were similar among the four groups, while the fat percentage was higher in G4 followed by G3, G2 and group G1. In weeks 3 and 4 fat was more in the G4 and G3 groups respectively. In the third week pH, acidity and specific gravity of milk was not changed significantly in all the groups. In the fourth week pH was slightly reduced in G3, while acidity and specific gravity remained the same in all the groups. In group G5 pH and specific gravity of milk was comparatively high in group G1 followed by G2 and G3, while acidity was more in G1 as compared to other groups. Similarly pH and specific gravity of milk in the sixth week was slightly more in G1 followed by other three groups. In the seventh week fat was more in G4 followed by G3, G2 and G1. In week 1 levels of MDA were high in G4 followed by G3, G2 and G1. Similar results were found in weeks 2, 3, 4, 5 and 8. In the sixth and seventh weeks the level of MDA was high in G4 followed by G2, G3 and G1. It was concluded that changes in feeding time significantly increased milk yield in those groups which were fed in the cooler part of the day. Milk composition and its physiochemical properties were also affected due to variation in milk yield in the morning and evening times. Hence change in feeding time during the summer season in tropical areas is a good husbandry practice to mitigate heat stress effects on the production performance of dairy cattle.

KEY WORDS feeding time, malondialdehyde, milk composition, mitigation strategy, summer stress.

INTRODUCTION

The accumulation of greenhouse gases in the Earth's at mosphere has led to predictions that global surface tem

peratures will increase between 1 and 6 °C during the 21st century (Climate Change, 2007). The heat dissipation limits hypothesis (HDL) (Speakman and Król, 2010) suggests that lactating mammals produce heat directly during milk syn-

thesis and are limited in their capacity to dissipate the excess heat load.

Because heat dissipation capacity depends on ambient temperature, and a lactating animal's milk yield is predicted to decline as it gets warmer. With a reduction in milk yield, profits suffer on a global scale.

A condition in which body temperature is so high that it interacts with normal homeostatic processes affecting the respiratory, circulatory and solution balances leading to collapse and death.

There are three temperature zones as follow: zone of physical condition, zone of normal body temperature and zone of hyperthermia (heat stress).

Warming and the breeding of selected animals that are more and more susceptable to surrounding effects have made this process, named heat stress (HS), particularly relevant even in moderate areas (Ferreira, 2013; Nardone *et al.* 2010).

Summer heat may affect milk yield either by direct or indirect actions arbitrates with change of proportions of energy. Milking cow have a balance between energy, ambient heat, milk yield, suckling stage and intake of dry matter, which decrease release of luteinising hormones and size of the recessive follicle during the postnatal period (Ronchi *et al.* 2001).

Heat stress increases body temperature, respiration rate, and reduces feed intake and milk production (Hahn, 1999; Omeenaski *et al.* 2002; West, 2003). Monitoring animal performance in measured thermal environments over longer periods of time (weeks to months) permits development of response functions for growth, milk production or other performance measures as functions of air temperature (Hahn *et al.* 2003).

Malondialdehyde (MDA) is a reactive species of electrophile and is a reactive aldehyde compound which produces toxic substances in tissue cells due to formation of covalent protein draws wounded to end-products of progress lipoxidation, interference to end product of progress glycation (Farmer and Davoine, 2007).

MATERIALS AND METHODS

This study was conducted in the Cattle Breeding and Dairy Farm Harichand District Charsadda and the laboratory work was performed in the Department of Animal Health, the University of Agriculture Peshawar. The study was performed during the hot months of the year (i.e. June to August) to explore the effect of different feeding times on the milk yield, milk composition and physiochemical properties of milk in Holstein Friesian (HF) dairy cattle during the summer season.

Animals

Twenty Holstein Friesian cows (HF) with a live weight 400 to 450 kg and body condition score (BCS) 3.0 to 4.0 of the same parity and mid lactation were used. The animals selected for study were further divided into four groups G1, G2, G3 and G4 (five animals in each group).

