Effects of Dietary Protein Sources and Levels on Heat Production and Thermoregulatory Responses of Sheep Exposed to a High Ambient Temperature

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ABSTRACT : Four Suffolk ewes were used in Latin Square switch over design to study the effects of varying levels and sources of protein on heat production and thermoregulatory responses at daytime high (33℃) temperature. They were fed Italian ryegrass hay supplemented with fishmeal and/or urea, providing three different levels of crude protein (CP) (low/unsupplemented: 7.9, medium: 11.6, and high: 15.8%) at 1.5×maintenance. Feeds were distributed at 0900 (30%) and 1700 (70%). Urea diet caused higher heat production and increased vaginal temperature compared to fishmeal and fishmeal-urea mix diets. Time spent standing, skin temperature and respiration rate of sheep fed urea were similar with those of sheep fed fishmeal. Sheep fed diet with low CP level had higher heat production, increased vaginal and skin temperature than sheep fed diet with medium CP content. Sheep on high CP diet produced significantly more heat than sheep fed medium CP diets. Their vaginal temperatures were similar with those of sheep fed medium CP diet but lower than those of sheep fed low CP diet. Respiration rates of sheep and time spent by them for standing on all diets did not differ significantly. These results suggest that urea is inferior protein supplement for thermoregulation of animal at hot environment, as it induced higher heat production than fishmeal and fishmeal-urea mix. Thermoregulatory response on fishmeal-urea mix diet was similar to fishmeal diet. Increasing CP of the diet from low to medium gives advantage for thermoregulation of animal. Increasing CP further to high level was not beneficial as it resulted in the responses of sheep similar to those on low protein diet. *(Asian-Aus. J. Anim. Sci. 2000. Vol. 13, No. 11 : 1523-1528)*

Key Words : Protein Sources, Protein Levels, Heat Production, Thermoregulatory Responses, Sheep, High Ambient Temperature

INTRODUCTION

Most of forages grown in hot regions are low in quality, indicated mainly by high fiber and low protein content. Inadequate supply of protein reduces the efficiency of ME utilization, both for maintenance and for production (Shirley, 1986). In addition, under hot environment, as the body temperature of animal increases above that of normal for thermoneutral conditions, the metabolic rate, maintenance energy requirements and protein catabolism may increase (Blaxter, 1962). This suggests that animal in hot environments is easily subjected to protein deficiency.

Protein is an expensive nutrient, hence, its optimum utilization is necessary. Information concerning heat production and thermoregulatory responses in sheep as affected by dietary protein, especially under hot environment, which may influence animal production through the level of feed intake and efficient feed utilization, is scanty. The objective of the present experiment was to examine the effect of different protein sources and levels on heat production and thermoregulatory responses of sheep exposed to a hot ambient temperature.

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MATERIALS AND METHODS

Animals and diets

Four 15 months old well trained Suffolk ewes (35.4±0.6 kg) with about 11 cm fleece length were used and kept in individual cage (125×60 cm) in a controlled temperature room. Room temperature was set high at the daytime and low at night-time by switching control button to 23 and 33℃ at 1600 and 0800, respectively. Lighting schedule of 12L:12D was imposed between 0700 and 1900 (L=105 lux) and 1900 and 0700 (D=8 lux) and maintained throughout the experiment.

During the study, animals were fed Italian ryegrass hay-based diets supplemented with fishmeal and/or urea. The experimental diets are shown in table 1. Diets 2, 3, and 4 contained urea (U), fishmeal+urea (FU), and fishmeal (FM) as protein sources respectively, and had similar protein level. These were used for analyzing the effects of protein source. Diet 3, 1 and 5 containing medium, low and high levels of crude protein, respectively, were used for analyzing the effects of protein level. Diet 1 was unsupplemented diet (control), while diet 3 and 5 have similar dietary crude protein proportion provided by urea and fishmeal, respectively, i.e. 27:23. Adaptation to each diet was given for 6 days at maintenance level of 35 kg ewes. During parameter measurements, on 7th day, feeds were given at 1.5 of maintenance level (NRC, 1985a) distributed at 0900 and 1700 for 30 and 70% of daily meal, respectively. Animal had free access to drinking (tap) water, except during in the hood where it was given water twice a day at 0800 and 2100 for about 5 min each, during when the inlet fresh air was measured. Time spent for standing (TSS), heat production (HP), vaginal temperature (Tv) and skin temperature (Ts) were measured for 23h. Respiration rate (RR) was measured at 0800 and 1600, one hour before feeding. Methods of measurements of HP, Tv, TSS, and RR were similar to the previous experiment (Sudarman and Ito, 2000). Skin temperatures (Ts) were measured in three different locations of upper part of the body (withers, back and rump) using thermocouple. The locations were clipped and rubbed with tissue paper to remove grease and sweat. Thermocouples adhered on a piece of surgical white tape (Nichiban, W25-9) and pasted with a clear bond (Konishi bond G17) were attached firmly to the skin by pressing with finger.

