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RESEARCH ARTICLE

Dietary Supplementation of Seaweed (*Ulva lactuca*) to alleviate the Impact of Heat Stress in Growing Lambs

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ABSTRACT

Several environmental and nutritional management approaches have been used to mitigate heat stress and improve performance of farm animals in semi-arid and arid regions. The present study was designed with the intention to alleviate the negative effects of heat stress and to promote the performance of growing lambs reared under hot environmental conditions. The study was conducted on 18 male Naimey lambs with average body weight of 22.78±0.49 kg, and 4-5 months old. The animals were randomly divided into 3 equal groups (A, B and C), and fed diets containing different concentrations of seaweed (*Ulva lactuca*) for 90 days. Group A served as control and was offered diet containing 0.0% seaweed. Groups B and C served as treated groups and were offered diets containing 3.0 and 5.0% seaweed, respectively. Dietary inclusion of seaweed to the diet of growing lambs exposed to heat stress (max T_a 43.9^{o}C, max RH 81.1%, max THI 84.6) neither influenced (P>0.05) the thermo-physiological parameters (rectal and skin temperatures), nor affected (P>0.05) the performance parameters (feed intake, body weight gain, feed conversion efficiency). Furthermore, dietary seaweed supplementation did not alter (P>0.05) blood constituents or blood antioxidant capacity. However, dietary seaweed supplementation significantly (P<0.05) reduced respiratory rate, and increased serum potassium concentration. Based on the data of the present study, seaweed (*Ulva lactuca*) supplementation to the diets of growing lambs reared under heat stress conditions did not show any indication of promoting their production performance or heat tolerance.

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INTRODUCTION

One of the major problems facing sheep production in the arid and semi-arid regions like Saudi Arabia is the heat stress resulting from high ambient temperature and solar radiation causing the effective temperature of the environment to exceed the thermo-neutral zone, particularly during summer season (Ali *et al.,* 1999). Hot environmental conditions are well known to negatively affect livestock productive and reproductive performance (Nardone *et al.,* 2010; Isani *et al.,* 2012; Baumgard and Rhoads, 2013). This could result in a tremendous economic loss for sheep industry. Elevated body and skin temperatures, accelerated respiratory and heart rates and post absorptive metabolic changes are the most important characteristics for heat stress in ruminants (Bernabucci *et al.,* 2010; Al-Haidary *et al.,* 2012; Abdoun *et al.,* 2012).

Several approaches have been used to alleviate heat stress and improve performance in farm animals, including management and feeding strategies. Although no feed additives have effectively reduced heat stress, some evidence exists that brown seaweed (*Ascophyllum nodosum*) may decrease core body temperature (T_{core}) for the short term (Spiers *et al.,* 2004; Archer *et al.,* 2007) and improve immune function and antioxidant capacity during heat stress, without affecting performance (Ruperez *et al.,* 2002; Saker *et al.,* 2004; Okab, 2007). Furthermore, it has been shown that brown seaweed fed at 1% of diet DM had a short-term effect in reducing T_{core} of beef steers exposed to heat stress (Spiers *et al.,* 2004; Williams *et al.,* 2009). There are controversy reports concerning effects of seaweeds on the performance of livestock. Brown seaweed inclusion in the diet of dairy cattle at 0.25 and 0.5% DM had no effect (P>0.10) on overall DMI, DMI per BW, or milk production (Pompeu *et al.,* 2011).

Moreover, dietary supplementation of brown seaweed to the diets of male goats did not show significant effect on weight gain (Yates *et al.,* 2010). In contrast, Kellogg *et al.* (2006) found that brown seaweed supplemented at 0.25% of diet increased milk production of cows during heat stress. While, Williams *et al.* (2009) reported that dietary supplementation of brown seaweed to steers at 1% DM had lowered DMI. However, other species of seaweed have been observed to increase feed intake and body weight gain (Al-Shorepy *et al.,* 2001; Turner *et al.,* 2002).

To our knowledge, there is a lack of information pertaining to dietary inclusion of seaweed to the diet of growing lambs; therefore, the objective of the present study was to evaluate the ability of green seaweed (*Ulva lactuca*) to alleviate the effects of heat stress and promote performance of growing lambs under hot summer conditions.

