The Journal of Animal & Plant Sciences, 26(2): 2016, Page: 366-372
ISSN: 1018-7081

APOLIPOPROTEIN B100 AND SOME LIPID COMPONENTS DURING TRANSITION PERIOD IN COWS UNDER HOT AND THERMONEUTRAL ENVIRONMENTAL CONDITIONS

Fatma Edrees Ibrahim Teama

Biological Applications Department, Radioisotopes Applications Division,
Nuclear Research Center, Atomic Energy Authority, Inshas, Cairo, Egypt, P.O. 13759.
Corresponding Author E-mail: fatema58us@yahoo.com

ABSTRACT

Study conducted to investigate the biochemical changes in the blood (apolipoprotein B100 and some lipid components) of crossbred cows (Brown Swiss × Baladi) during the transition period under hot and thermoneutral environments. Twelve cows were divided in to two equal groups; hot and thermoneutral environment. Blood samples were collected and ApoB100, total protein, glucose, cholesterol, triglycerides, non-esterified fatty acid (NEFA), β-hydroxybutyric acid (BHBA), and thyroid hormones (T_3 and T_4) were estimated. Temperature-humidity index as well as milk yield and composition were also recorded in each season. The levels of almost all of studied parameters including ApoB100, total protein, cholesterol, triglycerides, glucose, and thyroid hormones detected in the thermoneutral group significantly exceeded those detected in the other group (hot environment). On the other hand, the levels of NEFA and BHBA detected in the blood of the cows in the hot environment group significantly increased than those of the thermoneutral group. In addition, the blood contents of the two lipid components (NEFA and BHBA) were negatively correlated with the level of ApoB100. Milk yield and composition were also among the parameters affected negatively by heat stress, and were positively correlated with the level of ApoB100 postpartum. The studied parameters may present a valuable assess for the prediction of the disturbance during this critical period.

Keywords: ApoB100; hot environment; transition period; crossbred cows.

INTRODUCTION

The Egyptian climate is characterized by high ambient temperatures, relative humidity, and radiant energy during the summer season. This climate impairs the potential of farm animals (Kamal, 1976), decreasing their milk production and reproductive performance (Sharma *et al*., 1988). Changes in hormonal levels, especially of thyroid hormones, which play an important role in animal adaptation, have been reported in dairy animals under heat stress (Marai and Habeeb, 2010; Haque *et al*., 2012).The main physiological responses that appear in dairy cows as a result of exposure to hot conditions include sweating, high respiratory rate, reduced metabolic rate, decreased dry matter intake, and altered water metabolism. All of these changes may have a negative impact on the production as well as reproduction of dairy cows (West, 1999). It is well known that the transition period of dairy cows (3 to 2 wk prepartum until 2 to 3 wk postpartum) is a very critical period during which tremendous physiological, metabolic, and nutritional changes take place. Previous studies have shown that there are changes in the number of physiological parameters that occur in the transition of cows that modify their metabolism, including hormonal changes (Grummer, 1995), especially under heat stress. The rapidly increasing requirements of the fetus as well as the development of the mammary glands for the synthesis of milk components are also among the main causes of such changes in this period (Bell, 1995). During the periparturient period, requirements of energy and proteins increase dramatically to support the onset of milk production. This period is also associated with a reduction of dry matter intake (DMI). These energy requirements are met, in part, by mobilization of fatty acids from fat depots, which increases the amount of circulating non-esterified fatty acids (NEFA). It has been found that NEFA in liver extracts in proportion to their concentration in the bloodstream may lead to hepatic lipidosis (Grummer, 1993).This could explain reduced capacity of the liver to synthesize apolipoprotein B100 (ApoB100). Apolipoprotein B100 is the core component of the very low-density lipoproteins (VLDL), and is also necessary to stabilize the VLDL particles. Reduced synthesis and secretion of ApoB100 are usually associated with liver fat accumulation and impaired function (Itoh *et al*., 1997).Therefore, this study was designed to investigate the changes occurring in some biochemical components including ApoB100 and levels of T_3 and T_4 hormones in dairy cows during their transition period under a thermoneutral or hot environment.