Data were subjected to the analysis of variance (ANOVA) for Latin Square Design switch over, treatment means were compared by LSD (Steel and Torrie, 1980) and significance was declared at $p \leq 0.05$.

RESULTS AND DISCUSSION

Heat production

Protein source and level significantly affected total 1 day heat production (HP) of sheep. The influence of different levels of dietary protein on heat production and thermoregulatory responses of sheep are summarized in table 2. Sheep fed low CP level (control) had significantly higher and similar total 1 day HP than those fed medium and with those fed high, respectively. Heat production of sheep fed high CP level was higher than that of those fed medium. When the data were analyzed in different periods of ambient temperature, the significant difference was observed only between HP of medium and high protein groups during cool period (table 2). This difference due to the decreasing rate of HP of medium group was greater than the other two groups. Heat production during daytime did not differ on varying levels of protein which may be the compound effect of diet and hot temperature. In general, HP of sheep fed medium CP level was the lowest follow by those fed low and high, respectively.

According to Leng (1989), in hot environment more calorigenic substrate is available for maintenance and production. However, such substrate cannot be utilized for production due to the limitation of essential amino acids availability. He further argued that in case of the excess substrate is oxidized by the ruminant, it will produce more heat. Lengs argument supports the present results that sheep fed low protein level or unsupplemented diet had higher heat production than the other two groups of diet.

In contrast, when protein is provided in excess, it is catabolized and utilized as an energy source (Blaxter, 1962). Utilizing protein as an energy source is not economical since it produces high heat. In addition, an excessive protein N intake has to be excreted and is energetically costly process. In cows it was estimated to be 30 kJ per g protein N (Oldham and Smith, 1982). This expense of energy is likely to contribute to the total heat production and in hot environment it is certainly bound to increase heat load to the animal. As the rule of thumb, therefore, it is suggested to give protein in optimum level. But this level needs to be clarified, since it depends on other factors, mainly energy content of the diet. Results of the present study shows that the medium level of CP (11.6%) in sheep diet prevents a greater heat load to the animal.

The influences of different sources of dietary protein on heat production and thermoregulatory responses of sheep are summarized in table 3. Sheep fed U diet had significantly higher total 1 day HP than those fed FM and FU diet. However, HP on later two diets was not different. During cool period, sheep fed urea diet had significantly higher HP than the other two groups (table 3) and this part greatly contributed to cause significant differences in total 1 day HP. During hot daytime, sheep on diets with all protein sources had similar but high HP compared to

Table 1. Ingredient and nutritional composition of experimental diets

Diet	1	2	3	4	5
Ingredient composition (% dry matter)[1]					
Italian ryegrass hay	86.1	85.0	85.0	84.9	83.6
Corn flaked	12.9	12.7	10.8	7.6	7.8
Fish meal	-	-	2.4	6.5	6.0
Urea	-	1.3	0.8	-	1.6
Premix	1.0	1.0	1.0	1.0	1.0
Chemical composition					
Dry matter (%)	86.2	86.3	86.4	86.5	86.6
Composition of dry matter					
ME (kcal/kg)	2,293	2,263	2,261	2,260	2,226
Crude protein (%)	7.9	11.6	11.6	11.6	15.8
Crude fiber (%)	26.9	26.5	26.5	26.5	26.1
P:E ratio[2]	34.5	51.3	51.3	51.3	69.9

[1] Calculated values based on AFFRC (1995).
[2] Calculated crude protein (g) relative to ME (Mcal/kg).

