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Plasma profile of hormones and energy metabolites in low and high producing periparturient Sahiwal cows during summer and winter season

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ABSTRACT

In order to study the plasma hormonal and metabolic changes in Zebu (Sahiwal) cattle during summer and winter season, five each of periparturient high and low yielding cows were selected from Livestock Research Centre, ICAR-NDRI, Karnal. Blood samples were collected on days -45, -30, -15 (prior to parturition), 0 (day of parturition) and 15, 30, 45 (postpartum) during both the seasons and plasma was separated. Plasma cortisol levels were significantly higher on day of parturition and during postpartum period in both the groups during both the seasons. Plasma insulin levels were significantly ($P<0.05$) lower during summer in high yielder group. On the day of calving, plasma T_3 and T_4 levels were significantly ($P<0.05$) lower in high yielders during both seasons. Plasma leptin levels were lower ($P<0.05$) on day of parturition in high yielders than low yielders during both seasons. Plasma glucose levels were significantly lower ($P<0.05$) on day of parturition and remained low during postpartum period in both groups during both seasons comparel to prepartum period. Plasma NEFA levels of both groups of cows were higher on the day of calving and remained higher during postpartum period over pre calving values. High yielder cows showed more deviation in stress markers *viz*. cortisol and NEFA than low yielder cows. Therefore, special attention should be given to feeding and management of the lactating animals (especially high yielding cows during summer) around parturition for sustained productivity and maintenance of homeostasis.

Key words: Cortisol, Energy metabolites, Leptin, Periparturient, Sahiwal.

INTRODUCTION

All organisms are occasionally or regularly exposed to extreme environmental conditions that challenge the physiological functioning at circulatory levels. When environmental effect becomes severe it is considered as stressful and require counter measures in order to maintain the body homeostasis. After parturition, the cow requires high amount of energy from body reserves, which leads to the loss of body weight. At the beginning of lactation, dairy cows have to cope with the high energy and protein demands for milk synthesis, but nutrient intake is low. Early lactating dairy cows are likely to undergo negative energy balance, metabolic and digestive disturbances (Nielsen, 1999). The continuous increase in milk production has created new challenges for high-producing dairy cows, especially during the transition period. Adaptation of glucose metabolism in early lactation leads to increased gluconeogenesis in the liver and to direct glucose into the mammary gland for lactose synthesis (Reynolds *et al.* 2003). Jacob *et al.* (2001) reported higher ($P< 0.01$) serum cortisol concentration in cows on the day of parturition than non-pregnant cows. The hormonal activity of the thyroid gland has an important role in the transition period for determining the cell metabolism intensity, metabolism of lipids and carbohydrates (Nikolic *et al.* 1997). A reduction in plasma leptin level by approximately 50% was found after calving and remained depressed during lactation, despite a gradual improvement in energy balance (Block *et al.,* 2001). Similarly, Liefers (2004) also observed high leptin concentrations during late pregnancy that declined rapidly at parturition. Although it is clear that a variety of endocrine or paracrine signals modify the heat stress response to stressors, but still the information on these hormones and energy metabolites especially in Sahiwal cows (tropical breed) around parturition during different season is scanty. Therefore, the present study was designed to document the levels of hormones and energy metabolites in periparturient Sahiwal cows and to establish possible relationships between these parameters during summer and winter season.

MATERIALS AND METHODS

Five each of periparturient low (<7 lit./day) and high(>10 lit./day) milk producing (based on previous lactation) Sahiwal cows were selected separately during summer and winter season from Livestock Research Centre (LRC) of ICAR-National Dairy Research Institute (NDRI), Karnal (India). Experimental animals were maintained as per the standard conditions of feeding (fodder and concentrate) and management at the LRC. Fresh tap water was available round the clock *ad lib* to all the animals for drinking.