Experimental layout

Animals were housed in an open shed, having water trough and open yards. Group one (G1) received 80% of total allowance of a mixed ration between 8 p.m. to 6 a.m. and the remaining 20% was offered at 11 a.m. to 4 p.m. Group two (G2) was provided with 50% feed between 8 p.m. to 6 a.m. and the remaining 50% was offered from 6 a.m. to 8 p.m. The 100% feeding to group three (G3) was offered from 8 p.m. to 6 a.m. Group four (G4) was provided with 100% feed from 6 a.m. to 6 p.m. (Table 1).

Table 1 Ex	perimental	layout is	shown	in	this	table
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S.No	Groups	Number of animals	Feeding D/N (%)*
1	G1	5	20/80
2	G2	5	50/50
3	G3	5	0/100
4	G4	5	100/0

D: day time and N: night time.

* G1: 20% feed at day time and 80% feed at night time; G2: 50% feed at day time and 50% feed at night time; G3: 100% feeding in night time and G4: 100% feeding in day time.

The experimental diet contained green forages, wheat straw and concentrates. Water was provided ad libitum.

Milk yield

Data regarding milk yield from each group was recorded at morning and evening milkings.

Milk composition and physiochemical properties

Milk samples were collected in clean plastic bottles. These bottles were labeled with sample number, group number, date of collection etc. Samples were analyzed through EKO milk (ultrasonic milk analyzer) for fat and solids-not-fat (i.e. protein plus lactose percentage).

Determination of MDA in blood serum

A serum sample (0.2 mL) was added into the mixture containing 0.2 mL SDS, 1.5 mL acetic acid solution adjusted to pH 3.5 with Na OH and 1.5 mL aqueous solution of TBA, adding 0.6 mL distal water and then heated at 95 °C for 60 min. After cooling with tap water 0.1 mL distill water and 5.0 mL mixture of n- butanol and pyridine (15:1, v/v) was added and shaken vigorously. The organic layer was taking after centrifugation at 4000 rpm for 10 min and MDA was quantified using a spectrophotometer at 532 nm (Ohkawa *et al.* 1979).

Statistical analysis

Each parameter was recorded from the beginning of the experiment on a daily and weekly basis. At the end, all the recorded data were subjected to analysis of variance by using a completely randomized design (CRD) model in SPSS program (AOAC, 1990).

 $Y_{ij} = \mu + a_i + \Sigma_{ij}$

Where:

 $\begin{array}{l} Y_{ij} : \mbox{ milk + milk properties (independent variable).} \\ \mu : \mbox{ population constant.} \\ a_i : \mbox{ effect of time.} \\ i: \mbox{ day, night.} \\ \Sigma_{ij} : \mbox{ random residuals.} \end{array}$

RESULTS AND DISCUSSION

Mean morning and evening milk yield observed in all four groups during 1st, 2nd, 3rd, 4th, 5th, 6th, 7th and 8th week of the trial is shown in Table 2 a, b, c and d. In week one morning milk yield was significantly (P<0.01) higher in G1 followed by G2, G3 and G4 respectively. In the evening milking, G1, G4, G2 and G3 had the highest milk yield as compared to other groups. Milk yield during the second week of morning lactation G2 had the highest mean value followed by G1, G3 and G4. While in evening time, G1 had the highest yield followed by G4, G3 and G2. Total milk yield was highest in G1 followed by G2, G3 and G4 during the first and second weeks as shown in Table 2 (a). Milk yields during the third and fourth weeks are shown in Table 2 (a). In week 3 morning milk yield was significantly higher in G2 followed by G1, G3 and G4 respectively, while it was highest in G1 followed by G4, G3 and G2, in the evening. Similarly milk yield during the morning milking in week 4 was highest in G2 followed by G1, G3 and G4, respectively. While in evening time it was highest in G1 followed by G4, G2 and G3 groups. Total milk yield was highest in G1 followed by G2, G3 and G4 during week 3, while it was more in G2 followed by G1, G3, and G4 during week 4.

