**REGULAR ARTICLES** 



# Ideal proportion of roughage and concentrate for Malpura ewes to adapt and reproduce in a semi-arid tropical environment

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Abstract The study was designed to identify the most appropriate roughage to concentrate ratio for Malpura ewes under semi-arid tropical environments. The study was conducted for a period of 35 days and included 30 (2 years old) non-pregnant Malpura ewes weighing between 30 and 35 kg. Estrus synchronization was carried out in all the animals using indigenously developed intravaginal sponges impregnated with progesterone. The ewes were randomly divided into three groups (n=10/group) namely R50 (n=10; roughage to concentrate, 50:50), R60 (*n*=10; roughage to concentrate, 60:40), and R70 (n=10; roughage to concentrate, 70:30). Individual feed and water intake was recorded on a daily basis throughout the course of the study. Growth variables, physiological responses, blood metabolites, and endocrine responses were estimated at weekly intervals. Results of the study indicated that nutritional treatment significantly influenced growth variables including body weight (p < 0.01), BCS (p < 0.01), feed intake (p < 0.05), and water intake (p < 0.01) among the different treatment groups. The highest growth variables were recorded in R50 whereas the lowest were in R70. Both respiration rate (p < 0.01) and plasma estradiol levels (p < 0.05) also showed a similar trend as that of the growth variables. However, growth hormone (p < 0.01), T<sub>3</sub> (p < 0.01), Hb (p < 0.01), glucose

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(p<0.05), and estrus duration (p<0.05) demonstrated a reverse trend with highest values recorded in R70 and lowest in R50. However, the nutritional treatment did not influence pulse rate, rectal temperature, T<sub>4</sub>, progesterone, PCV, total cholesterol, total protein, albumin, estrus%, and estrus cycle length. Since the additional concentrate supplementation in R50 and R60 did not improve the production variables, it can be concluded that providing 70 % roughage and 30 % concentrate could be a more appropriate and economically feasible ration composition for Malpura ewes reared in semi-arid tropical environments.

**Keywords** Adaptation · Roughage to concentrate ratio · Malpura ewes · Reproduction · Sheep

# Introduction

Livestock in general and small ruminants (sheep and goat) in particular play an important role in the rural economy of India where small, marginal, and landless farmers are the main beneficiaries (Maurya et al. 2010). Sheep raised in these areas are generally confronted with severe nutritional deficits during the periods of feed scarcity which in turn exacerbates disease and consequently leads to low productive and reproductive performances. Therefore, under the prevailing circumstances, exploration, and evaluation of new feed resources and their efficient utilization should be a continual process to overcome this problem of feed scarcity. Roughage to concentrate ratio is an important factor to be considered for improving feed efficiency (Liu et al. 2005).

The economically important production traits which are difficult to measure in many sheep flock are strongly influenced by management decisions (Notter 2000). The interaction between nutrition and reproduction has direct implication

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for productivity (Sejian et al. 2010). Many reviews have been published on the influence of optimum nutrition on reproduction in ruminants with special reference to sheep (Martin et al. 2004; Scaramuzzi et al. 2006). However, such studies were mostly on temperate breeds and may not be applicable to breeds in tropical arid and semi-arid conditions.

In arid and semi-arid areas, sheep are one of the primary livelihood securities of most farmers. Hence, the farmers need to be more informed about the importance of appropriate ration composition that would be economically feasible to optimize production in sheep. However, current information regarding nutritional requirements of indigenous breeds for optimum production and fertility are limited, and available information is mostly extrapolated from studies using exotic breeds. A basic understanding of nutrition and dietary requirements under local environmental conditions is therefore required before one can consider and adopt possible improvements.

Small ruminants are critical to the development of sustainable and environmentally sound production systems primarily due to low investment and less contribution to greenhouse gases per unit of feed consumed when compared to large ruminants (Naqvi et al. 2013). Small ruminants in particular offer many benefits to millions of farmers in the semi-arid tropical environment. These animals are integral to rural livelihoods and culture, providing wool, meat, milk, cheese, and easily accessible income (Marai et al. 2007; Naqvi et al. 2013). The socioeconomic role of sheep and goat in communities living in semi-arid regions will be maintained over the forthcoming decades. Therefore, efforts should be intensified to improve productive and reproductive performances of these animals using simple and low-cost strategies. This study aims to identify the most appropriate cost-effective roughage to concentrate rations for sheep reared under semi-arid tropical conditions.