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Blood samples were collected prior to parturition on day 45th, 30th and 15th and post partum on day 15th ,30th and 45th from experimental animals using heparinized vacutainer tubes during summer and winter seasons. Soon after collection, blood samples were brought to the laboratory in chilled icebox and plasma was separated by centrifuging at 1200×g at 4°C for 25 minutes. Plasma samples were stored at -20°C till estimation of biochemical and hormonal parameters. The environmental parameters *viz*. dry and wet bulb temperatures and relative humidity were recorded in the shed and temperature humidity index (THI) was calculated and presented in Table 1.

Glucose and cortisol levels were estimated in plasma samples using "Glucose assay kit" and cortisol EIA kit respectively procured from Cayman Company, USA. NEFA was estimated in plasma by "Bovine ELISA kit" from Cusabio Biotech Co., Ltd., China. Insulin was determined in plasma of cows by "Bovine ELISA kit" from Endocrine Technologies, USA. Tri iodothyronine (T_3) and Thyroxine (T_4) in blood plasma were quantified using Radioimmunology assay (RIA) kits supplied by Bhabha Atomic Research Centre (BARC), Leptin was estimated in plasma using "Bovine leptin kit" from Cusabio Biotech Co., Ltd.

Data of present study was normally distributed as checked by Shapiro-Wilk test in SAS system. Data was analyzed by analysis of variance using SAS software, version (9.1) of the SAS system for window, Copyright© (2011) SAS Institute Inc., Cary, NC, USA. Results were expressed as the means ±SEM. A difference with value $P<0.05$ was considered statistically significant. The correlation coefficient was also carried out among different parameters.

RESULTS AND DISCUSSION

Plasma hormones

Cortisol: During summer season, the mean levels of cortisol in blood plasma of high and low yielding Sahiwal cows were 3.27±0.23 and 2.61±0.75 ng/ml, respectively, on 45th day of prepartum. The respective values increased ($P< 0.05$) to 7.21±0.30 and 6.32±0.18 ng/ml on the day of calving. The cortisol did not reach to pre calving levels even after 45th day of parturition (Table 2). During winter season, the mean plasma cortisol levels of high and low yielding Sahiwal cows on 45th day of prepartum were 2.48±0.18 and 2.96±0.75 ng/ml, respectively. On the day of calving, the respective values increased ($P< 0.05$) to 5.37±0.20 and 5.76±0.58 ng/ml (Table 2).The levels of cortisol remained higher during summer compared to winter season in both the groups of cows. Further the levels of cortisol were higher in high yielders

Table 1: Environmental parameters recorded during the study

Months	Temperature (°C)				Relative Humidity	Temperature
	Maximum	Minimum	Dry bulb	Wet bulb	%	Humidity Index
Summer season						
April	34.0	17.0	33.2	20.5	24.0	79.31
May	38.4	23.2	37.5	24.5	32.0	85.24
June	35.0	24.9	34.1	26.3	52.0	84.15
July	33.1	26.3	31.7	27.1	71.0	82.95
August	32.1	25.7	31.6	27.6	73.	83.28
Average	34.5	23.4	33.6	25.2	50.4	82.90
Winter season						
October	31.9	17.1	31.6	22.2	43.0	79.35
November	28.1	12.8	27.8	19.0	40.0	74.35
December	21.8	6.9	21.5	14.8	46.0	66.72
January	17.3	6.2	16.7	12.6	61	61.67
February	20.7	6.9	20.5	14.3	45.0	65.60
Average	23.9	9.9	23.6	16.5	47.0	69.50

Table 2: Plasma cortisol levels (ng/ml) in periparturient Sahiwal cows during summer and winter season

Season	Summer		Winter	
Days	High Yielder	Low Yielder	High Yielder	Low Yielder
-45	3.27±0.23 ^{f}	2.61±0.75 ^{f}	2.48±0.18 ^{f}	2.96±0.75 ^{f}
-30	3.88±0.16 ^{ed}	2.86±0.39 ^{ed}	2.89±0.10 ^{ed}	2.64±0.42 ^{ed}
-15	4.65±0.16 ^{cd}	3.84±0.46 ^{cd}	2.94±0.29 ^{cd}	3.23±0.65 ^{cd}
0 (calving)	7.21±0.30 ^{a}	6.32±0.18 ^{a}	5.37±0.20 ^{a}	5.76±0.58 ^{a}
15	5.03±0.42 ^{b}	3.71±0.16 ^{b}	4.21±0.26 ^{b}	3.11±0.37 ^{b}
30	5.01±0.51 ^{cb}	3.47±0.52 ^{cb}	3.47±0.36 ^{cb}	3.30±0.52 ^{cb}
45	3.53±0.36 ^{ef}	3.02±0.22 ^{ef}	3.28±0.36 ^{ef}	2.80±0.29 ^{ef}