Table 2. Heat production and thermoregulatory responses of sheep influenced by different crude protein levels in the diet

Measurements[1]		Period	Diets[2]		
			High	Medium	Low
HP	(kJ/kg$^{0.75}$/h)	Hot	24.5 ± 0.1	23.3 ± 0.5	23.5 ± 0.3
	(kJ/kg$^{0.75}$/h)	Cool	21.3 ± 0.2^{b}	19.7 ± 0.3^{a}	20.9 ± 0.4^{ab}
	(kJ/kg$^{0.75}$/23h)	1 day	515.2 ± 3.9^{b}	481.4 ± 5.9^{a}	500.9 ± 4.1^{b}
HP_{cor}	(kJ/kg$^{0.75}$/23h)	1 day	497.5 ± 2.6^{b}	461.3 ± 5.5^{a}	483.8 ± 3.3^{b}
TV (℃)		Hot	40.4 ± 0.03	40.3 ± 0.02	40.4 ± 0.04
		Cool	39.3 ± 0.03^{a}	39.4 ± 0.02^{a}	39.6 ± 0.02^{b}
Ts (℃)		Hot	39.3 ± 0.06^{b}	38.9 ± 0.08^{a}	39.3 ± 0.07^{b}
		Cool	37.3 ± 0.14^{b}	36.8 ± 0.11^{a}	37.9 ± 0.10^{c}
RR (breath/min)		Hot	207 ± 5	207 ± 7	225 ± 14
		Cool	61 ± 5	50 ± 7	55 ± 8
TSS (min)		Hot	395 ± 24	457 ± 6	407 ± 7
		Cool	640 ± 55	710 ± 25	587 ± 46

[a,b,c] Values (mean ± SE) within the same row with different superscript differ significantly ($p \leq 0.05$).

[1] HP: heat production; HP_{cor}: heat production corrected for the energy cost of standing; Tv: vaginal temperature; Ts: skin temperature; RR: respiration rate; TSS: time spent for standing.

[2] High: 15.8% CP (diet 5); Medium: 11.6% CP (diet 3); Low: 7.9% CP (diet 1).

Table 3. Heat production and thermoregulatory responses of sheep influenced by different crude protein sources in the diet

Measurements[1]		Period	Diets[2]		
			FM	FU	U
HP	(kJ/kg$^{0.75}$/h)	Hot	23.1 ± 0.3	23.3 ± 0.5	23.6 ± 0.4
	(kJ/kg$^{0.75}$/h)	Cool	19.6 ± 0.3^{a}	19.7 ± 0.3^{a}	21.6 ± 0.4^{b}
	(kJ/kg$^{0.75}$/23h)	1 day	478.8 ± 2.8^{a}	481.4 ± 5.9^{a}	512.2 ± 9.0^{b}
HP_{cor}	(kJ/kg$^{0.75}$/23h)	1 day	461.6 ± 2.0^{a}	461.3 ± 5.5^{a}	493.2 ± 8.3^{b}
TV (℃)		Hot	40.2 ± 0.02^{a}	40.3 ± 0.02^{ab}	40.5 ± 0.03^{b}
		Cool	39.5 ± 0.03	39.4 ± 0.02	39.6 ± 0.06
Ts (℃)		Hot	39.1 ± 0.11	38.9 ± 0.08	39.2 ± 0.12^{b}
		Cool	37.7 ± 0.15^{b}	36.8 ± 0.11^{a}	37.4 ± 0.12^{b}
RR (breath/min)		Hot	241 ± 10	207 ± 7	238 ± 2
		Cool	98 ± 10	50 ± 7	59 ± 8
TSS (min)		Hot	392 ± 12^{a}	457 ± 6^{b}	405 ± 9^{a}
		Cool	610 ± 44^{a}	710 ± 25^{b}	706 ± 39^{b}

[a,b] Values (mean ± SE) within the same row with different superscript differ significantly ($p \leq 0.05$).

[1] HP: heat production; HP_{cor}: heat production corrected for the energy cost of standing; Tv: vaginal temperature; Ts: skin temperature; RR: respiration rate; TSS: time spent for standing.