# Materials and methods

# Location

The study was conducted after obtaining approval from the Institutional Ethical Committee. The experiment was carried out at the Central Sheep and Wool Research Institute farm, Rajasthan, India, which is located in the semi-arid region of the country at longitude of 75° 28' E, latitude of 26° 26' N, and altitude of 320 m above mean sea level. The prevailing ambient temperature ranges from 6 to 46 °C, and the relative humidity (RH) ranges from 20 to 85 %. The annual rainfall in this area ranges from 200 to 400 mm with an erratic distribution throughout the year. The temperature and RH ranged from 31 to 44 °C and 15 to 43 %, respectively, during the

study period. The experiment was carried out during February-March.

## Animals

Malpura is a triple purpose (meat, wool, milk) sheep breed native of the arid and semi-arid areas of Western tropical India. The study was conducted using 30 (2 years old) non-pregnant Malpura ewes weighing between 30 and 35 kg. The animals were housed in well-ventilated sheds made up of asbestos roofing and maintained under proper hygienic conditions. Prophylactic measures against sheep diseases like sheep pox, peste des petits ruminants, enterotoxaemia, and endoparasitic and ectoparasitic infestations were carried out as prescribed by the health calendar of the institute.

# **Estrus synchronization**

In order to study estrus cyclicity in the ewes, estrus synchronization was carried out in all the animals using indigenously developed intravaginal sponges impregnated with progesterone (Naqvi et al. 2001). The synchronization procedure started on the first day of the 35-day experiment. Each vaginal sponge was imbibed with 0.35 gm progesterone (CDH Laboratory reagent, New Delhi). The sponge was inserted and kept in situ in the vagina for a period of 12 days. On the day of sponge removal, ewes were given a single dose of equine chorionic gonadotrophin (Folligon, Intervet International, Netherlands) at 200 IU/ewe intramuscularly. Estrus was detected 1 day after sponge removal. Estrus in each ewe was detected by parading aproned rams of proven vigor for 30 min at every 6-h intervals (6:00 AM, 12:00 noon, 6:00 PM, and 12:00 midnight). The estrus duration was calculated from the first time the ewes allowed the aproned ram to mount them until the end of receptivity in the ewes. Estrus cycle length was calculated from the day of sponge insertion until the estrus ended.

#### **Experimental procedure**

The study was conducted for a period of 35 days. The ewes were randomly divided into three groups namely R50 (n=10; roughage to concentrate, 50:50), R60 (n=10; roughage to concentrate, 60:40), and R70 (n=10; roughage to concentrate, 70:30). Allocation of animals to each group was carried out to ensure matching body weight (BW) and body condition score (BCS) between the groups. The animals had ad libitum access to feed and water. At 8:00 PM, the residues of both feed and water were recorded. This feeding pattern was supported by providing 24 h lighting in the animal shed. The composition of the diet includes roughage (*Cenchrus ciliaris*) and concentrate mixture (barley 65 %, groundnut cake 32 %, mineral mixture 3 %, common salt 1 %). The roughage was provided as dry

fodder in chopped form. The crude protein content of roughage and concentrate was 8.4 and 14 %, respectively. The neutral detergent fiber content of roughage and concentrate was 72.6 and 48.5 %, respectively. Similarly, the digestible energy of roughage and concentrate was 7.37 and 12.1 MJ, respectively. The metabolizable energy of roughage and concentrate was 6.05 and 10.1 MJ, respectively. The crude protein (CP) of the feed sample was determined by Kjeldahl technique (AOAC 1995). Neutral detergent fiber (NDF) was determined by Van Soest et al. (1991) without sodium sulphite or amylase, whereas acid detergent fiber (ADF) and acid detergent lignin (ADL) were determined according to the method as described by Robertson and Van Soest (1981). The diet was supplied as a total mixture of roughage and concentrate according to different proportion for each group. Table 1 describes the chemical composition, energy, and nutrient content of the diet provided to the animals. The animals were housed in a wellventilated shed in east west orientation with the dimensions  $9.1 \times 3.7 \times 2.4$  m for length, width, and height, respectively. The area allocated to each animal was 4.2 m<sup>2</sup>. The roof of the shed was made up of asbestos sheet with wire mesh sides and clay sand floor. The shed had a stocking density of 20 animals. The animals were acclimatized to different feeding proportions as per their treatment groups 15 days prior to the start of the trial. Individual feed and water intake was recorded on daily basis for all groups.