MATERIALS AND METHODS

The current study was conducted in Cairo, Egypt. Twelve heavy pregnant crossbred cows (Brown Swiss × Baladi) at the third parity were utilized in the study. During the transition period, these animals were divided into two groups thermoneutral (28ºC and 40 RH % during May) and a hot environment (39ºC and 55 RH% during July) according to the season. Each group was comprised of six animals.

Cows were housed in a free-stall barn with shad during day and night. Animals were maintained on a similar feeding program. Cows were offered daily a concentrate basal diet at 2% of their average body weight. The concentrate consisted of 27% undecorticated cottonseed meal, 35% wheat bran, 30% yellow corn, 5% soybean, 1.5% limestone, and 1.5% sodium chloride. Rice straw was available at all times. Mineral-vitamin blocks were supplied (Tithebarn limited, Winsford, Cheshire CW7 3PG.U.K.). All animals had free access to tap water during the experimental period. Rectal temperature and respiratory rate were recorded. Climatic data were recorded during the experiment period, in which air temperature (°C) and relative humidity (%) values were recorded using a thermo-hygrometer and were later utilized to calculate the temperature-humidity index (THI) according to Armstrong (1994).

Blood samples were collected from each animal before the morning feeding by jugular venipuncture in vacutainer tubes. Plasma glucose level was determined enzymatically using commercial kits (Biodiagnostic, Egypt). Serum total protein, total cholesterol, and triglycerides were determined spectrophotometrically using a commercial kit (Biodiagnostic, Egypt).β-hydroxybutyrate (BHBA) and non-esterified fatty acids (NEFA) were also determined calorimetrically using commercial kits (Randox Laboratories). The serum level for both thyroxin (T_4) and triiodothyronine (T_3) hormones was estimated by the radioimmunoassay (RIA) technique using solid-phase coated tubes kits labeled with (I^{125}) (Diagnostic Systems Laboratories, Inc. Webster, Texas, USA). Apolipoprotein B100 was determined using a bovine ApoB100 ELISA kit (Biomedical Bmassay, China).

Cows were milked twice daily one week after delivery. The milk yield of the individual cows was recorded at each milking session on all test days. A composite sample from the two milking sessions (200 mL) from days 7, 15, and 20 was analyzed for the contents of protein, fat, and solids non-fat (SNF), and milk yield was recorded. Total solids, fat, and protein contents of the milk were estimated according to Gupta *et al* (1992).The solids non-fat content was calculated by subtracting the fat content from the total solids content.

Data were analyzed statistically using the General Linear Model procedure of SPSS (v.16, Inc., Chicago, IL, USA). A two-way ANOVA was run, according to the following model:

$$Y_{ij} = \mu + E_i + P_j + (E\times P)_{ij} + e_{ijk},$$

Where; μ= overall mean; E_i = fixed effect of environmental conditions (thermoneutral and hot environments); P = fixed effect of transition period (prepartum and postpartum); E×P= interactions between the two factors; and e_{ijk} = random error. Significances for differences in the results were verified by Duncan's test (1955). Differences between the mean values of milk seasons were tested using unpaired "t" test according to Graph Pad Prism (v.5). Pearson's simple correlation analyses were performed.

RESULTS

The temperature-humidity index is usually used as an indicator of thermal stress in cows (Armstrong, 1994). The mean (± standard error) THI recorded in this study for the thermoneutral and hot environments were 70±1 and 91±1, respectively. These data indicate that the cows were exposed to a higher thermal load in the hot environment. The recorded rectal temperature was higher in the hot environment compared with the thermoneutral conditions, and mean values were 41.1±0.1°C and 38.1±0.2°C, respectively. Additionally, the respiratory rate was higher under hot conditions (88±3 breaths/min) compared with the thermoneutral conditions (56±1 breaths/min).