[2] High: 15.8% CP (diet 4); Medium: 11.6% CP (diet 3); Low: 7.9% CP (diet 2).

nighttime HP. The high HP of sheep fed urea diet may be due to urea as a nitrogen source is highly degradable in the rumen. Thus host animal received only limited amount of CP, as explained above that animal fed a diet of low protein content resulted in higher HP.

Urea is highly degraded and is converted in the rumen to ammonia. It is utilized by microbes or is absorbed across the rumen wall. In the liver it is converted back to urea, a mechanism to avoid to reach its toxic level in the blood. This conversion requires approximately 12 kcal/g nitrogen (Tyrrell et al., 1970). This cost, certainly, will be greater with a higher supply N in the diet. It may explain why sheep fed urea had higher heat production than the other two groups.

Fishmeal besides rich in CP is degraded slowly in the rumen (ARC, 1980; NRC, 1985b) and is often called as by pass protein. Ruminal CP degradation of fishmeal ranges from 30 to 70% (Hussein and Jordan, 1991). Therefore, much of its protein is available postruminally for animal. In hot environment, calorigenic substrates cannot be converted into useful products (milk, meat, etc.) when protein supply is in adequate (Leng, 1989). Balancing the diet (protein: energy ratio) with undegradable protein source such as fishmeal results in optimum rumen fermentation as a consequence of which low heat is produced as observed in the study.

In feeding trial, Cotrill and Osbourn (1977) reported that young castrated calves grew faster when fed corn silage-based diets supplemented with fishmeal than with urea. Similarly Oldham · and Smith (1982) reported that young growing heifers had greater daily gain when fed diets supplemented with fishmeal than those fed diets supplemented with urea or soybean meal. In lambs, Hovell et al. (1983) found that live-weight gains of lambs fed NaOH-treated straw increased when provided with a fishmeal supplement rather than with urea. These responses were attributed to increased amino acids flow to the small intestine and/or improved ruminal fermentation (Cotrill and Osbourn, 1977). It may, therefore, be concluded that fishmeal is utilized efficiently for protein synthesis rather than urea since it supplies more complete amino acids that can be absorbed in intestine and utilized by animal. Inefficient utilization of urea may lead to high heat production as observed in the present experiment. In addition, it has been reported that fishmeal supplementation increases digestion of fibrous components of the diet (Hussein et al., 1991; McAllan, 1991; Stritzler et al., 1992; Dixon et al., 1999) due to the provision of substrates by the fishmeal for microbes (Dixon et al., 1999).

There were significant differences in HP in different temperature periods. On hourly basis, HP during hot-period was significantly higher than that during cool-period (table 2 and 3), although feeding times were distributed in each of these periods. The differences in HP between hot and cool period for sheep fed urea, fishmeal and fishmeal-urea were to the tune of 2.0, 3.5 and 3.6 $kJ/kg^{0.75}$ per h, respectively. While the differences in HP between hot and cool for sheep fed high, medium and low protein levels were to the tune of 3.2, 3.6 and 2.6 $kJ/kg^{0.75}$ per h, respectively. This indicates that sheep fed urea or low CP diet would still receive heat burden even during night-cool period. This is likely to be more deleterious when temperature difference between day and night is little, because of difficulty to dissipate extra heat load.

Skin temperature

Compared to skin temperature (Ts) at cool period, it was higher at hot period. Increase in skin temperature at hot environment is a physiological mechanism in order to increase heat dissipation, because a great temperature gradient is favorable for transferring heat from body surface to environment. This is achieved by increasing blood flow which carries heat from internal body to peripheral tissues. Increased ambient temperature and effect of feeding cause skin temperature of sheep to increase.

Skin temperature of sheep fed medium CP level was lower than that of those fed low or high during both hot and cool periods. During hot period, Ts of two later groups was not significantly different, but during cool period, sheep fed low level of protein had significantly higher Ts than those fed high level of protein (table 2). Sheep fed low protein level experienced a greater heat load as indicated by their higher Tv. This implied that the higher Ts of this group was a physiological necessity to dissipate the extra heat burden. Thus, dissipation of heat via skin in animals fed low protein diet was more effective during cool as indicated by their higher Ts.

