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Impact of seasonal variation on Physio-biochemical attributes of goats

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Abstract
Physiological and blood biochemical parameters were evaluated twice a day (morning and afternoon) in each season to investigate the goat's adaptation profile in three different seasons (winter, spring, and summer). The temperature and relative humidity of the air were measured, and rectal temperature, respiration rate, and heart rate were among the physiological data measured. The findings of this study revealed that season and period had a substantial impact on all environmental factors. In all seasons, physiological indicators such as rectal temperature, respiration rate, and heart rate had greater values in the afternoon. The season of the year has a substantial impact on blood biochemical markers. The biochemical features of blood fluctuate with the seasons, with metabolism slowing under heat stress and speeding up during cold stress. These physiological and blood biochemical indicators were shown to be involved in the adaption process in this investigation.

Keywords: Biochemical attributes, goat, physiological responses, Seasonal stress

1. Introduction
Goats are warm-blooded animals with the ability to keep their body temperature within tight ranges. To do so, they must adapt their metabolism for internal heat production in addition to exchanging with the environment (Nardone *et al.*, 2010) [22]. Few research on the adaptive character of local goat populations, particularly in rural marginal areas, have been done, and these researches are critical for the development of management and conservation measures. Even in some affluent countries, many local populations were not sufficiently defined in this regard (Ribeiro *et al.*, 2015) [31]. The connection between animals and the environment must be examined in order to increase farming efficiency; knowledge of climate factors and their impacts on physiological, haematological and blood biochemical responses is essential for optimising livestock-raising systems.

Animals respond to extreme temperature changes in various ways, altering their physiology and behaviour in the process (Silva *et al.*, 2010) [38]. Physiological parameters (rectal temperature, heart rate, respiratory rate, and surface temperature) (Marai *et al.*, 2007) [20] and blood biochemical parameters (Abdelatif *et al.*, 2009) [27] are just a few examples. Diet, age, physiological health, breed, degree of production, handling, and, most importantly, climate stress can all affect these parameters within the same species. This study aimed to use physiological and blood biochemical, data to assess the adaptation profile of the goat in three different seasons (Winter, spring and summer).

2. Materials and Methods
The present study was conducted on goats (*Capra hircus*) to identify the adaptations of native goat breeds of tropical climatic conditions of Northern India, to understand the seasonal variation on physiological and blood biochemical parameters profile in different seasons. The experiments were conducted on the goats at ICAR NDRI, Karnal, Haryana. Goats used in the study were non-lactating, non-gestating female and male goats. The goats were of 2-3 years old and had an average weight of 25-30 kg. The whole experiment was conducted in three seasons.

Experiment 1: Conducted during the winter season (December to January)
Experiment 2: Conducted during thermo-neutral zone (March)
Experiment 3: Conducted during summer season (May)

2.1. Climatological Measurements

The experiments were conducted in winter (December – January), thermo-neutral zone (March) and summer (May) seasons. The climatic data recorded during the experimental period were analyzed and THI were calculated. The following environmental parameters were recorded twice daily shown in table 1.

1. Maximum and Minimum Temperature
2. Dry bulb and Wet bulb temperature were recorded using dry and wet bulb thermometers.

Temperature Humidity Index (THI) was calculated from the dry bulb and wet bulb thermometer using the equation: THI = 0.72 x (Cdb + Cwb) + 40.6 (Mc Dowell, 1983), where, Cdb = Average dry bulb (°C), Cwb = Average wet bulb (°C).

Table 1: Environmental parameters in different seasons during the experimental days

Environmental Variable	Winter	Spring (Thermoneutral)	Summer
Dry bulb Temperature (0C)	14.26(9.44 to 19.44)	23.80(22.77 to 24.44)	36.67(31.66 to 40.55)
Wet bulb Temperature (0C)	11.67(9.4 to 14.4)	18.38(17.77 to 19.44)	23.61(21.11 to 26.66)
Relative Humidity (%)	76.97(53.5 to 100)	59.75(53.9 to 66.4)	36.57(16.3 to 58.6)
THI	59.27(54.2 to 65)	70.97(69.8 to 71.8)	84(81.4 to 86.2)

2.2. Physiological Responses

2.2.1. Rectal Temperature

Rectal temperature (°C) was recorded with a digital thermometer by placing it in the rectal lumen, assuring that it touches the epithelial layer.