#### Blood collection and plasma separation

Blood samples were collected on days 0, 7, 14, 21, 28, and 35 after feeding at 11:00 AM from the external jugular vein in

tubes with heparin anticoagulant. Blood samples were divided into two aliquots. One aliquot was used for estimation of hemoglobin (Hb) concentration and packed cell volume (PCV) while the other was used for plasma separation. Plasma was separated by centrifugation at 3,500 rpm for 15 min at room temperature. The plasma was then divided into aliquots in microcentrifuge tubes and kept frozen at -20 °C till further analysis. Plasma samples were used to determine biochemical and endocrine variables.

# Variables studied

Growth variables such as body weight (BW) and body condition score (BCS) (1-5 point scale) were recorded at 6 h and 30 min before feeding and watering at weekly interval. Body weight was recorded by subjecting the animals to stand individually on the weighing machine (Balance Avery, Bombay, India) with a maximum capacity of 50 kg. The BCS method described by Russel et al. (1969) was used to allocate animals in a scale of 1–5. BCS was assessed by careful palpation of the spinous and transverse process in the loin area, immediately behind the last rib. Feed intake (FI) and water intake (WI) was recorded on daily basis. The animals were offered both feed and water on individual basis in the morning at 8:00 AM. Care was taken to ensure some quantity of feed and water was always left in the feed and water troughs. The following day before feeding and watering, the previous day leftover residues were recorded to know the daily feed and water intake in individual animals. Physiological responses such as respiration rate (RR), pulse rate (PR), and rectal temperature (RT) were recorded twice daily (8:00 AM and 14:00 PM) at weekly

Table 1 The chemical composition, energy, and nutrient contents of the diet provided to the animals

| Nutrient contents           | Roughage          | Concentrate   |
|-----------------------------|-------------------|---|
| Ingredients                 | Cenchrus ciliaris | Barley, 650 g/kg; groundnut cake, 320 g/kg; minerals 30 g/kg including 10 g/kg NaCl |
| Dry matter (%)              | 92.7              | 93.3  |
| Crude protein (%)           | 8.4               | 14.0  |
| Ether extract (%)           | 1.9               | 3.2   |
| Neutral detergent fiber (%) | 72.6              | 48.5  |
| Acid detergent fiber (%)    | 46.4              | 9.0   |
| Acid detergent lignin (%)   | 7.9               | 6.23  |
| Digestible energy (MJ)      | 7.37              | 12.1  |
| Metabolizable energy (MJ)   | 6.05              | 10.1  |
|                             |                   |   |

 $TDN/kg=digestible crude protein/kg+digestible carbohydrate/kg+2.25 \times digestible ether extract=0.4$ , where digestible carbohydrate=fiber+nitrogen free extract

DE and ME are calculated as per the following formula:

DE(Mega calories)/kg=.4×4.4=1.76

DE(Mega Joule)/kg=DE(Mega calories)×4.19=1.76×4.19=7.37

ME(Mega Calories)/kg= DE(Mega calories)×0.82=1.76×.82=1.443

ME(Mega Joule)/kg=ME (Mega calories)× 4.19=1.443×4.19=6.047

interval. Respiration rate was recorded based on the flank movements at the paralumbar fossa of the ewes using a stopwatch and represented by number of breaths per minute. Pulse rate was recorded based on the pulsations noted in the femoral artery per unit of time and represented by number of beats per minute. Rectal temperature was recorded using a clinical rectal thermometer and represented in °F per minute.