Milk yields during the fifth and sixth weeks of lactation are shown in Table 2 (c). In week 5, morning time milk yield was significantly high in G2 followed by groups G1, G3 and G4 respectively, while it was more in G4, G3, G2 and G1 respectively in the evening, milk yield during morning during the week 6 of lactation was high in G2 followed by G1, G3 and G4, respectively while in evening; it was significantly higher in group G1 followed by G4, G2 and G3, respectively. Total milk yield was highest in G2 followed by groups G1, G4 and G3 during week 5 of lactation while it was more in group G1, G2, G3 and G4 during week 6 of lactation. Milk yields during the seventh and eighth weeks of lactation are shown in Table 2 (d). In week 7, morning milk yield was significantly higher in group G2 followed by groups G1, G3 and G4 respectively, while it was more in G1, followed by G4, G3 and G2 respectively in the evening.

Similarly milk yield during the morning in week 8 was high in G2 followed by G1, G3 and G4, respectively. In the evening it was highest in G1 followed by G4, G3 and G2. Total milk yield was highest in G1 followed by G2, G3 and G4 during the seventh and eighth weeks.

Ominski et al. (2002) tested the response of morning-fed and evening-fed cows to short-term heat stress and found that evening feeding did not alleviate production losses associated with heat stress. Beede and Collier (1986) identified three management strategies to minimize the effects of heat stress. The present study was comparable to the study of Igono et al. (1992) and Tahmasbi et al. (2012). They observed that the milk production was more in morning as compare to evening due to heat stress. Change feed intake of high-production dairy cattle in summer time from day to night hours resulted in a decline of energy expenditure during the hot daytime hours, with no compensation for this decline during night hours (Aharoni et al. 2005). Estimated milk yield reduction was 0.32 kg per unit. Mean milk composition observed in all four groups during the 1st, 2nd, 3rd, 4th, 5th, 6th, 7th and 8th week of trials is shown in Table 3 a, b, c and d.

In the first and second weeks of lactation, protein and lactose contents remained constant in all groups, while the fat content was highest in G4 followed by group G3, G2 and G1 respectively. Ash content was the same in all groups as shown in Table 3 (a). In the third and fourth weeks fat content was highest in G4 and G3; it was comparatively less in G1 and G2 (Table 3 b).

Mean milk composition of all the experimental groups in the fifth and sixth weeks is shown in Table 3 (c). In week 5, lactose content was the same in all the four groups. Fat was highest in G4 followed by G3, G2 and G1. Protein was highest in G1 as compared to other groups. No variation was observed in ash content in all four groups. In the fifth week fat content was highest in G4 followed by group G3, G2 and G1, while in week 6 it was highest in G1 followed by G4, G3 and G2 respectively. Ash content remained the same in all the groups in week 6.

Mean milk composition of all the experimental groups in the seventh and eighth weeks is shown in Table 3 (d). In the seventh week of lactation, fat content was highest in G4 followed by G3, G2 and G1. Protein, lactose and ash contents were same in all groups. Similar results for fat, protein, lactose and ash contents were found in the eighth week.

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	Mea	n±SE	_	Mean±SE					
Groups	Wee	ek 3 rd	Total	Weel	κ 4 th	Total			
	М	Ε	-	М	Е				
G1	58.20 ^a ±0.37	48.40 ^a ±0.24	106.6 ^a ±0.50	61.70 ^b ±0.30	50.40°±0.24	112.10 ^a ±0.50			
G2	54.20 ^b ±0.37	26.40°±0.40	80.6 ^b ±0.74	66.20 ^a ±0.37	20.20 ^d ±0.20	87.00 ^b ±0.54			
G3	50.70 ^c ±0.30	21.00 ^d ±0.44	71.70 ^c ±0.48	58.20°±0.37	29.40°±0.40	87.60 ^b ±0.74			
G4	22.40 ^d ±0.40	40.00 ^b ±0.54	62.00 ^d ±2.91	23.40 ^d ±0.40	42.20 ^b ±0.37	65.60°±0.60			

G1: 20% feeding in day time and 80% in night; G2: 50% feeding in day and 50% during night; G3: 100% feeding in night time; G4: 100% feeding in day time. M: morning time and E: evening time.

The means within the same column with at least one common letter, do not have significant difference (P>0.05).

SE: standard error.