The present study analyzed the effect of hot climate on ApoB100, plasma glucose, total protein, total cholesterol and triglycerides, β-hydroxybutyrate (βHBA), non-esterified fatty acids (NEFA), and thyroid hormones (T_4 and T_3) during the transition period of crossbred cows. A highly significant decrease was observed in the levels of glucose, total protein, and ApoB100 in the hot environment group compared with the thermoneutral group (Table 1). The same results were observed post-calving compared with pre-calving. A significant reduction in the level of total protein was recorded when the combined effect of heat stress and transition period was considered compared with the effect of either heat stress or transition period independently (E×P statistical test).

Serum cholesterol and triglycerides levels were also affected by hot conditions, with a significant decline recorded in the levels of these two parameters in the hot condition group compared with the thermoneutral group ($P<0.0001$). This was also the case when the cholesterol and triglycerides levels post-calving and pre-calving were compared, as the same decline in their levels was detected post-calving (Table 2). However, the combined effect of heat stress and transition period failed to achieve any significant change on either lipid.

The serum levels of NEFA and BHBA were highly elevated in hot conditions as well as in the post-calving period compared with the thermoneutral environment and pre-calving period, showing statistical significance (Table 2). A significant combined effect of both factors (hot climate and transition period) was also observed on the NEFA and BHBA levels (Table 2).

Table 1. Apolipoprotein B100 (ApoB100), total protein, and glucose levels in crossbred cows in hot and thermoneutral environments during the transition period (Mean ± SE)

Items	ApoB100(µg/mL)	Total protein (g/dL)	Glucose (mg/dL)
Environmental condition (E)			
Thermoneutral	138.40A±7.8	6.73A±0.1	60.63A±0.59
Hot	109.77B±8.0	5.92B±0.07	56.75B±0..64
P-value	0.01	0.0001	0.0001
Transition period (P)			
Pre-calving	158.31A±7.89	6.80A±0.2	65.13A±0.58
Post-calving	89.85B±1.81	5.85B±0.3	52.25B±0.64
P-value	0.0001	0.0001	0.0001
Interactions (E × P)			
Thermoneutral			
Pre-calving	175.71±13.9	7.31a±0.1	67.79a±1.1
Post-calving	101.03±8.5	6.15b±0.13	53.46c±0.89
Hot			
Pre-calving	140.89±11.4	6.29b±0.11	62.46b±0.9
Post-calving	78.66±4.8	5.56c±0.10	51.0d±0.87
P-value	0.050	0.050	0.010

SE- standard error.
Means followed by different letters (A and B; or a, b, c, and d) in the same column within each classification are significantly different (P<0.05 or P<0.01), respectively.

Table 2. Cholesterol, triglycerides, NEFA, and βHBA levels in crossbred cows in hot and thermoneutral environments during the transition period (Mean ± SE)

Item	Cholesterol (mg/dL)	Triglycerides (mg/dL)	NEFA (µmol/L)	B-HBA (µmol/L)
Environmental condition (E)				
Thermoneutral	110.03A±2.0	118.94A±0.9	62.87B±16.40	391.27B±15.8
Hot	87.42B±1.97	107.62B±1.1	449.75A±15.98	563.64A±16.5
P-value	0.0001	0.0001	0.0001	0.0001
Transition period (P)				
Pre-calving	113.81A±1.8	117.21A±1.11	300.74B±15.9	414.23B±16.48
Post-calving	83.67B±1.79	109.35B±0.97	511.9A±16.41	540.68A±16.50
P-value	0.0001	0.0001	0.0001	0.0001
Interactions(E × P)				
Thermoneutral				
Pre-calving	123.57±2.7	123.27±1.3	287.24c±6.4	365.85d±23.31
Post-calving	96.51±2.6	114.61±1.5	438.5b±22.8	416.69c±23.32
Hot				
Pre-calving	104.02±2.8	111.13±1.28	314.23c±23.25	462.61b±13.6
Post-calving	70.83±3.1	104.10±1.5	585.2b±22.97	664.67a±15.2
P-value	0.020	0.050	0.010	0.010

NEFA - non-esterified fatty acid; βHBA - β-hydroxybutyric acid; SE - standard error.
Means followed by different letters (A and B; or a, b, c, and d) in the same column within each classification are significantly different (P<0.05 or P<0.01), respectively.