Blood metabolites such as Hb, PCV, plasma glucose, total cholesterol, total protein, and albumin were estimated at weekly intervals. All biochemical variables were estimated using *Span* diagnostic kits, India, as per standard method (Sejian et al. 2014a, b) using the UV-visible recording spectrophotometer (UV-160A; Shimadzu Corporation, Japan). Reproductive performance such as estrus% (the number of animals in each group exhibiting symptoms of estrus), estrus duration, and estrus cycle length were recorded for two estrus cycles.

Endocrine variables such as growth hormone (GH), thyroxine  $(T_4)$ , tri-iodo-thyronine  $(T_3)$ , estradiol, and progesterone were estimated at weekly intervals. Hormones analyzed were plasma GH (analytical sensitivity, 0.10 mIU/l; the intra-assay and inter-assay coefficient of variations were below or equal to 1.5 and 14 %, respectively), T<sub>3</sub> (analytical sensitivity 0.1 nmol/l; intra-assay and inter-assay coefficient of variations 3.3 and 8.6 %, respectively),  $T_4$  (analytical sensitivity 13 nmol/l; intra-assay and inter-assay coefficient of variations 5.1 and 8.6 %, respectively), estradiol (analytical sensitivity 6 pg/ml; intra-assay and inter-assay coefficient of variations were 5.8 and 9.0 %, respectively), and progesterone (analytical sensitivity 0.05 ng/ml; intra-assay and inter-assay coefficient of variations were 12.1 and 11.2 %, respectively). All hormonal variables were determined by radio immuno assay (RIA) using gamma counter (PC-RIA MAS, Stretec, Germany) by employing RIA kits supplied by Immunotech, France.

#### Data analysis

The data were analyzed by GLM (SPSS 16.0) repeated measures analysis of variance. The analysis took into account the data from the same animals across time. Effect of fixed factors namely treatment (R50, R60, and R70) and days (longitudinal time over which experiment was carried out=day 0, day 7, day 14, day 21, day 28, and day 35) and also interaction of treatment and days were analyzed on the various variables studied. Comparison of means of the different subgroups was made by Duncan's multiple range tests as described by Kramer (1957). Differences in estrus response were evaluated by Chi-square test (Snedecor and Cochran 1994). Differences among groups for estrus duration and estrus cycle length were evaluated by one-way analysis of variance test.

#### Results

# Body weight, body condition score, feed intake, and water intake

A significantly (p<0.01) lower body weight was recorded in R70 and R60 ewes as compared to R50 ewes (Fig. 1.). As the experiment progressed, the body weight of both R50 and R60 significantly (p<0.01) increased, but in R70, it reduced and remained constant for the entire study period (Fig. 1). Furthermore, BCS also was significantly (p<0.01) lower in R70 as compared to R50 and R60 ewes (Table 2). In addition, there were significant (p<0.05) interactions between groups and experimental days for BCS indicating the difference between the groups continued over time. The lowest feed intake (p<0.05) was recorded in R70 as compared to both R50 and R60 (Table 2). Similar trend was observed for water intake (Table 2).

#### **Physiological responses**

Among the physiological responses, different roughage to concentrate ratio had significant effect only on RR (p<0.01) in the morning and afternoon (Table 3). Furthermore, experimental week significantly (p<0.01) influenced RR during morning and afternoon and PR during afternoon and RT during morning (Table 3). In addition, there were significant (p<0.05) interactions between groups and experimental days for RT in the morning indicating the animals trying to cool themselves during night hours in order to cope up with heat stress during day time. This is the common adaptive mechanisms of sheep reared under hot semi-arid environments.

# **Endocrine variables**

The nutritional treatments in the study showed significant (p < 0.05) effect on plasma GH concentration between the groups (Table 4). The highest concentration of plasma GH was recorded in R70 and the lowest in R50 (Table 4). Plasma T<sub>3</sub> was significantly (p < 0.01) higher in R70 (Table 4). The nutritional treatments also had a significant (p < 0.05) effect on plasma estradiol level (Table 4). The highest plasma estradiol concentration was recorded in R50, while the lowest in R70. The nutritional treatment did not significantly influence plasma progesterone levels (Table 4). Different roughage to concentrate ratio altered the thyroid hormone concentrations throughout the study period.