Table 2 b Mean milk yield (liter) per group during the 3rd and 4th weeks of the experimental trials

	Mea	n±SE				
Groups	Wee	ek 3 rd	Total	Wee	k 4 th	Total
	М	Ε	-	М	Ε	
G1	58.60 ^b ±0.60	50.60 ^a ±0.24	109.20 ^a ±0.73	59.40 ^b ±0.40	49.20 ^a ±0.37	108.60 ^b ±0.60
G2	79.80 ^a ±0.37	21.60°±0.40	101.40 ^b ±0.67	88.20 ^a ±0.70	26.20 ^b ±0.70	114.40 ^a ±0.67
G3	50.60°±0.24	22.60°±0.60	73.20°±0.58	51.20°±0.37	25.20 ^b ±0.37	76.40°±0.50
G4	22.80 ^d ±0.20	28.30 ^b ±0.43	51.10 ^d ±0.40	22.80 ^d ±0.20	30.38°±0.49	53.18 ^d ±0.56

G1: 20% feeding in day time and 80% in night; G2: 50% feeding in day and 50% during night; G3: 100% feeding in night time; G4: 100% feeding in day time. M: morning time and E: evening time.

The means within the same column with at least one common letter, do not have significant difference (P>0.05). SE: standard error.

Table 2 c Mean milk yields (liter) per group during the 5th and 6th weeks of the experimental trials

	Mea	n±SE		Mea	n±SE	
Groups	Week 5 th		Total	Wee	k 6 th	Total
	Μ	Ε		Μ	Ε	
G1	60.40 ^b ±0.24	49.40 ^a ±0.40	109.80 ^b ±0.58	59.50 ^b ±0.44	48.30 ^a ±0.24	108.10 ^a ±0.60
G2	86.60 ^a ±0.30	23.40 ^d ±0.80	110.00 ^a ±0.31	77.40 ^a ±0.50	28.00°±0.44	105.40 ^b ±0.74
G3	59.60 ^b ±0.24	27.40°±0.40	87.00 ^c ±0.40	49.60°±0.24	26.60°±0.24	76.20°±0.48
G4	20.30°±0.24	29.80 ^b ±0.58	50.40 ^d ±0.67	14.84 ^d ±0.21	31.00 ^b ±0.44	45.84 ^d ±0.51

G1: 20% feeding in day time and 80% in night; G2: 50% feeding in day and 50% during night; G3: 100% feeding in night time; G4: 100% feeding in day time. M: morning time and E: evening time.

The means within the same column with at least one common letter, do not have significant difference (P>0.05).

SE: standard error.

Table 2 d Mean milk yield (liter) per group during the 7th and 8th weeks of the experimental trials

Mean±SE Mean±SE							
Groups	Wee	k 7 th	Total	Wee	k 8 th	Total	
	М	Ε	_	М	Ε		
G1	59.20 ^b ±0.37	50.40 ^a ±0.24	109.60 ^a ±0.40	54.80 ^b ±0.20	47.60 ^a ±0.40	102.40 ^a ±0.40	
G2	70.20 ^a ±0.37	25.20 ^d ±0.37	95.40 ^b ±0.74	74.20 ^a ±0.37	26.40 ^d ±0.40	100.60 ^b ±0.50	
G3	47.40°±0.40	29.40°±0.40	76.80°±0.73	50.40°±0.24	34.40°±0.40	84.80°±0.37	
G4	20.60 ^d ±0.24	36.60 ^b ±0.74	57.20 ^d ±0.96	18.60 ^d ±0.24	36.60 ^b ±0.24	55.20 ^d ±0.37	

G1: 20% feeding in day time and 80% in night; G2: 50% feeding in day and 50% during night; G3: 100% feeding in night time; G4: 100% feeding in day time.

M: morning time and E: evening time.

The means within the same column with at least one common letter, do not have significant difference (P>0.05).

SE: standard error.

The results of fat content in the present study are in agreement with Charnobai *et al.* (1999) who reported that milk yield affects the fat content of cows milk. They stated that heat stress can change the chemical composition. Similar findings were observed by Haenlein (1996) who studied the effect of lactation on physico-chemical properties of local cows milk. The protein and lactose contents in the current study are in agreement with Antunac *et al.* (2001).