The values are the mean and SEM of five observations on five animals.
Value with different superscript in column differed significantly ($P<0.05$)

compared to low yielder groups (Table 2). Acute stressors activates hypothalamopituitary-adrenal axis, resulting in an increased cortisol levels (Dantzer and Mormede 1983) and involved in adaptation to short and long term heat stress (Beraidinell *et al.*1992). The plasma cortisol levels of high and low yielding Sahiwal cows during summer season increased by 120 and 142%, respectively on the day of calving from pre calving (45 day) values. During winter season, the respective values were increased by 116.5 and 94.5% on the day of calving from pre calving (45 day) values. Similar results i.e. lower plasma cortisol levels during prepartum than postpartum period in Holstein Frisian cows have been reported by Nikolic *et al.* (1997). The results of the present study are in agreement with Jacob *et al.* (2001) who reported significantly higher (P<0.01) serum cortisol concentration in cows on the day of parturition than non-pregnant cows. It has also been reported by Marai *et al.* (2010) that blood plasma cortisol levels increased significantly due to increase in ambient temperature from 17.5^{0}C to 37.1^{0}C. Cortisol levels increased in chronically heat-stressed cows, if heat stress intensiûes (Kadzere *et al.* 2002). Schaubli *et al.* (2008) stated that the bovine intercaruncular uterine wall expresses glucocorticoid receptors and exhibits a cell type-specific distribution pattern during late pregnancy and parturition.

Insulin: In high and low yielding Sahiwal cows, the mean plasma insulin levels during summer season were 1.26±0.02 and 1.23±0.03 ng/ml, respectively, on 45th day of prepartum. The respective values decreased to 0.72±0.01 and 0.85±0.03 ng/ml on the day of calving. During winter season, the mean plasma levels of insulin in high and low yielding Sahiwal cows were 1.51±0.08 and 1.53±0.10 ng/ml, respectively on 45th day of prepartum and the respective values decreased (p< 0.05) to 0.90±0.07 and 1.10±0.05 ng/ml on the day of calving (Table 3). In general the levels of insulin remained higher during winter compared to summer in both group of cows. During summer season, the plasma levels of insulin in high and low yielding Sahiwal cows decreased to 42.8 and 30.8%, respectively on the day of calving from pre calving (45 day) values. The respective values during winter season decreased to 40.3 and 28.1% on the day of calving from the pre calving (45 day) values. The results of the present study corroborates with those of Holtenius and Holtenius (2007) who observed the lowest insulin concentration during the first 3 weeks after parturition. They further reported the lower (P<0.01) blood insulin levels in lactating ewes compared to pregnant ewes. Similarly, greater (P<0.01) concentration of insulin was reported in non-lactating than in lactating ewes (Hatfield *et al.* 1999). The results of the present study are in general agreement with Khan and Ludri (2002) who reported low plasma insulin concentrations on the day of kidding in goats.