2.2.2. Respiration Rate

Respiration rate was recorded by observing flank movement, and each subsequent inward and outward movement was recorded and taken as a breath per minute.

2.2.3. Heart Rate

Heart rate was recorded using a stethoscope at the medial side of the left forelimb between the 3rd and 5th rib and counted for one minute.

2.3. Biochemical Components

2.3.1. Total Protein

Biuret method: Protein binds with copper ions in an alkaline medium of the Biuret reagent and produces a purple-coloured complex, whose absorbance is proportional to the protein concentration. The components were mixed well and incubated at 37°C for 5 min. The absorbance of the test and standard were measured against the reagent blank at 546nm (530- 570nm).

2.3.2. Albumin

Bromocresol green (BCG) method: The method is based on the specific binding of bromocresol green (BCG), an anionic dye, and the protein at acid pH produce a colour change of the indicator from yellow-green to green-blue with the resulting shift in the absorption wavelength of the complex. The intensity of the colour formed is proportional to the concentration of albumin in the sample.

2.3.3. Plasma Urea

Blood urea nitrogen in the plasma sample was estimated using a RECOMBIGEN® UREA UV (liquid stable) kit purchased from Arkray Healthcare Pvt Ltd.

2.3.4. Creatinine

Jaffe's modified method (End Point & Kinetic Method): (ALKALINE PICRATE METHOD) Reagent kit for quantitative estimation of CREATININE in serum or urine. Creatinine kit is based on Jaffe's Kinetic method. A drawback of Jaffe's endpoint reaction is the interference due to non-specific substances such as proteins, ascorbic acid and ketoacids. Various modifications are aimed at either eliminating or minimizing this interference. Before the picrate creatinine complex formation is monitored in the kinetic method, a delay minimizes interference from the fast**-reacting** substances such as ketoacids and hence subsequent measurements up to 120 seconds, broadly refers to actual creatinine values only. Other advantages of the kinetic reaction include no deproteinization, rapid method and low sample volume.

3. Result and Discussion

3.1. Physiological Responses of goats during different seasons

3.1.1. Respiration Rate (breathe/minute): The Mean ± S.E.M. values of respiration rate of goats during different seasons are depicted in the Fig. 1 and table 2. The respiration rate (breaths per minute) during the winter, spring and summer seasons was 22.33 ± 0.28, 23.26 ± 0.41 and 36.86 ± 0.21, respectively. A significant (*P*<0.05) increase in respiration rate was observed during summer when compared with the winter season. However, no significant difference was observed when the spring values were compared with that of the winter season. The per cent values of respiration rate were found to be increased by 58.46% from spring to summer and a 3.99% decrease from spring to winter. Whereas an elevation of about 65.06% in respiration rate was observed from winter to summer season. Therefore, an increase in the average respiration rate by 14.53 breathe/minute was observed in goats from the winter to summer.

Table 2: Mean ±S.E.M. values of Physiological reactions in goats during different seasons

Parameter	Winter	Spring	Summer
Respiration Rate (Breathe/min)	$22.70^{b} \pm 0.28$	$23.00^{b} \pm 0.41$	$36.86^{a} \pm 0.21$
Heart Rate (Beats/min)	$74.66^{b} \pm 0.90$	$73.40^{b} \pm 0.38$	$83.20^{a} \pm 0.51$
Rectal Temperature (°C)	$39.11^{a} \pm 0.04$	$39.20^{a} \pm 0.06$	$39.24^{a} \pm 0.05$

Means with different superscripts (a, b and c) within same rows differ significantly (*P*< 0.05)