They found that heat stress can cause reduction in milk production, which ultimately resulted in variation in protein and lactose composition of total milk. The ash content in the present study is similar to that of Aganga *et al.* (2002) who investigated the relation of milk yield with ash content. He proved that there was no significant relation of milk production and ash content of milk. The present results is also in line with the reports of Gordon and Forbes (1970), who found that variation in feeding time and milk yield can greatly influence the basic milk composition of dairy animals. Similar relation between heat stress and milk composition was investigated by Klusmeyer *et al.* (1990) in dairy cattle and they found significant changes in milk composition in dairy cows due to heat stress.

C		Week 1 st				Week 2 nd				
Group		Mean±SE			Mean±SE					
	Fat	Protein	Lactose	Ash	Fat	Protein	Lactose	Ash		
G1	3.32°±0.03	3.56±0.02	4.58±0.05	$0.78^{a} \pm 0.02$	3.30±0.03	3.52±0.02	4.62±0.02	$0.74^{a} \pm 0.02$		
G2	3.52 ^b ±0.02	3.36±0.02	4.48±0.03	$0.74^{a}\pm 0.02$	3.46±0.02	3.32±0.03	4.48±0.03	0.74 ^a ±0.02		
G3	3.58 ^b ±0.02	3.36±0.02	4.30±0.03	$0.70^{a} \pm 0.03$	3.56±0.02	3.38±0.05	4.28±0.03	0.70°±0.02		
G4	3.74 ^a ±0.04	3.36±0.04	4.26±0.04	0.66 ^b ±0.02	3.76±0.05	3.36±0.02	4.24±0.06	0.72 ^b ±0.02		

Table 3 a Mean milk composition during the1st and 2nd weeks of the experimental trials

G1: 20% feeding in day time and 80% in night; G2: 50% feeding in day and 50% during night; G3: 100% feeding in night time; G4: 100% feeding in day time. The means within the same column with at least one common letter, do not have significant difference (P>0.05). SE: standard error

Table 3 b Mean milk composition during the 3rd and 4th weeks of the experimental trials

C		Week 3 rd				Week 4 th			
Group		Mean±SE				Mea	n±SE		
	Fat	Protein	Lactose	Ash	Fat	Protein	Lactose	Ash	
G1	3.32±0.03	3.56±0.02	4.58±0.03	0.72 ^c ±0.04	3.36±0.02	3.54±0.02	4.56±0.04	0.70 ^d ±0.06	
G2	3.56 ± 0.02	3.34 ± 0.05	4.48±0.03	$0.74^{b}\pm 0.04$	3.56±0.02	3.34 ± 0.05	4.52±0.03	0.76 ^b ±0.02	
G3	3.50±0.04	3.34±0.04	4.28±0.02	$0.76^{a} \pm 0.02$	3.52±0.03	3.36 ± 0.02	4.32±0.03	$0.78^{a}\pm0.02$	
G4	3.62±0.06	3.42±0.03	4.24±0.05	$0.76^{a} \pm 0.02$	3.60±0.04	3.34±0.04	4.26±0.06	$0.72^{\circ}\pm 0.02$	

G1: 20% feeding in day time and 80% in night; G2: 50% feeding in day and 50% during night; G3: 100% feeding in night time; G4: 100% feeding in day time. The means within the same column with at least one common letter, do not have significant difference (P>0.05). SE: standard error.

Table 3 c Mean milk compositions during the 5th and 6th weeks of the experimental trials

C		Wee	ek 5 th		Week 6 th				
Group		Mea	n±SE		Mean±SE				
	Fat	Protein	Lactose	Ash	Fat	Protein	Lactose	Ash	
G1	3.38±0.02	4.54 ^a ±0.05	4.54±0.05	0.72 ^b ±0.06	3.38±0.02	3.54 ± 0.02	4.58±0.02	0.72 ^c ±0.05	
G2	3.46±0.02	3.36 ^b ±0.06	4.58±0.02	$0.74^{a}\pm 0.00$	3.52±0.02	3.34±0.04	4.46 ± 0.04	$0.76^{a}\pm0.04$	
G3	3.56±0.04	3.38 ^b ±0.02	4.32±0.03	$0.70^{b} \pm 0.04$	3.68 ± 0.04	$3.40{\pm}0.04$	4.30±0.03	0.74 ^b ±0.04	
G4	3.72±0.03	3.40 ^b ±0.05	4.36±0.02	0.70 ^b ±0.02	3.86±0.02	3.42 ± 0.02	4.28±0.04	0.72°±0.02	