Serum levels of triiodothyronine (T_3) and thyroxin (T_4) in cows were also affected significantly by environmental conditions and transition period. In fact, the thyroid hormones levels were markedly reduced in the hot environment and in the post-calving period compared with thermoneutral conditions and pre-calving period, respectively (Table3). In addition, heat stress and transition period together affected the concentration of thyroid hormones even more than any of them did independently ($P<0.01$ and $P< 0.05$ for T_3 and T_4, respectively) (Table3). Milk composition data showed a highly significant decrease in the values of each component measured in the hot environment group compared with the thermoneutral conditions (Table 4).

The correlation between the level of ApoB100 and the levels of other parameters was tested using Pearson's simple correlation. There was a positive correlation ($P<0.01$) between the serum level of ApoB100 and that of glucose, total protein, cholesterol, T_3, T_4 and triglycerides, whose values were 0.824, 0.698, 0.768, 0.608, 0.842, and 0.262, respectively. In contrast, the ApoB100 level was negatively correlated with the NEFA and βHBA levels for both seasonal variation ($P<0.05$) and transition period ($P<0.01$). The ApoB100 level was also positively correlated with milk composition including milk fat, milk protein, and solid non fat under thermoneutral conditions ($P<0.01$), whose values were 0.719, 0.878, and 0.958, respectively. In the case of hot environment, the level of ApoB100 was negatively correlated with milk composition (fat and total solids, $P<0.01$), whose values were -0.998 and -0.628, respectively. These data indicate dramatic changes in biochemical parameters and milk composition as well as a negative correlation between the levels of ApoB100 and NEFA observed in the blood of crossbred cows during their transition period in two environmental groups.

Table 3. T_3 and T_4 hormones in crossbred cows under hot and thermoneutral environments during the transition period (Mean ± SE)

Item	T_3 (nmol/L)	T_4 (nmol/L)
Environmental condition (E)		
Thermoneutral	2.64A±0.11	60.81A±0.71
Hot	1.68B±0.08	56.57B±0.68
P-value	0.0001	0.0001
Transition period (P)		
Pre-calving	2.36A±0.08	61.28A±1.21
Post-calving	1.96B±0.09	56.10B±0.68
P-value	0.001	0.0001
Interactions (E × P)		
Thermoneutral		
Pre-calving	2.68a±0.11	64.51a±1.11
Post-calving	2.59a±0.10	57.11b±0.96
Hot		
Pre-calving	2.03b±0.14	58.1b±1.12
Post-calving	1.32c±0.11	55.1b±0.87
P-value	0.01	0.05

SE - standard error.
Means followed by different letters (A and B; or a, b, c, and d) in the same column within each classification are significantly different ($P<0.05$ or $P<0.01$), respectively.

Table 4. Composition of milk from crossbred cows in hot and thermoneutral environments (Mean ± SE)

Item / Milk composition	Milk yield, kg	Fat, %	Solids non-fat,%	Total solids, %	Protein, %
Thermoneutral	8.09±0.29	3.90±0.13	8.82±0.11	12.72±0.19	3.16±0.08
Hot	6.94±0.13	3.35±0.12	8.32±0.100	11.67±0.12	2.77±0.11
P-value	0.005	0.011	0.0085	0.0009	0.021

SE - standard error. Significant at $P<0.05$.

DISCUSSION

During the transitional period, in hot climatic environments such as the summer season in Egypt, dairy cows suffer from metabolic and health problems that may reflect in their production and reproduction performance.