MATERIALS AND METHODS

Animals, management and experimental design: The current study was conducted on growing Naimey male lambs and lasted for 90 days (May-August 2012). A total of eighteen (4-5 months old) Naimey male lambs with an average live body weight of 22.78±0.49 kg were included in this study. Two weeks prior to commencement of the study, animals were fed alfalfa hay, ear tagged, and treated with anthelmintics. Naimey lambs were randomly allocated into 3 equal groups (A, B and C), fed twice daily at 3% of their body weight and had free access to clean fresh tap water. Lambs in group A were offered a diet containing commercial feed without seaweed supplementation and assigned as control diet, those in groups B and C were offered control diet supplemented with 3 and 5% seaweed, respectively. The ingredients and chemical composition of the experimental diets are shown in Table 1.

Seaweed collection and preparation: Seaweed (*Ulva lactuca*) was collected from costal zones, washed with water, sun-dried and further dried at 60°C for 72 h in oven. The oven-dried seaweed was grounded through a 1 mm stainless-steel screen using Wiley mill grinder and chemically analyzed. Finally, seaweed was incorporated in the experimental diets at 3 and 5% DM.

Experimental measurements: Ambient temperature (T_a) and relative humidity (RH) were recorded continuously at 30 min interval using 2 data loggers (HOBO Pro Series data logger, Model H08-032-08, ONSET Co., USA) mounted at a height of approximately 3 m from the ground, and placed away from direct sources of heat, sunlight and water. Special data logging software (BoxCar Pro 4, ONSET Co., USA) was applied for programming the loggers and for data analysis. Temperature-humidity index (THI) was calculated using T_a and RH measurements to estimate the environmental severity on the animals according to Moran (2005).

Feed was offered twice daily and then adjusted to the actual intake to minimize the refusal. Individual lamb's feed intake was measured daily by subtracting the refusal weight from the total feed weight offered. Individual lamb's weight was recorded weekly before morning meal.

Table 1: The ingredients and proximate analysis of the experimental diets*

	Diet A	Diet B	Diet C
Ingredients (%):	40	37	35
Alfalfa hay			
Seaweed	0	3	5
Barley	25	25	25
Corn	25	25	25
Soybean meal	7	7	7
Sodium chloride	1	0.8	0.7
Lime stone	1	1	1
Sodium bicarbonate	0.8	1	1.1
Vitamins	0.2	0.2	0.2
Proximate analysis (%):			
Dry matter (DM)	93.38	93.18	93.47
Crude protein (CP)	15.87	15.75	16.07
Ether extract (EE)	2.52	2.47	2.43
Ash	8.74	9.72	10.3
Neutral detergent Fiber (NDF)	27.59	27.16	26.46
Acid detergent fiber (ADF)	16.62	16.38	16.14

*Diet A = 0.0% seaweed; Diet B = 3.0% seaweed; Diet C = 5.0% seaweed.

Daily weight gain as well as feed conversion ratio was calculated.

Rectal (T_r) and skin (T_{sk}) temperatures as well as respiratory rate (RR) were determined once per week at 11:00h throughout the experiment. Blood samples were collected through jugular venipuncture into EDTA tubes (for haematological analysis) and into plain tubes (for serological analysis). Collected samples were immediately placed in an ice box and transferred to the laboratory. Within 1 h of collection, EDTA tubes were used to measure packed cell volume using haematocrit tubes and haematocrit centrifuge (4000 g for 5 min). Serum samples were prepared by centrifuging plain tubes at 1500 g for 10 min. The obtained serum samples were stored at -20°C for further analysis of total protein, albumin, glucose, total lipids, cholesterol, total antioxidant capacity, sodium and potassium concentrations using commercial kits (United Diagnostics Industry, Dammam, KSA). Serum globulin concentration was calculated as the difference between total protein and albumin concentrations. The osmolality of serum samples was measured using Vapor Pressure Osmometer (Vapro® Model 5600, Wescor Inc, USA).

Statistical analysis: Statistical analysis was performed by analysis of variance (ANOVA) using the general linear model (GLM) of Statistical Analysis System (SAS). Completely randomized design was utilized to analyze the experimental data. Means showing significant differences in ANOVA were tested using the PDIFF option.

RESULTS

Ambient conditions and performance parameters: The average daily values for maximum, mean, and minimum ambient temperatures (T_a) were 43.9, 29.6, and 15.6°C, respectively, and the maximum, mean, and minimum RH were 81.1, 23.5, and 04.7%, respectively. The computed daily values for maximum, mean, and minimum temperature humidity index (THI) were 84.6, 73.3, and 59.8, respectively. Seaweed inclusion in the diet at 3 and 5% DM had no effect (P>0.05) on average daily feed intake, daily body weight gain or feed conversion efficiency of growing Naimey lambs (Table 2).