 G_1 : 20% feeding in day time and 80% in night; G_2 : 50% feeding in day and 50% during night; G_3 : 100% feeding in night time; G_4 : 100% feeding in day time. The means within the same column with at least one common letter, do not have significant difference (P>0.05).

SE: standard error.

Table 3 d Mean milk composition during the 7th and 8th weeks of the experimental trials

C		We	ek 7 th		Week 8 th				
Group		Mea	n±SE		Mean±SE				
	Fat	Protein	Lactose	Ash	Fat	Protein	Lactose	Ash	
G1	3.32±0.03	3.54±0.02	4.60±0.03	0.74 ^b ±0.02	3.46±0.02	3.58±0.02	4.60±0.03	0.74 ^b ±0.02	
G2	3.42±0.03	3.36 ± 0.02	4.50±0.04	0.76 ^a ±0.03	3.52 ± 0.02	3.40 ± 0.04	4.56±0.02	$0.76^{a}\pm0.04$	
G3	3.52±0.03	3.40 ± 0.04	4.32±0.03	0.72°±0.02	3.68 ± 0.02	3.36 ± 0.02	4.30±0.03	0.72°±0.03	
G4	3.82±0.04	3.32±0.02	4.30±0.04	0.72°±0.02	3.88±0.07	3.40±0.04	4.34±0.04	0.74 ^b ±0.02	
G1 . 20% feed	ing in day time and	80% in night: G2: 5	0% feeding in day a	nd 50% during night	· G3 · 100% feeding	in night time: GA:	100% feeding in da	v time	

G1: 20% feeding in day time and 80% in night; G2: 50% feeding in day and 50% during night; G3: 100% feeding in night time; G4: 100% feeding in day time

The means within the same column with at least one common letter, do not have significant difference (P>0.05).

SE: standard error

The mean physiochemical properties of milk observed in the four groups during the 1st, 2nd, 3rd, 4th, 5th, 6th, 7th and 8th weeks is shown in Table 4 (a), b, c and d. In the first week pH of milk was highest in G1, G2, and G4 followed by G3 respectively. In the second week pH of milk was slightly higher in G1 followed by the other groups. Similar results were found for acidity and specific gravity in the first and second weeks (Table 4 a). Mean physiochemical properties of all the experimental groups in the third and fourth weeks are shown in Table 4 (b).

In the third week pH, acidity and specific gravity values of milk were not significantly changed in all experimental groups. In the fourth week, pH was slightly reduced in G3, while acidity and specific gravity remained the same in other groups. In G5 pH and specific gravity values of milk were comparatively highest in G1 followed by G2 and G3 while acidity was highest in G1 as compared to other groups. Similarly pH and specific gravity of milk in the sixth week was slightly higher in group G1 followed by other groups.

	Table 4 a Mean physiochemical	properties of milk during the 1st and 2nd	^d weeks of the experimental trials
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Group		Week 1 st			Week 2 nd	
		Mean±SE			Mean±SE	
	pН	Acidity	Specific gravity	pН	Acidity	Specific gravity
G1	$6.60^{a} \pm 0.07$	0.16 ± 0.00	1.033 ± 0.00	$6.68^{a} \pm 0.08$	0.15±0.00	1.032 ± 0.05
G2	6.56 ^a ±0.02	0.15±0.00	1.022 ± 0.00	6.48 ^b ±0.04	0.16±0.00	1.023 ± 0.00
G3	6.46 ^b ±0.04	0.12 ± 0.00	1.013 ± 0.00	6.46 ^b ±0.04	0.15±0.00	1.013 ± 0.00
G4	6.52 ^a ±0.03	$0.14{\pm}0.00$	1.001 ± 0.00	6.46 ^b ±0.02	0.15±0.00	1.002 ± 0.00