Apo-lipoprotein B100 is one of the two major Apo-protein fractions in bovine lipoprotein. It plays an important role for stabilizing the VLDL by binding to triglycerides (Gibbons, 1990), and is responsible for exporting triglycerides from the liver to extra-hepatic tissues (Shin and Norio, 1997). During the postpartum period of cows, a marked decrease in plasma ApoB100 level was recorded in the present study compared with the prepartum period, which is consistent with previous research (Myamoto *et al*., 2006; Bernabucci *et al*., 2010; Basirico *et al*., 2011). This decrease in the ApoB100 level may be attributed to mobilization of fats and muscles by dairy cows during early postpartum, as described by Wathes *et al*. (2007). This mobilization generates a negative feedback effect that down-regulated the ApooB100 expression by liver and consequently its secretion. A reduction of the level of ApoB100 was also detected during the hot period as compared with thermoneutral period in the current study. This reduction in the liver ApoB100 during the hot season, therefore, provides additional support for the theory that hot environments down-regulate the expression of the ApoB100 gene and hence its protein product in transition dairy cows (Bernabucci *et al*., 2004; Myamoto *et al*., 2006).

In our study, the low level of total protein recorded in the postpartum period of cows under hot environment reflects the high maternal requirement of proteins needed for production of milk and immunoglobulins (Roubies *et al*., 2006; Mohri *et al*., 2007). It was also reported that these low levels of plasma proteins observed during the first days after calving could be due to the muscular development of the fetus (Karapehlivan *et al*, 2007) and the high energy needed for milk synthesis in animals during early lactation (Yokus *et al*., 2006).

Glucose levels were also reduced in the postpartum period in the hot environment according to the present study, which is consistent with the previous work by our group (Teama and Gad, 2014). This reduction in glucose level may attributed to the mobilization of glucose into fetal circulation through an active transport process, as the energy requirement in late pregnancy is mostly met by placental uptake of maternal glucose (Bell, 1995). Tainurier *et al*. (1984) attributed this decrease in glucose level to the higher energy requirement for fetal metabolism and/or progressive appearance of fetal insulin, which was found to pass into the maternal blood. However, this finding contradicts the early work by Tainurier *et al*. (1984), who demonstrated an increase in glucose level in other ruminants during this period. The authors attributed this increase to the secretion of glucocorticoids and epinephrine, which stimulate glycogenolysis in the liver.

Cholesterol and triglycerides are two main parameters of lipid profile in blood. In dairy cows, during the transition period, the cholesterol and triglycerides levels were reduced according to previous studies. This decrease can be explained by the increased demand for the mechanisms involved in milking (Krajnicakova *et al*., 2003). The regulation of both lipolysis and lipogenesis processes was found to increase the lipid reserve during pregnancy that is utilized following parturition and for the start of lactation (Roche *et al*., 2009). A significant reduction in the levels of cholesterol and triglycerides was found in this study. This reduction was observed during the transition period in the cows reared in the hot environment. This decrease may be attributed to physiological mode or negative energy balance in cows as a result of the decreased dry matter intake and changes in the liver activity that characterize the transition period in dairy cows (Bobe *et al*., 2003). It is well known that cholesterol is a constituent of several lipoproteins and its reduction during this period could reflect the possible alteration of liver synthesis and presence of fatty liver (Artekar and Desai, 2003). It was also shown that the low cholesterol levels reported may be attributable to its role in ovarian steroidogenesis processes (Piccione, 2009).

In the hot environment, a significant decrease in the levels of cholesterol and triglycerides was also detected in the present study. This decline in their levels is probably related to metabolic disorders associated with the exposure of cows to a hot environment. This heat exposure may accelerate body fat catabolism and lipid mobilization (Abeni *et al*., 2007). A reduction in liver activity reported during heat exposure (Ronchi *et al*., 1999) could also explain the lower cholesterol level during summer (hot environment). In addition, the increase in total body water content and the consequent dilution of the blood components (hemodilution) may provide another explanation for the reduction of cholesterol and triglycerides levels (Habeeb et. al., 1992).

The level of NEFA was also estimated in the current study. During the transition period, the NEFA level was higher postpartum than prepartum. It has been reported that there is an increase in NEFA level around the time of calving (Teama and Gad, 2014) due to the increased requirement of nutrients for the growing fetus. Therefore, body fat reserves may be mobilized and, as a consequence, the plasma NEFA content increases near the end of gestation (Bell *et al*., 1995).