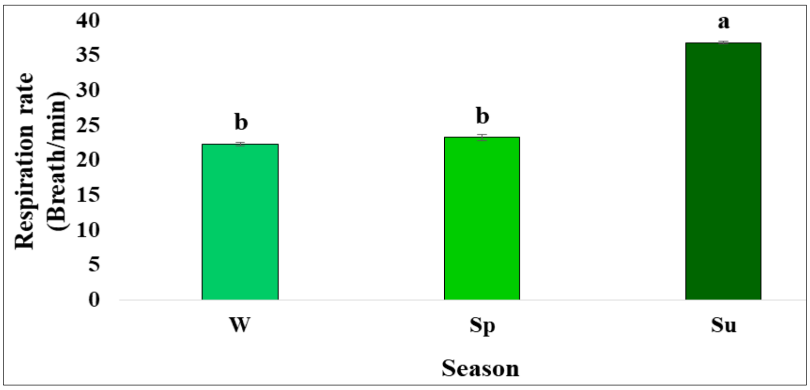

Fig 1: (Mean ± SEM) values of Respiration rate in goats during different seasons
Bars with different alphabets differ significantly (P< 0.05)

3.1.2. Heart rate (beats per minute)

The Mean ± S.E.M values of goats' heart rate during different seasons were presented in Fig 2 and table 2. The average heart rate (beats per minute) of experimental animals recorded during winter, spring and summer seasons were 74.66 ± 0.90, 73.40 ± 0.38 and 83.20 ± 0.51 breath/minute, respectively. The heart rate was significantly (P< 0.05) increased in summer compared to the winter and spring season, whereas the values non-significantly varied between spring and winter. The per cent values of heart rate were found to be increased by 13.35% from spring to summer and 1.17% increase from spring to winter. Whereas an elevation of about 11.43% in heart rate was observed from winter to summer. Therefore, an increase in the average heart rate by 8.54 beats/minute was observed in goats from winter to summer.

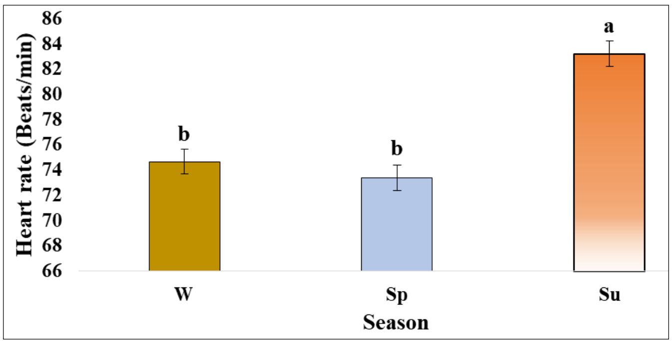

Fig 2: (Mean ± SEM) values of Heart rate (M in goats during different seasons

Bars with different alphabets differ significantly (P< 0.05)

3.1.3. Rectal Temperature (C)

The Mean ± S.E.M. values of rectal temperatures of goats during different seasons are presented in table 2 and depicted in Fig. 3. In the experimental animals, the average rectal temperature (°C) of 39.11 ± 0.04, 39.20 ± 0.06 and 39.24 ± 0.05 °C were observed during winter, spring and summer seasons, respectively. No significant variation was observed in the average rectal temperature of the animals during all seasons, and values of rectal temperature was found to be within the normal range of the goat. The per cent value of rectal temperature increased by 0.10% from spring to summer and 0.22% decreased from spring to winter. Whereas an elevation of about 0.33% in rectal temperatures was observed from the winter to summer season.