Skin temperature of sheep fed different sources of protein was not significantly different during hot period. During cool period, however, sheep fed urea and fishmeal diets had significantly higher Ts than sheep fed FU diet (table 3). Heat production of urea fed animals was higher than those of the others and in order body temperature not to increase greatly these animals should be able to do a better heat loss. Sheep fed FM had similar HP but higher Ts than sheep fed FU. Standing time data of these two groups indicate that of sheep fed FU was standing for longer time than those fed fishmeal diet. This probably shows that amount of heat loss via skin per unit time of sheep fed FM is greater than those fed FU.

Respiration rate

Respiration rates of sheep fed different protein levels and sources did not differ significantly during both hot and cool periods (tables 2 and 3). Respiration, as one of the avenues for evaporative heat loss, usually will be effectively operated when ambient temperature approach or exceed body temperature. However, in this condition sensible heat loss is usually no longer effective. In experiment using closely clipped sheep, Blaxter et al. (1959) found decreased sensible heat loss as environmental temperature was increased. While evaporative heat loss was constant below environmental temperatures of 23℃, it was increased above this. They also found that at environmental temperatures of about 30℃, the curves of both heat loss partitions cross each other indicating that at environmental temperatures above 30℃, latent (evaporative) heat loss become more important than sensible heat loss.

Vaginal temperature

Sources as well as levels of protein in the diet significantly affected vaginal temperature (Tv) of sheep. At different CP levels, vaginal temperatures of sheep during hot period were not different, however, during cool period sheep fed low level of protein had significantly higher Tv than those fed medium and high levels. Vaginal temperatures of later two groups of sheep did not differ significantly. In general, Tv of sheep decreased as the level of CP in the diet increased. The present result was not in agreement with Ahmed and Abdellatif (1995) who fed rams with groundnut cake as a main source of CP at different proportion so as to obtain different CP levels of experimental diets. They reported that though the temperature measured in the morning (0800 h) was similar, rectal temperature measured in the afternoon (1300 h) was increased as the level of CP in the diet increased from 9.8 (39.5℃) to 12.8% (39.9℃). However, there was no further increase in rectal temperature when the CP level in the diet increased to 14.7%. This discrepancy may be due to the difference in source of protein used, since the difference in source of protein cause difference in the body temperature response as shown in the present result. However, the increased CP content in the study of Ahmed and Abdellatief (1995) seemed to be accompanied by the increased energy content of the diets and this is likely to be the major factor affecting the higher rectal temperature of sheep fed higher CP diet.

During hot period, Tv of sheep fed urea diet was significantly higher than that of sheep fed fishmeal but was not different from those fed fishmeal-urea (table 3). The higher Tv of sheep fed urea diet was possibly caused by a higher heat production on this diet. During cool period, Tv of sheep was not significantly different.

Time spent standing

By standing, the surface of animal body become broader and therefore during standing greater heat loss by sensible means (via skin) occurred than during lying. Hence, it is a normal response that animal will stand longer when they receive a great heat burden in order to easily dissipate heat, although standing requires extra energetic cost over lying. In sheep and cattle this cost accounts 0.42 kJ/kg per hour (ARC, 1980). Standing time did not change significantly daily HP of sheep fed different sources and levels of CP (tables 2 and 3).

During hot daytime, source of protein significantly affected standing time, but level did not (tables 2 and 3). Sheep fed FU diet had significantly higher standing time than those fed FM diet, but it did not differ significantly from those fed U diet. Standing time on FM and U diet similar. As sensible heat loss during standing is more, sheep fed FU diet with higher standing time may have more heat loss than the other two groups. But in fact Ts of FU is lower than those of the other two groups. This implies that heat loss via skin of all groups was probably not greatly different. In addition, the sheep fed FU diet did not need to dissipate more heat since they produced low HP.

About 60% of 1 day standing time was spent during the cool period as cool period was longer (16 h) than hot period (8 h), however, the intensity of standing (min/h) was greater during hot period, i.e., 51 vs. 43 min/h for hot and cool period. This is clearly shows that the animals tried to dissipate extra heat during hot-day by making body surface broader.

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