With the increased energy requirements for both maintenance functions and lactogenesis, the ketone β-HBA tends to increase following the day of calving until about 3 week (wk) postpartum. Thereafter, levels begin to decline as the cow recovers from hypophagia and the negative energy balance (Vasquez-Anon *et al*., 1994). An increased concentration of β-HBA is considered as a sign of negative energy balance (Re´mond *et al*., 1991), which revealed that the cows may have been in greater negative energy balance during early lactation.

The hormonal activity of the thyroid gland has an important role in the animals' adaptation to environmental changes. In our data, the low level of thyroid hormones (T3, T4) during the transition period of crossbred cows was significant in the hot environment, postpartum and interaction between (E*P). This decline in their levels may be attributed to the metabolic disorder during this period under hot environment. In early postpartum, cows are in the state of metabolic and heat stress, in order to meet the increased energy of the mammary gland and the adjustment of the neuroendocrine system of cows to new metabolic needs of their body (Nikolić *et al*., 1997). The decrease in thyroid hormones may support the mammary gland in partitioning of nutrients between mammary and non-mammary tissue. This decrease also attenuates the adverse effect of heat stress and nutrient deficiency in body tissue at the onset of lactation. This more dramatic decline in hormones levels could influence the mammary tissue development and postpartum milk yield (Collier *et al*., 1982). Under circumstances of negative energy balance and high lipid mobilization, the blood level of thyroid hormones is significantly lower in the transitional period, with a marked decline in the blood triiodothyronine (T_3) level shortly before and after calving (Nikolić *et al*., 1997).

Milk yield and the proportion of milk components including fat, protein, total solids, and solids non-fat were among the factors that were significantly reduced under heat stress according to the present study. The variation in composition of milk from the cows was found to be dependent upon multiple factors including seasonal variation and udder health. Heat stress is generally associated with a decline in the production performance of dairy cows. Indeed, during heat stress circumstances, animals activate thermo-regulation mechanisms to avoid hyperthermia and to maintain their vital functions (Nardone *et al*., 2006) that could affect their performance. Our data are therefore consistent with previous studies as the milk yield and other fractions were high during thermoneutral environment compared with the hot environment (Haenlein, 2003). For instance, the percentages of fat, protein, and all nitrogen fractions were influenced by the seasonal variations according to an early study by Lacroix *et al*. (1996). In another study, Casati *et al*. (1998) demonstrated marked changes in milk yield and composition induced by changing the light-to-dark ratio. In addition, a reduction in fat and protein contents of milk has also been detected in response to a high light-to-dark ratio, which could be attributed to increased secretion of prolactin, whose plasma level is higher in the summer than in the winter (Tucker, 1989).

Conclusions: The biochemical changes observed in dairy cows during the transitional period under hot environmental conditions provide further evidence for the adverse effect on their performance. The studied parameters may present a valuable assess for the prediction of disturbances during this critical period.

Acknowledgments: The authors would like to thank Prof. Dr. Kamel El-Masry, (Animal Physiology; Nuclear Research Center), for providing animals from the Experimental Farm Project; Dr. Mostafa Abaas, (Assistant Lecturer) for his help in collecting samples; and Dr. Hamdy Abouzid for revising the manuscript.

REFERENCES

Abeni, F., L. Calamari and L. Stefanini (2007). Metabolic conditions of lactating Frisian cows during the hot season in the Po Valley. 1. Blood indicators of heat stress. International J. Biomet. 52: 87-96.

Armstrong, D. V. (1994). Heat stress interaction with shade and cooling. J. Dairy Sci. 77: 2044-2050.

Artekar, T. M. and H. G., Desai (2003). Fatty liver with elevated transaminase levels due to heterozygous apolipoprotein B deficiency. Trop. Gastro. 24:35–36.

Basiricò, L., P. Morera, N. Lacetera, B. Ronchi, A. Nardone and U. Bernabucci (2011). Down-regulation of hepatic ApoB100 expression during hot season in transition dairy cows. Livestock Sci. 137: 49-57.

Bell, A. W. (1995). Regulation of organic nutrient metabolism during transition from late pregnancy to early lactation. J. Anim. Sci. 73:2804-2819.