Rectal temperature, skin temperature and respiratory rate: Inclusion of seaweed in the diet of lambs had no

Table 2: Feed intake, growth performance and feed conversion ratio of lambs fed diet supplemented with seaweed* (mean±SE, n=6)

Parameters	Diet A	Diet B	Diet C	P value
Daily feed intake (kg)	01.35±0.01	01.33±0.01	01.34±0.01	0.3964
Daily weight gain (g)	243.3±8.0	228.3±8.0	236.7±8.0	0.4336
Feed conversion ratio	05.56±0.88	05.83±0.88	05.68±0.88	0.9412

*Diet A = 0.0% seaweed; Diet B = 3.0% seaweed; Diet C = 5.0% seaweed.

Table 3: Blood constituents of growing lambs fed seaweed supplemented diets* (mean±SE, n=6)

Parameters	Diet A	Diet B	Diet C	P value
PCV (%)	34.08±0.78	31.75±0.67	32.17±0.80	0.0970
Total protein (g/dl)	05.99±0.14	06.23±0.14	06.12±0.13	0.4588
Albumin (g/dl)	02.58±0.03	02.65±0.07	02.67±0.05	0.5071
Globulin (g/dl)	03.41±0.14	03.58±0.19	03.45±0.17	0.5409
Glucose (mg/dl)	78.83±6.15	85.91±3.39	84.66±1.99	0.4664
Cholesterol (mg/dl)	59.09±8.23	68.25±1.60	60.28±2.05	0.4058
Total lipids (mg/dl)	223.6±11.85	254.2±13.90	266.1±11.86	0.0978
Na (mmol/l)	112.5±3.84	121.4±0.97	114.1±3.65	0.1488
K (mmol/l)	05.09±0.13[a]	05.99±0.24[b]	06.08±0.15[b]	0.0063
Osmolality (mosmol/l)	297.1±1.89	294.1±4.16	294.4±5.82	0.8658
TAC (mmol/l)	01.70±0.03	01.72±0.08	01.71±0.13	0.9930

*Diet A = 0.0% seaweed; Diet B = 3.0% seaweed; Diet C = 5.0% seaweed; [a, b] Mean values within the same row bearing different superscripts are significantly different at P<0.05.

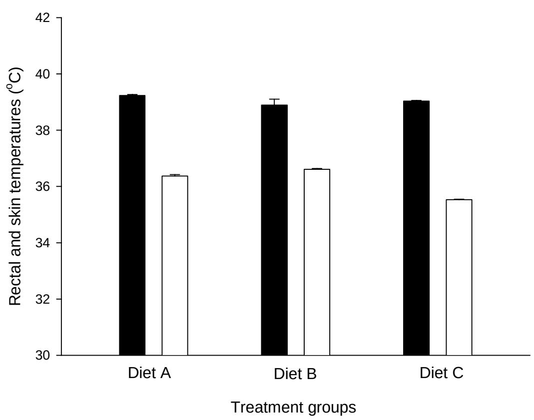

Fig. 1: Means (±SE) of rectal (Black bar) and skin (White bar) temperatures for lambs fed on diets supplemented with different levels of seaweed (Diet A = 0.0% seaweed; Diet B = 3.0% seaweed; Diet C = 5.0% seaweed).

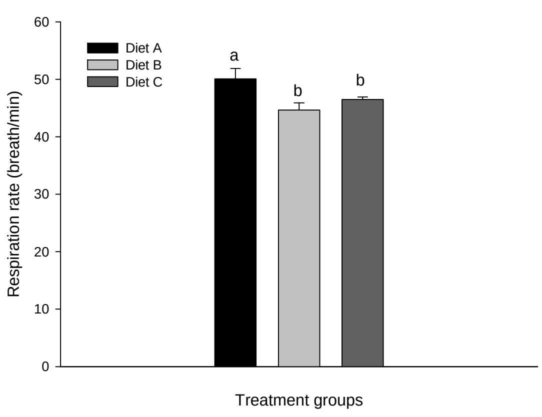

Fig. 2: Means (±SE) of respiration rate (RR) for lambs fed on diets supplemented with different levels of seaweed (Diet A = 0.0% seaweed; Diet B = 3.0% seaweed; Diet C = 5.0% seaweed; means with different letters are significantly different at P<0.05).

effect (P>0.05) on average rectal and skin temperatures (Fig. 1). However, dietary seaweed inclusion had significantly (P<0.05) reduced the respiratory rate compared with no seaweed (Fig. 2).