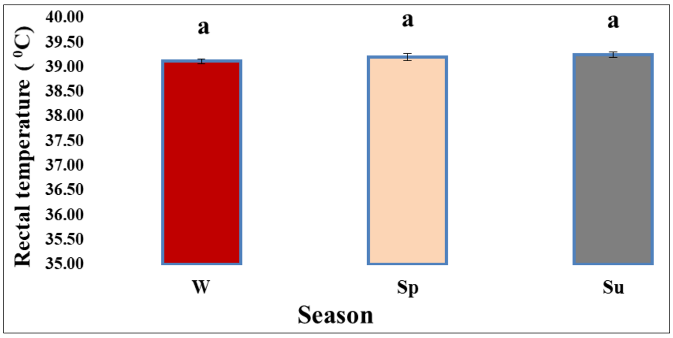

Fig 3: (Mean ± SEM) values of rectal temperature in goats during different seasons

Bars with different alphabets differ significantly (P< 0.05)

3.2. Blood Biochemical components of goats during different seasons

3.2.1. Total protein (gm/dL)

The Mean ± S.E.M. values of total plasma protein concentrations during different seasons in goats are presented in table 3 and depicted in Fig. 4. The average total protein concentrations in plasma recorded during winter, spring, and summer seasons were 8.11 ± 0.06, 7.27 ± 0.09 and 6.24 ± 0.06 gm/dL, respectively. The total protein concentration during winter was significantly (P< 0.05) higher than that of spring and summer season values. However, the plasma protein concentrations during summer were significantly lower than the spring values. The per cent values of total plasma proteins decreased by 14.16% from spring to summer and 11.55% increase from spring to the winter season. Whereas a decline of about 23.05% in total plasma protein was observed from the winter to summer.

Table 3: Mean ± S.E.M. values of Blood Biochemical parameters of goats during different Seasons

Parameter	Winter	Spring	Summer
Total Protein (gm/dL)	$8.11^{a} \pm 0.06$	$7.27^{b} \pm 0.09$	$6.24^{c} \pm 0.06$
Urea (mmol/L)	$4.51^{c} \pm 0.20$	$6.67^{b} \pm 0.08$	$8.70^{a} \pm 0.16$
Creatinine (mg/dL)	$1.38^{a} \pm 0.04$	$1.41^{a} \pm 0.04$	$1.45^{a} \pm 0.03$
Albumin (gm/dL)	$4.07^{a} \pm 0.04$	$3.59^{b} \pm 0.04$	$2.51^{c} \pm 0.08$

Means with different superscripts (a, b and c) within same rows differ significantly (P<0.05)

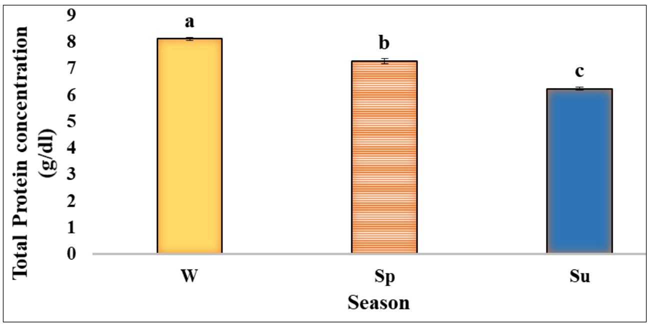

Fig 4: (Mean ± SEM) values of Plasma Total Protein concentration in goats during different seasons

Bars with different alphabets differ significantly (*P*<0.05)

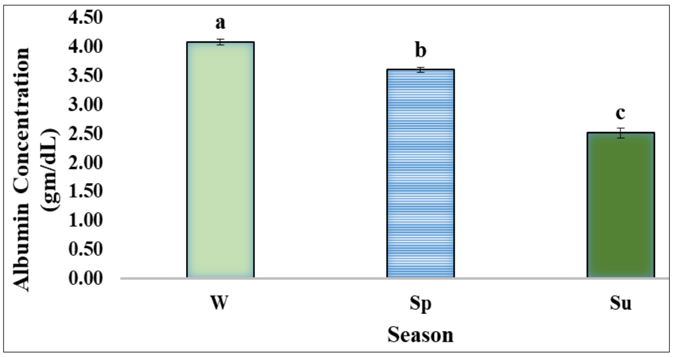

Fig 5: (Mean ± SEM) values of Plasma Albumin concentration in goats during different seasons

Bars with different alphabets differ significantly (*P*< 0.05)