G1: 20% feeding in day time and 80% in night; G2: 50% feeding in day and 50% during night; G3: 100% feeding in night time; G4: 100% feeding in day time. The means within the same column with at least one common letter, do not have significant difference (P>0.05). SE: standard error

Table 4 b Mean physiochemical properties of milk during the 3rd and 4th weeks of the experimental trials

Group		Week 3rd			Week 4 th	
		Mean±SE			Mean±SE	
	рН	Acidity	Specific gravity	pН	Acidity	Specific gravity
G1	6.60±0.07	$0.14{\pm}0.00$	1.033±0.00	6.56 ^a ±0.06	0.15±0.00	1.032 ± 0.00
G2	6.48±0.03	0.15 ± 0.00	1.013 ± 0.00	6.56 ^a ±0.04	0.16±0.00	1.013 ± 0.00
G3	6.56 ± 0.04	0.13±0.00	1.013 ± 0.00	6.48 ^b ±0.03	$0.14{\pm}0.00$	1.013 ± 0.00
G4	6.52±0.02	$0.14{\pm}0.00$	1.012 ± 0.00	6.50 ^a ±0.03	0.15±0.00	1.002 ± 0.00
G1 · 20% feedir	ng in day time and 80%	in night: G2: 50% feedi	ng in day and 50% during nigh	t: G3: 100% feeding in	night time: G4: 100% f	eeding in day time

The means within the same column with at least one common letter, do not have significant difference (P>0.05). SE: standard error.

Table 4 c Mean physiochemical properties of milk during the 5th and 6th weeks of the experimental trials

C		Week 5 th			Week 6 th	
Group		Mean±SE			Mean±SE	
	pН	Acidity	Specific gravity	pН	Acidity	Specific gravity
G1	6.72 ^a ±0.03	0.62 ^b ±0.07	1.032 ^a ±0.00	6.60 ^a ±0.08	0.03 ^b ±0.00	1.032 ^a ±0.00
G2	6.54 ^b ±0.04	$0.15^{a} \pm 0.00$	1.023 ^b ±0.00	6.58 ^b ±0.03	$0.16^{a} \pm 0.00$	1.013 ^b ±0.00
G3	6.48°±0.04	$0.14^{a} \pm 0.00$	1.013 ^b ±0.00	6.52 ^b ±0.04	0.13 ^a ±0.00	1.012 ^b ±0.00
G4	6.52 ^b ±0.03	0.15 ^a ±0.00	1.002 ^b ±0.00	6.50 ^b ±0.03	$0.14^{a}\pm 0.00$	1.000 ^b ±0.00

G1: 20% feeding in day time and 80% in night; G2: 50% feeding in day and 50% during night; G3: 100% feeding in night time; G4: 100% feeding in day time. The means within the same column with at least one common letter, do not have significant difference (P>0.05). SE: standard error.

Table 4 d	Mean	physio	chemical	properties	of milk	during	the 7 th	¹ and 8 ¹	th weeks	of the ex	perimental tr	ials
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Group		Week 7 th			Week 8 th		
		Mean±SE		Mean±SE			
	pH	Acidity	Specific gravity	рН	Acidity	Specific gravity	
G1	6.72 ^a ±0.03	0.15±0.01	1.032 ± 0.00	6.56 ^a ±0.06	0.16±0.00	1.023±0.00	
G2	6.48 ^b ±0.04	0.15±0.00	1.023 ± 0.00	6.54 ^a ±0.02	0.15±0.00	1.013 ± 0.00	
G3	6.02 ^c ±0.00	0.15±0.00	1.013 ± 0.00	6.48 ^b ±0.03	0.13±0.00	1.012 ± 0.00	
G4	6.50 ^{ab} ±0.03	0.15±0.00	1.00 ± 0.00	$6.56^{a} \pm 0.02$	0.14±0.00	1.000 ± 0.00	

G1: 20% feeding in day time and 80% in night; G2: 50% feeding in day and 50% during night; G3: 100% feeding in night time; G4: 100% feeding in day time. The means within the same column with at least one common letter, do not have significant difference (P>0.05).