Tri-iodothyronine (T_3): During summer season, the mean levels of T_3 in blood plasma of high and low yielding Sahiwal cows were 0.91±0.05 and 0.90±0.03 ng/ml, respectively, on 45th day of prepartum. This plasma T_3 levels decreased to 0.54±0.02, and 0.64±0.01 ng/ml, respectively on the day of calving. During winter season, the mean T_3 levels of high and low yielding Sahiwal cows on 45th day of prepartum were 1.14±0.06 and 1.11±0.05 ng/m, respectively. The respective values decreased to 0.55±0.07 and 0.63±0.04 ng/ml on the day of calving. The T_3 levels remained higher during winter season compared to summer season irrespective of low or high yielding group of cows. Numerically the values of T_3 remained higher in high producing group compared to low producing during both the seasons (Table 4). The plasma T_3 levels of high and low yielding Sahiwal cows during summer season decreased by 40.6 and 28.8% respectively on the day of calving from the pre calving (45 day) values. During winter season, the pre calving values (45 day) of plasma T_3 levels of high and low yielding Sahiwal cows were also reduced to 51.7 and 43.2%, respectively on the day of calving. Significantly lower (P<0.01) levels of T_3 in the blood of lactating ewes compared to non-pregnant and pregnant ewes has been observed by Antunovic *et al.* (2011). Rasooli *et al.* (2004) also reported depressed thyroid activity in Holstein heifers during heat stress. Cold environment could be a stimulus to augment thyroid hormone secretion to increase basal metabolic rate in order to maintain body temperature, which accompanies with high level of blood metabolites (Kataria *et al.* 1993). Samanc *et al.* (2010) reported lower T_3 levels during the late dry (day 2) and early

Table 3: Plasma insulin levels (ng/ml) in periparturient Sahiwal cows during summer and winter season

Season	Summer		Winter	
Days	High Yielder	Low Yielder	High Yielder	Low Yielder
-45	1.26±0.02^{a}	1.23±0.03^{a}	1.51±0.08^{a}	1.53±0.10^{a}
-30	1.12±0.03^{a}	1.16±0.03^{a}	1.42±0.07^{a}	1.54±0.07^{a}
-15	0.98±0.03^{cb}	0.97±0.03^{cb}	1.40±0.05^{cb}	1.41±0.08^{cb}
0 (calving)	0.72±0.01^{e}	0.85±0.03^{e}	0.90±0.07^{e}	1.10±0.05^{e}
15	0.78±0.04^{ed}	0.90±0.02^{ed}	1.00±0.06^{ed}	1.31±0.12^{ed}
30	0.84±0.04^{cd}	0.96±0.01^{cd}	1.11±0.06^{cd}	1.30±0.07^{cd}
45	1.03±0.02^{b}	1.10±0.02^{b}	1.43±0.11^{b}	1.40±0.04^{b}

The values are the mean and SEM of five observations on five animals.
Value with different superscript in column differed significantly (P<0.05)

Table 4: Plasma triiodothyronine (T_3) levels (ng/ml) in periparturient Sahiwal cows during summer and winter season

Season	Summer		Winter	
Days	**High Yielder**	**Low Yielder**	**High Yielder**	**Low Yielder**
-45	0.91±0.05 [a]	0.90±0.03 [a]	1.14±0.06 [a]	1.11±0.05 [a]
-30	0.83±0.04 [b]	0.80±0.06 [b]	0.99±0.07 [b]	0.90±0.05 [b]
-15	0.72±0.04 [cb]	0.77±0.04 [cb]	0.73±0.03 [cb]	0.80±0.03 [cb]
0 (calving)	0.54±0.02 [e]	0.64±0.01 [e]	0.55±0.07 [e]	0.63±0.04 [e]
15	0.56±0.05 [ed]	0.70±0.05 [ed]	0.77±0.13 [ed]	0.73±0.07 [ed]
30	0.61±0.05 [cd]	0.69±0.05 [cd]	0.80±0.08 [cd]	0.70±0.06 [cd]
45	0.62±0.04 [cd]	0.70±0.05 [cd]	0.74±0.06 [cd]	0.79±0.04 [cd]

The values are the mean and SEM of five observations on five animals.
Value with different superscript in column differed significantly (P<0.05)

lactation (day12) periods in cows with severe fatty liver. Present findings are also in accordance with Horowitz (2001) who stated that heat acclimation decreases endogenous levels of thyroid hormones (in an attempt to reduce endogenous heat production) and those mammals adapted to warmer climates follow this pattern.