3.2.2. Plasma Albumin (gm/dL)

The Mean ± S.E.M. values of plasma albumin concentrations during different seasons in goats are presented in table 3 and depicted in Fig. 5. The average albumin concentrations in plasma during winter, spring and summer seasons were recorded4.07 ± 0.04, 3.59 ± 0.04 and 2.51 ± 0.08 gm/dL, respectively. The albumin concentration during winter was significantly (*P*<0.05) higher than that of spring and summer values. However, the plasma albumin concentrations during spring were significantly higher than in summer values. The per cent values of plasma albumin concentrations were found to be decreased by 30.08% from spring to summer and 13.37% increase from spring to winter. Whereas a decline of about 38.32% in plasma albumin concentrations were observed from winter to summer.

3.2.3. Blood Urea (mmol/L)

The Mean ± S.E.M. values of urea concentrations in goats during different seasons are presented in table 3 and depicted in Fig. 6. The average urea concentrations in the blood of the animals during winter, spring and summer seasons were recorded 4.51 ± 0.20, 6.67 ± 0.08 and 8.70 ± 0.16 mmol/L, respectively. During summer, the average blood urea concentration was significantly (*P*< 0.05) higher than the spring and winter values. However, the urea concentration during winter was significantly lower than in spring values. The per cent values of plasma urea concentration increased by 30.43% from spring to summer and 32.38% increase from spring to winter. Whereas a elevate of about 92.90% in urea concentrations was observed from winter to summer season.

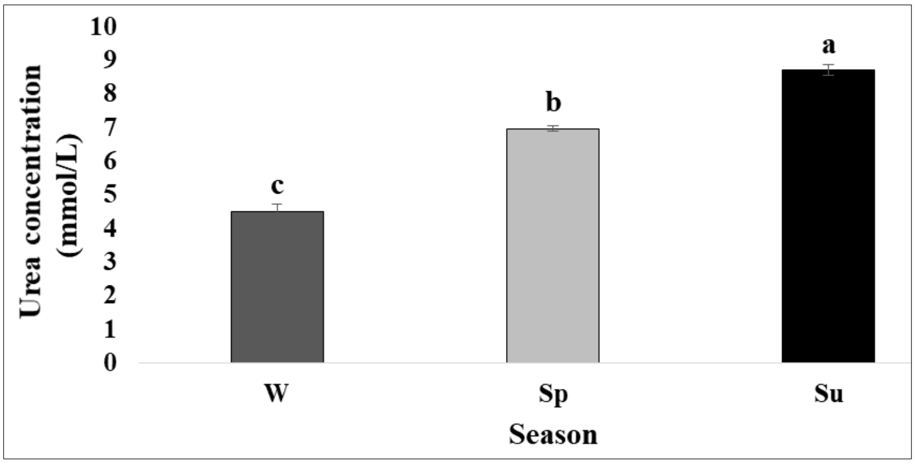

Fig 6: (Mean ± SEM) values of Plasma Urea concentration in goats during different season
Bars with different alphabets differ significantly (*P*<0.05)

3.2.4. Blood creatinine (mg/dL)

The Mean ± S.E.M. values of creatinine concentration in goats during different seasons are presented in table 3 and depicted in Fig. 7. The average creatinine concentrations in the blood of the animals during winter, spring and summer seasons were 1.38 ± 0.04, 1.41 ± 0.04 and 1.45 ± 0.03 mg/dL, respectively. The average creatinine blood concentration creatinine during all seasons were found to be non-significant (*P*< 0.05) and within the normal range of creatinine level in goats. The per cent values of plasma creatinine concentrations were found to be increased by 2.83% from spring to summer and a 2.12% decrease from spring to winter. Whereas an elevate of about 5.07% in creatinine concentrations were observed from winter to summer season.