SE: standard error

Acidity was also increased in G1 followed by G2, G3 and G4 respectively (Table 4 (c)). The means of the physiochemical properties of all the experimental groups in the seventh and eighth weeks are shown in Table 3 (b). In the seventh and eighth weeks, pH of milk was significantly highest in G1, while the acidity and specific gravity of milk were not significantly changed in all the experimental groups.

The pH of cow milk ranges from 6.57 to 6.84 and is not influenced by month, lactation number, or season of calving (Minieri *et al.* 1965). Acidity varies from 0.05% to 0.20%.

The findings of the present study are in agreement with Arguello *et al.* (1998).

Bhosale *et al.* (2009) found that the pH, acidity and specific gravity similar to that of present study. The results of the present study are also comparable to the study of Asif Mahmood *et al.* (2010) who found that pH values were in the range of 6.53-7.00 in buffalo milk, 6.59-6.67 in cow milk, 6.48-6.64 in goat milk and 6.55-6.68 in sheep milk. Specific gravity was found in the range for cow milk of 1.028-1.032. The specific gravity of cow milk was similar that cited by Jenness *et al.* (1974).

The mean levels of malondialdehyde (MDA) in blood serum observed in the experimental groups during the 1st, 2nd, 3rd, 4th, 5th, 6th, 7th and 8th weeks of the trial are shown in Table 5.

Table 5 Mean levels of malondialdehyde (MDA) in blood serum of the experimental groups

Group -	Mean±SE									
	Week 1 st	Week 2 nd	Week 3rd	Week 4 th	Week 5 th	Week 6 th	Week 7 th	Week 8 th		
G1	$0.15^{d} \pm 0.01$	$0.15^{d} \pm 0.01$	$0.19^{d} \pm 0.02$	0.15 ^d ±0.01	$0.14^{d} \pm 0.01$	$0.15^{d} \pm 0.01$	$0.15^{d} \pm 0.01$	$0.15^{d} \pm 0.00$		
G2	$0.28^{c}\pm 0.02$	0.29 ^e ±0.01	$0.28^{c} \pm 0.01$	0.28°±0.02	0.28 ^c ±0.02	0.61 ^b ±0.18	0.49 ^b ±0.13	0.28°±0.02		
G3	0.49 ^b ±0.03	0.49 ^b ±0.03	0.49 ^b ±0.02	$0.48^{b} \pm 0.026$	$0.48^{b} \pm 0.05$	0.50°±0.03	0.41°±0.02	0.50 ^b ±0.02		
G4	$0.88^{a} \pm 0.18$	0.90 ^a ±0.18	0.87 ^a ±0.17	0.88 ^a ±0.14	$0.78^{a}\pm 0.07$	0.89 ^a ±0.18	0.82 ^a ±0.11	$0.88^{a} \pm 0.17$		

G1: 20% feeding in day time and 80% in night; G2: 50% feeding in day and 50% during night; G3: 100% feeding in night time; G4: 100% feeding in day time. The means within the same column with at least one common letter, do not have significant difference (P>0.05).

SE: standard error.

In the first week of lactation, the level of MDA was high in G4 followed by G3, G2 and G1. Similar results were found in the second, third, fourth, fifth and eighth weeks of lactation. In the sixth and seventh weeks the level of MDA was highest in G4 followed by groups G2, G3 and G1. The increase in the levels of MDA is an indicator of heat stress generated because of exposure of animals to hot weather and feeding of animals in hot times of the day.

CONCLUSION

The following conclusions were drawn from the present study. Changes in feeding time during the summer affects the production performance of dairy animals. Offering 80% feeding at night time and 20% at day time during the summer season mitigates heat stress and increases milk production. In hot weather conditions changes in feeding time decreases heat stress which ultimately led to adecrease level of MDA in plasma. The present study recommends the following points: A) to minimize heat stress feeding times of dairy animals should be changed during hot weather conditions; B) day time feeding should be replaced with night time feeding and C) to improve production performance of dairy animals in summer feeding times should be changed.

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