Thyroxine (T_4): On 45th day of prepartum, the mean levels of T_4 in blood plasma of high and low yielding Sahiwal cows during summer season were 27.1±1.5 and 21.4±2.6 ng/ml, respectively. The respective values decreased to 15.5±0.9 and 13.9±0.7 ng/ml on the day of calving. During winter season, the mean T_4 levels of high and low yielding Sahiwal cows on 45th day of prepartum were 23.0±2.1 and 30.1±2.6 ng/ml, respectively. On the day of calving, these values reduced to 14.4±1.8 and 15.6±1.5 ng/ml, respectively (Table 5). Almost similar levels of T_4 were attained by both the groups of cows after 45th day of post partum as recorded 45th day pre partum. During summer season, the plasma T_4 levels of high and low yielding Sahiwal cows decreased to 42.8 and 35%, respectively on the day of calving from the pre calving values (45 day). During winter season, the respective values decreased by 37.3 and 48.1% on the day of calving from the pre calving values (45 day). Sinka *et al.* (2008) reported significant drop in T_4 levels during early lactation and remained low up to peak lactation. T_4 levels were found to be low during the early lactation period in cows with severe fatty liver (Stojic *et al.* 2001). The lowest levels of plasma T_4 on the day of kidding compared to postpartum period have been reported by Khan and Ludri (2002). The results of the present study are in accordance to those of Rasooli *et al.* (2004) who observed that cold environment may be a stimulus to increase the thyrotrophic hormone output thereby resulting in a higher concentration of thyroid hormone in serum.

Leptin: The mean levels of leptin in blood plasma of high and low yielding Sahiwal cows during summer season were 5.81±0.14 and 5.54±0.10 ng/ml, respectively on 45th day of prepartum. The lowest values ~~i.e. to~~ 3.85±0.05 and 4.42±0.09 ng/ml in high and low yielding cows respectively reached on the 15th day (postpartum) of calving. During winter season, the mean plasma leptin levels of high and low yielding Sahiwal cows were 6.12±0.10 and 5.90±0.21 ng/ml, respectively, on 45th day of prepartum. The respective values reduced (P< 0.05) to minimum 3.98±0.08 and 4.62±0.25 ng/ml in high and low producing cows on the day of calving (Table 6). The plasma levels of leptin in high and low yielding Sahiwal cows decreased to 33.7 and 20.2%, respectively on 15th day (postpartum) of calving from the pre calving (45th day) values during summer season. During winter season, the respective values of plasma leptin decreased by 34.9 and 21.6% on the day of calving. The result of the present study is in accordance with those of Sauerwein *et al.* (2004) who reported a decline in plasma leptin levels towards parturition and the decline is probably caused by the decline in adiposity and insulin concentrations and after parturition its level remained low up to 30 days in ruminants. The plasma leptin

Table 5: Plasma thyroxine (T_4) levels (ng/ml) in periparturient Sahiwal cows during summer and winter

Season	Summer		Winter	
Days	**High Yielder**	**Low Yielder**	**High Yielder**	**Low Yielder**
-45	27.1±1.5[a]	21.4±2.6 [a]	23.0±2.1 [a]	30.1±2.6 [a]
-30	23.5±1.6[ba]	18.4±1.9 [ba]	23.7±1.6 [ba]	27.7±1.6 [ba]
-15	21.9±1.8[c]	17.6±0.5 [c]	20.2±1.8 [c]	20.2±2.2 [c]
0 (calving)	15.5±0.9[d]	13.9±0.7 [d]	14.4±1.8 [d]	15.6±1.5 [d]
15	16.2±0.6 [d]	15.1±1.4 [d]	18.3±1.8 [d]	18.7±1.6 [d]
30	19.2±0.8[c]	17.8±0.6 [c]	22.8±1.3 [c]	20.3±0.6 [c]
45	22.0±0.8[b]	19.4±1.4 [b]	23.3±0.9 [b]	24.0±1.0 [b]

The values are the mean and SEM of five observations on five animals.
Value with different superscript in column differed significantly (P<0.05)

Table 6: Plasma leptin levels (ng/ml) in periparturient Sahiwal cows during summer and winter season