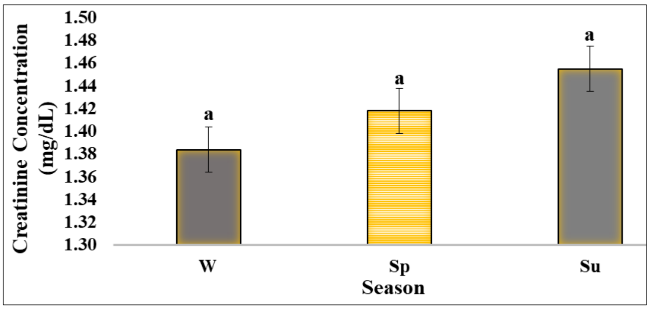

Fig 7: (Mean ± SEM) values of Plasma Creatinine concentration in goats during different seasons

Bars with different alphabets differ significantly (*P*< 0.05)

4. Discussion

4.1. Physiological Responses of goats during different seasons

In the current study's findings during different seasons, the mean respiration rate, rectal temperature, and heart rate are within the ranges as reported by Phulia *et al.* (2010) [25] and Darcan *et al.* (2008) [7]. An animal adapts to stressful conditions by altering the physiological mechanisms of its body (Gupta *et al.*, 2012) [12]. Francis, 2017 reported a significant (p< 0.01) increase in the rectal temperature, pulse rate, respiration rate, and skin temperature from winter to the summer season in Sirohi and Barbari goats. Among the physiological variables commonly assessed in adaptability studies in small ruminants are rectal temperature, heart rate and respiratory rate. In general, physiological variables change depending on the season, age, sex, time of day, physiological stage, exercise, water consumption, food intake and digestion (Otoikhian *et al.*, 2009; Phulia *et al.*, 2010; Sharma and Kataria 2011; Leite *et al.*, 2012; Lucena *et al.*, 2013; Ribeiro *et al.*, 2016, 2018) [24, 25, 37, 17, 18, 29, 30]. Increased respiration rate is due to an increase in demand for oxygen by the body during environmental stress. The initial response to heat stress increases its respiratory rate, causing heat dissipation by evaporation (Renaudeau *et al.*, 2012) [28]. Raised in heart rate makes the blood flow to reach peripheries and body extremities, so heat can be conducted to the outer environment. A decrease in the rectal temperature, respiration rate, and heart rate was observed when animals were exposed to lower environmental temperature (Appleman and Deuouche in 1958) [4].

Saini *et al.*, (2013) [34] observed variations in the heart rate in Marwari sheep in response to the water deprivation with the advancement of thirst period, the mean values of heart rate gradually increased.

Higher physiological responses during summer in goats and a lower physiological response during the winter season denotes its greater flexibility to alter its physiological responses to overcome environmental stress during different seasons of animals. According to Swenson and Reece (2015) [40], the rectal temperature in goats varies from 38.3°C to 40.0°C and has frequently been used as an indicator of the animals' body temperature, although there is a variation in body temperature in different parts of the body throughout the day. The daily variation in rectal temperature ranges from 0.3°C to 1.9°C (Piccione and Refinetti 2003) [26]. In different environmental conditions, the rectal temperature is maintained within the proper range for goats (Rocha *et al.*, 2009; Salles *et al.*, 2009; Phulia *et al.*, 2010; Aiura *et al.*, 2010; Leite *et al.*, 2012; Lucena *et al.*, 2013; Ribeiro *et al.*, 2018) [32, 35, 25, 2, 17, 18, 30]. During heat exposure, animals access respiration as a mechanism to avoid increased rectal temperature, maintaining homeothermy. The increase in respiratory frequency and heart rate can efficiently lose heat for short periods (Berbigier and Cabello 1990) [5]. Several studies were developed on this issue as those reported by Rocha *et al.* (2009) [32], Leite *et al.* (2012) [17], Lucena *et al.* (2013) [18], Ribeiro *et al.* (2018) [30].

Abdelatif *et al.* (2010) [1] reported an increase in the rectal temperature during dehydration in Nubian goats. Alamer and Al-hozab (2004) [3], Eyal (1963) [9], MacFarlane (1964) [19] and Taneja (1965) [41] have reported increased rectal temperature at times of water deprivation in small ruminants whereas, Hamadeh *et al.* (2006) [13] and Jaber *et al.*, (2004) have reported rectal temperature in the normal ranges.