Bernabucci**,** U., B. Ronchi, L. Basiricò, D. Pirazzi, F. Rueca, N. Lacetera and A. Nardone (2004). Abundance of mRNA of apolipoprotein B100, apolipoprotein E and microsomal triglycerides transfer protein in liver from periparturient dairy cows. J. Dairy Sci. 87:2881-2888.

Bernabucci, U., N. Lacetera, L. H., Baumgard, R. P. Rhoads, B. Ronchi and A. Nardone (2010). Metabolic and hormonal acclimation to heat stress in domesticated ruminants. Animal, 4: 1167–1183.

Bobe, G., B. N. Ametaj, J. W. Young and D. C. Beitz (2003). Effects of exogenous glucagon on lipids in lipoproteins and liver of lactating dairy cows. J. Dairy Sci. 86:2895–2903.

Casati, M. R., V. Cappa, L. Calamari, F. Calegari and G. Folli (1998). Effects of the season on milk yield and on some milk characteristics in cows. Scienza e TecnicaLattiero-casearia, 49: 7-25.

Collier, R.J., D.K. Beede, W.W. Thatcher, L. A. Israel and C. J. Wilcox (1982). Influences of environment and its modification on dairy animal health and production. J. Dairy Sci. 65:2213- 2227.

Duncan, D.B. (1955). Multiple range and multiple F-tests. Biometrics 11:1-42.

Gibbons, G. F. (1990). Assembly and secretion of hepatic very-low density lipoprotein. Biochemical J. 268:1–13.

Grummer, R. R. (1993). Etiology of lipid-related metabolic disorders in periparturient dairy cows. J. Dairy Sci. 76:3882-3896.

Grummer, R. R. (1995). Impact of changes in organic nutrient metabolism on feeding the transition dairy cow. J. Anim. Sci. 73:2820–2833.

Gupta, P.C., V.K. Khatta and A.B. Mandal (1992). Analysis Techniq in Animal Nutr.CCS HAU, Hisar, 46 p

Haenlein, G. (2003). Nutritional value of dairy products of ewe and goat milk. Retrieved January 28, fromhttp://ag.udel.edu/extension/information/goatmgt/gm-10.htm

Habeeb, A. A., F. M. Marai and T.H. Kamal, (1992). Heat stress. P.27-47. in: Farm Animals and the Environment, C. Phillips and D. Piggins, (Eds.). CAB International, Wallingford, UK.

Haque, N., A. Ludri, S. Hossain and M. Ashutosh (2012). Alteration of metabolic profiles in young and adult Murrah buffaloes exposed to acute heat stress. Int. J. Applied Anim. Sci. 1:23-29.

Itoh, H., K. Tamura, Y. Motoi and F. Kawawa (1997). Serum apolipoprotein B-100 concentrations in healthy and diseased cattle. J. Vet. Med. Sci. 59: 587-591.

Kamal, T. H. (1976).Indices for heat adaptability of domestic animals. In Progress in Animal Biometeorology. The effect of weather and climate on animals (H.D. Johnson, Ed.). Swets and Zeitlinger, Amesterdam, 1:470.

Karapehlivan, M., A. Atakisi, O. Atakisi, R. Yucayurt and S. M. Pancarci (2007). Blood biochemical parameters during the lactation and dry period in Tuj ewes. Small Rum. Res., 73: 267-271.

Krajnicakova, M., G. Kovac, M Kostecky, I. Valocky, I. Maracek, I. Sutiakova and L. Leonhard (2003). Selected clinic-biochemical parameters in the puerperal period of goats. Bulletin Veterinary Institute Pulawy. T.47. P. 177–182.

Lacroix, C., P. Verret and P. Paquin (1996). Regional and seasonal variations of nitrogen fractions in commingled milk. Int. J. Dairy, 6: 947-961.

Marai, I. F. M. and A. A. M. Haeeb (2010). Buffalo's biological functions as affected by heat stress - A review. Livestock Science 127:89-109.

Mohri, M., K. Sharifi and S. Eidi (2007). Hematology and serum biochemistry of Holstein dairy calves: age related changes and comparison with blood composition in adults. Res. Vet. Sci., 83: 30–39.