Blood composition and antioxidant capacity: Blood packed cell volume, serum total protein, glucose, cholesterol and total lipid concentrations did not differ significantly (P>0.05) due to dietary inclusion of seaweed (Table 3). Moreover, the blood antioxidant capacity, osmolality and sodium concentrations were not affected by seaweed inclusion in the diets of lambs. However, serum potassium concentration was significantly (P<0.05) increased in lambs fed on diet supplemented with both concentrations of seaweed compared to no seaweed supplementation (Table 3).

DISCUSSION

Temperature humidity index (THI) of 72.96 is considered as a threshold for extreme severe heat stress in sheep (Papanastasiou *et al.*, 2012). In the present study, the average THI was higher than 72.96 (73.3). Therefore, it can be claimed that growing Naimey lambs were exposed to severe heat stress during this study.

The findings that seaweed inclusion in the diet had no effect on daily feed intake, body weight gain or feed conversion efficiency are consistent with previous reports on goat (Yates *et al.*, 2010) and dairy cows (Pompeu *et al.*, 2011). In contrast, Kellogg et al. (2006) found that brown seaweed inclusion in the diet of dairy cows had increased milk production during heat stress; however, the average maximum T_a was higher than that observed in the study of Pompeu *et al.* (2011), which may explain the benefit of seaweed in reducing heat stress.

The results showing that dietary inclusion of seaweed had no effects on rectal temperature (T_r) or skin temperature (T_{sk}) of growing lambs in the current study are supported by previous observations derived from dietary inclusion of brown seaweed to dairy cows exposed to heat stress (Cvetkovic *et al.*, 2005; Pompeu *et al.*, 2011). In contrast, lambs fed 2% brown seaweed and exposed to T_a reaching 32.0°C, during transportation in a trailer, had lower maximum and average ear canal temperatures (Archer *et al.*, 2007); however, the lambs in that study were exposed to two stressors (heat and transportation). Similar to the finding of the present study, reduction of respiratory rate due to seaweed feeding has also been reported in beef steers (Spiers *et al.*, 2004; Williams *et al.*, 2009). In contrast, dietary supplementation of seaweed had no effect on respiratory rate for cows during the summer season (Cvetkovic *et al.*, 2005; Pompeu *et al.*, 2011). This suggests that feeding seaweed for some period before the event of heat stress, as it was the case in this study and that of Williams *et al.* (2009), may improve its benefits. In sheep, panting is the major evaporatory heat loss mechanism and respiratory frequencies tend to follow closely the heat loss by evaporation (Marai *et al.*, 2007). This physiological adjustment is essential to maintain normal body temperature and to prevent hyperthermia (Lowe *et al.*, 2001; Baumgard and Rhoads, 2013). Hence, the observed lower respiration rate in seaweed-supplemented lambs indicates that they were less stressed compared to control lambs which were not supplemented with seaweed.

In the current study, supplementation of seaweed to the diet of growing lambs had no effect on PCV, blood antioxidant capacity and serum metabolites and sodium concentrations. However, it had significantly increased serum potassium concentration. In contrast, seaweed has been reported to reduce blood glucose level in overweight subjects (Talbott *et al.,* 2012), total cholesterol and triglycerides in hypercholesterolemia rats (Wresdiyati *et al.,* 2008), and to increase antioxidant capacity of blood in lambs grazed on infected *Tall fescue* (Fike *et al*., 2001) and liver injured rats (Wong *et al.,* 2000). These, contradictory findings may be related to the health status of the experimental subjects. The observed increase in serum potassium concentration in the current study could be related to the reported increase in intestinal absorption of potassium released from the seaweed preparation (Krotkiewski *et al.,* 1991).

The conflicting findings concerning the effect of dietary inclusion of seaweed on livestock performance and thermo-physiology under heat stress may be related to the different types and concentrations of seaweed. Moreover, it could be related to the different species and types (dairy vs. beef) of livestock due to the different levels of metabolic heat production and adaptability to heat stress.

Conclusion: Dietary inclusion of seaweed (*Ulva lactuca*) at the level of 3 or 5% to growing lambs reared under heat stress conditions did not show any beneficial effect on feed intake, body weight gain or feed conversion efficiency. Further, rectal and skin temperatures, in addition to blood constituents and antioxidant capacity were not affected by seaweed supplementation. Therefore, we do not recommend inclusion of seaweed (*Ulva lactuca*) in the diet of growing lambs. Nevertheless, effects of seaweed supplementation on thermo-physiology and reproductive performance of adult sheep deserve to be investigated.

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