4.2. Blood Biochemical components of goats during different seasons

The plasma total protein concentration, observed in the current study, was significantly lower in summer than the winter values, and the trend is similar to the findings of Dangi SS *et al.*, 2012 [6] and Debbarma, 2019 [8]. A significant decrease in total protein and albumin concentration in goats have been reported during heat stress may be due to an increase in plasma volume resulting from heat stress, which results in a decrease in plasma protein concentration. In contrast, Helal A *et al.*, 2010 [14] reported that total plasma protein levels increase in goats subjected to heat stress could be due to dehydration which has been reported to occur due to increased respiration rate and sweating rate in hot weather. The concentration of total plasma protein is decreased during high ambient temperature due to reduced feed intake (Srikandakumar and Johnson, 2004) [39] and hemodilution conditions induced by heat stress (Gudev *et al.*, 2007) [11]. Farouk (2012) [10] did not find any significant differences in plasma total protein between the summer and winter seasons. Concentrations of serum total proteins and albumin have been used as indices for nutritional status. Serum total proteins and albumin concentration decreased during thermal stress in goats, and our results were similar to those of Sejian *et al.*, (2013) [36]. In the present study, plasma urea was significantly higher during the summer than the winter season. The increased concentration of urea may be due to AQP 3 that helps absorb urea into the medullary part of the nephron and helps concentrate the urine. Urea is very useful in concentrating the urine. A high protein diet intake causes more urea production, which may lead to more concentrated urine. The kidney filters the blood and help in the reabsorption and secretion of urea. Urea contributes to high solute concentration in the interstitial fluid (ISF) of the kidney medulla. The presence of urea is accomplished by a recirculation mechanism for urea between the collecting ducts and the loop of Henle. The recirculation mechanism and high concentration of urea in the medulla assist the counter current multiplier system and osmotic gradient and ensure excretion of urea when urine output is low (Reece, 2015) [40]. The other mechanism of increased urea concentration may also be due to the negative effect of increased body temperature during heat stress on rumen micro flora leading to increased plasma urea concentration (Gudev *et al.*, 2007) [11]. Joshi *et al.*, (2012) [16] reported highest value of serum urea during hot followed by cold and moderate ambience. In the present study, serum creatinine level is non-significant in different seasons. Similar results obtained by (Abdel-Samee *et al.*, 2008) that nonsignificant serum creatinine in blood under heat stress conditions. Regarding the results of serum creatinine found nonsignificant; and within normal range showing that kidney of goats was in healthy condition and renal function was normal as well as suitable muscle thickness. Creatinine is produced as the result of normal muscle metabolism. Phosphocreatine, an energy-storing molecule in muscle, undergoes spontaneous cyclization to form creatine and inorganic phosphorous. Creatine then decomposes to creatinine. Creatine is actually produced in the liver from amino acids (glycine and arginine) that the kidney has modified. In health, production and excretion of creatinine are relatively constant in an individual animal, resulting in low variation in an individual animal (Ruaux *et al.*, 2012, Hokamp and Nabity 2016) [33, 15]. If an increase in plasma creatinine level it indicates of kidney disfunction. Creatinine is a product of the breakdown of creatine phosphate in muscle and is expected to be high in more extensive or older animals with more skeletal muscle, reinforcing the view that excretion of creatinine is closely related to muscle mass and is high in bigger one's animals (Nonaka I *et al.*, 2008) [23].

5. Conclusions

The goat adapted to seasonal weather fluctuations in the environment it was found in research. The animals demonstrated a high ability to maintain a constant rectal temperature while slightly raising their respiratory rate during summer, as well as a large capacity for heat dissipation. They, on the other hand, respond quickly to cooler temperatures, increasing their heat-producing capability. Physiological and Biochemical properties alter during changing seasons, such as metabolism slowing during heat stress and speeding up during cold stress. As a result, proper assessment of the adaptive profile necessitates taking into account animals' physiological and biochemical parameters to environmental conditions.

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