Myamoto, T., Y. Sugiyama, J. Suzuki, T. Oohashi and Y., Takahashi (2006). Determination of bovine serum low-density lipoprotein cholesterol using N-geneous method. Vet. Res. Com., 30: 467-474.

Nardone, A., B., Ronchi, N. Lacetera and U. Bernabucci (2006).Climatic effects on productive traits in livestock.Vet. Res. Communications 30 (1): 75-81.

Nikolić, J.A., H. Šamanc, P. Begović, Z. Damjanović, R. Djoković, G. Kostić, J. Krsmanović and V. Resanović (1997). Low pripheral serum thyroid status independently affects the hormone profile of healthy and ketotic cow during the first week postpartum, Acta Vet., 47. 1, 3-14.

Piccione, G., G. Caola, C. Giannetto, F. Grasso, S. CalanniRunzo, A. Zumbo and P. Pennesi (2009). Selected biochemical serum parameters in ewes during pregnancy post-parturition, lactation and dry period. Animal Sci. 27: 321– 330.

Re´mond, B, M. Cisse, A. Ollier and Y. Chilliard (1991). Slow release somatotropin in dairy heifers and cows fed two levels of energy concentrate. J. Dairy Sci. 74: 1370–1381.

Roche, J.R., N.C Frggens, J. K Kay, M.W. Fisher., K.J Stafford and D.P. Berry (2009). Invited review: Body condition score and its association with dairy cow productivity health and welfare. J. Dairy Sci., 92: 5769–5801.

Ronchi, B., U. Bernabucci, N. Lacetera, A. VeriniSupplizi and A. Nardone (1999). Distinct and common effects of heat stress and restricted feeding on metabolic status of Holstein heifers. Zoot. Nutr. Anim. 25:11-20.

Roubies, N., N. Panouis, A. Fytianou, P.D. Koutsoubos, N. Giadinis and H. Karatzias (2006). Effects of age and reproductive stage on certain serum biochemical parameters of Chios sheep under Greek rearing conditions. J. Vet. Med. 53: P. 277–281.

Sharma, A. K., L. A. Rodriguez, C. J. Wilcox, R.J. Collier, K.C. Bachman and F.G. Martin (1988). Interactions of climatic factors affecting milk yield and composition. J. Dairy Sci., 71: 819-825.

Shin, O., and K. Norio (1997). Reduced concentrations of Apolipoproteins B-100 and A-I in serum from cows with retained placenta. Canada J. Vet. Res. 61: 312-314

Tainturier, D., J. P. Braun, A. G. Rico and J. P. Thouvenot (1984). Variation in blood composition in dairy cows during pregnancy and after calving. Res. Vet. Sci., 37: 129-131.

Teama, F. E. I and A.E Gad (2014). Leptin, thyroxin and cortisol hormones and some metabolic products during pre and postpartum periods in cows in relations to their body weight of newborn calves. Global Vet. 12 (1): 59-66.

Tucker, H. A. (1989). Photoperiod affects intake, growth and milk production of cattle. Feedstuffs, 61: 15-16.

Vazquez- Anon, M., S. Bertics, M. Luck, R. Grummer and J. Pinheiro (1994). Prepartum liver triglyceride and plasma metabolites in dairy Cows. J. Dairy Sci. 77: 1521-1528.

Wathes, D. C., Z. Cheng, N. Bourne, V. J. Taylor, M. P. Coffey and S. Brotherstone (2007). Differences between primiparous and multiparous dairy cows in the inter-relationships between metabolic traits, milk yield and body condition score in the periparturient period. Dom. Anim. En. 33: 203-225.

West, J. W. (1999). Nutritional strategies for managing the heat-stressed dairy cows. J. Anim. Sci., 77:21-35.

Yokus, B., D. U. Cakir, Z. Kanay, T. Gulten and E. Uysal (2006). Effects of seasonal and physiological variations on the serum chemistry, vitamins and thyroid hormone concentrations in sheep J. Vet. Medicine 53(6) 271-276.