Season	Summer		Winter	
Days	High Yielder	Low Yielder	High Yielder	Low Yielder
-45	5.81±0.14[a]	5.54±0.10[a]	6.12±0.10[a]	5.90±0.21[a]
-30	5.72±0.10[a]	5.66±0.12[a]	6.08±0.16[a]	6.11±0.18[a]
-15	5.53±0.16[a]	5.67±0.10[a]	5.25±0.30[a]	6.18±0.17[a]
0 (calving)	3.96±0.09[b]	4.47±0.26[b]	3.98±0.08[b]	4.62±0.25[b]
15	3.85±0.05[b]	4.42±0.09[b]	4.25±0.31[b]	4.74±0.03[b]
30	4.20±0.11[b]	4.88±0.24[b]	4.29±0.08[b]	4.78±0.18[b]
45	4.32±0.06[b]	5.13±0.18[b]	4.20±0.15[b]	4.78±0.11[b]

The values are the mean and SEM of five observations on five animals.
Value with different superscript in column differed significantly (P<0.05)

concentration was reduced by 50% after parturition and remained depressed during early lactation (Nikolic *et al.* 1997). Leptin concentrations were higher during pregnancy because of the high-energy intake, necessary for the coming lactation period (Garcia *et al.* 2000).

Leptin showed positive correlation with insulin and negative correlation with cortisol and NEFA. Chelikani *et al.* (2008) also reported a positive correlation of leptin with insulin and glucose and negative correlation of leptin with NEFA and cortisol in control and treatment group of cows around parturition.

Energy metabolites

Blood glucose: During summer season, the mean plasma glucose levels of high and low yielding Sahiwal cows varied from 41.1±0.87 mg/dl (0 day) to 54.5±1.74 mg/dl on 45th day of pre partum. Similar trend was observed in low yielding group (Table 7). During winter season, the mean levels of glucose in plasma of high and low yielding Sahiwal cows on 45th day of prepartum were 55.4±1.88 and 55.6±0.35 mg/dl, respectively. These respective glucose values reduced (P< 0.05) to 42.2±0.83 and 46.3±1.74 mg/dl on the day of calving (Table 7). The levels of plasma glucose start increasing from one month after calving in the both the groups and during both the seasons. During summer season, the pre calving (45th day) values of plasma glucose levels of high and low yielding Sahiwal cows decreased by 25 and 20%, respectively on the day of calving. The respective values were reduced by 22 and 17% during winter season. During the present investigation the blood glucose levels were well within the normal range (Table 7). Several authors reported a normal range of blood glucose from 45.81 to 70.77 mg/dl in ruminant animals. The results of the present study corroborates with those of Holtenius and Holtenius (2007) who reported lowest glucose during the first three weeks after calving and thereafter fluctuations were minimum. The lower levels of blood glucose levels during the present study may be probably as a result of the heat-induced increase in circulating basal insulin concentration. Similar pattern i.e. significantly (P<0.05) lower (3.22±0.21 mmol.l^{-1}) blood glucose level was reported at the start of lactation compared to middle of lactation (3.69±0.08 mmol.l^{-1}) and at the dry period (3.74±0.21 mmol.l^{-1}) in dairy cows (Filipejova and Kovacik, 2009).

Non-esterified fatty acids (NEFA): The mean levels of NEFA in blood plasma of high and low yielding Sahiwal cows varied from 149.5±10.1 to 308±9.6 uM/L and 128.1±6.1 to 286.5 ± 5.1 µM/L respectively during summer season on 45th day of prepartum (Table 8). During winter season also, NEFA followed the similar pattern i.e. lower levels during prepartum and higher levels on the day of parturition and post partum in both the groups. The NEFA levels remained higher in high producing cows than low producing irrespective of seasons (Table 8) The plasma NEFA levels of high and low yielding Sahiwal cows during summer season was increased from pre calving values (45 day) by 94.5 and 107.9 %, respectively on the day of calving and postpartum period (15th day). Similarly, during winter

Table 7: Plasma glucose levels (mg/dl) in periparturient Sahiwal cows during summer and winter season

Season	Summer		Winter	
Days	High Yielder	Low Yielder	High Yielder	Low Yielder
-45	54.5±1.74[a]	57.1±1.07[a]	55.4±1.88[a]	55.6±0.35[a]
-30	53.4±1.61[a]	54.5±1.34[b]	54.1±1.07[a]	54.3±1.26[a]
-15	49.2±0.92[b]	52.8±1.43[b]	50.3±0.81[b]	52.3±1.13[b]
0 (calving)	41.1±0.87[c]	45.9±1.83[c]	42.2±0.83[c]	46.3±1.74[c]
15	41.1±1.27[cd]	47.2±1.33[cd]	46.1±1.01[cd]	46.7±1.05[cd]
30	43.4±2.32[cd]	51.6±1.72[cd]	49.3±3.02[cd]	51.9±1.19[cd]
45	47.4±2.05[cb]	53.1±1.71[cb]	50.3±2.23[cb]	51.6±1.64[cb]

The values are the mean and SEM of five observations on five animals.
Value with different superscript in column differed significantly (P<0.05)

Table 8: Plasma Non- esterified fatty acid (NEFA) levels (μM/L) in periparturient Sahiwal cows during summer and winter season

Season	Summer		Winter	
Days	High Yielder	Low Yielder	High Yielder	Low Yielder
-45	149.5±10.1 [e]	128.1±6.1 [e]	107.5±9.0 [e]	102.3±10.1 [e]
-30	167.1±8.7 [e]	130.1±2.8 [e]	105.9±9.0 [e]	108.9±10.1 [e]
-15	193.1±8.6[d]	185.2±12.7 [d]	131.7±8.9 [d]	114.9±9.3 [d]
0 (calving)	290.9±2.9 [a]	286.5±5.1 [a]	261.4±8.5 [a]	212.7±18.2 [a]
15	308.0±9.6 [a]	284.1±9.1 [a]	256.5±17.3 [a]	222.0±23.3 [a]
30	263.8±21.3[b]	198.9±13.7 [b]	216.6±19.2 [b]	159.2±12.9 [b]
45	206.2±8.7[c]	151.3±12.9 [c]	198.8±7.5 [c]	149.4±18.2 [c]

The values are the mean and SEM of five observations on five animals.
Value with different superscript in column differed significantly (P<0.05)

season the respective values were increased by143 and 117.3% from pre calving values. The findings of the present study are in accordance with Bernabucci *et al*. (2005) who reported cows with greater BCS losses had higher NEFA levels and are sensitive to oxidative stress. Higher plasma NEFA (P<0.05) was observed after 14 days from calving compared to the values before parturition (Catalani *et al.* 2010). The dramatic increase in energy requirements needed for the onset of lactation in transition cows often accompanied by a decrease in voluntary dry matter intake that causes a negative energy balance. Energy requirements that cannot be met by the diet must then rely on tissue energy reserves. Therefore, negative energy balance during the periparturient period causes mobilization of fat from tissue stores and the release of nonesterified fatty acids (NEFA) into the blood stream. Similarly, Holtenius and Holtenius (2007) reported highest levels of the plasma free fatty within three weeks after calving and decreased gradually thereafter. The results of the present study are in accordance to those of Nikolic *et al.* (1997) and Karapehlivan *et al.* (2007) who reported higher NEFA levels during early lactation. Plasma NEFA had positive correlation with cortisol and negative correlation (P<0.05) with glucose, insulin, T_3, T_4 and leptin and THI.

CONCLUSIONS

Based on the results obtained during the present study, it can be stated clearly that the energy metabolites and hormonal levels deviated significantly on the day of parturition and not attained the same levels (pre calving) even after 45 days of post partum in Sahiwal cows. Further, the magnitude of deviation in these parameters on the day of parturition and post partum period than prepartum was more severe in high yielder group and during summer seasons. Therefore, special attention should be given to feeding and management the lactating animals (especially high yielding cows during summer) around parturition for sustained productivity and maintenance of homeostasis.

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