# **Biochemical variables**

The level of Hb was significantly (p<0.01) higher in R70 as compared to R50 and R60 (Table 5). However, PCV did not differ between the groups. Plasma glucose was significantly





duration was recorded in R70 while the shortest was in R60 ewes (Table 6). Furthermore, the length of estrus cycle also did not differ between the groups (Table 6).

# Discussion

# **Reproductive variables**

over time (Table 5).

Estrus response did not differ between the groups (Table 6). Estrus duration showed significant (p < 0.05) changes between the different nutritional treatment groups. The longest estrus

there were significant (p < 0.05) interactions between groups and experimental days for plasma glucose (p < 0.05) and total cholesterol levels (p < 0.05) indicating that variations persisted

Table 2Effect of differentroughage to concentrate ratio onbody condition scoring, feedintake, and water intake ofMalpura ewes

The results obtained on various adaptative and reproductive variables suggest that compared with roughage to concentrate ratio of 50:50 and 60:40, providing 70 % roughage and 30 % concentrate could be the ideal ration composition in semi-arid tropical environment. These findings could be applied to sheep reared under different semi-arid conditions.

| Attributes              | BCS                | FI (DMI gm/w^.75/day) | WI (L/DMI kg/day) |
|-------------------------|--------------------|-----------------------|-------------------|
| μ±SE                    | 3.51±0.03          | 65.96±0.49            | 2.25±0.03         |
| Treatment               | **                 | *                     | **                |
| R50                     | 3.83 <sup>a</sup>  | 67.02 <sup>a</sup>    | 2.47 <sup>a</sup> |
| R60                     | 3.53 <sup>b</sup>  | 66.94 <sup>a</sup>    | 2.17 <sup>b</sup> |
| R70                     | 3.18 °             | 63.93 <sup>b</sup>    | 2.11 <sup>b</sup> |
| Pooled SE for treatment | ±0.06              | ±0.85                 | ±0.05             |
| Days                    | NS                 | **                    | **                |
| 0                       | 3.63 <sup>a</sup>  | 61.68 <sup>d</sup>    | 2.27 <sup>b</sup> |
| 7                       | 3.62 <sup>a</sup>  | 63.48 <sup>cd</sup>   | 2.77 <sup>a</sup> |
| 14                      | 3.57 <sup>ab</sup> | 65.64 <sup>bc</sup>   | 2.60 <sup>a</sup> |
| 21                      | 3.50 <sup>ab</sup> | 68.20 <sup>ab</sup>   | 2.22 <sup>b</sup> |
| 28                      | 3.43 <sup>ab</sup> | 69.43 <sup>a</sup>    | 1.81 <sup>c</sup> |
| 35                      | 3.33 <sup>b</sup>  | 67.35 <sup>ab</sup>   | 1.81 <sup>c</sup> |
| Pooled SE for day       | $\pm 0.08$         | $\pm 1.20$            | $\pm 0.07$        |
| Treatment* day          | **                 | NS                    | NS                |

 $\mu$  indicates the overall mean for the parameter. Means with similar superscripts along the column do not differ significantly (*P*>0.05) from each other. Group I, R to C 50:50. Group II, R to C 60:40. Group III, R to C 70:30. Both body weight and BCS changes are averages of all 35-day values

*BCS* body condition scoring, *FI* feed intake, *WI* water intake, *NS* non-significant \**P*<0.05; \*\**P*<0.01

| Attributes              | RR (morning)        | RR (afternoon)     | PR (morning)       | PR (afternoon)      | RT (morning)        | RT (afternoon)       |
|-------------------------|---------------------|--------------------|--------------------|---------------------|---------------------|----------------------|
| μ±SE                    | 26.43±.43           | 46.69±.84          | 57.78±.44          | 64.63±0.49          | 100.11±0.12         | 101.75±0.04          |
| Treatment               | **                  | **                 | NS                 | NS                  | NS                  | NS                   |
| R50                     | 28.20 <sup>a</sup>  | 50.13 <sup>a</sup> | 57.97 <sup>a</sup> | 65.27 <sup>a</sup>  | 99.92 <sup>a</sup>  | 101.74 <sup>a</sup>  |
| R60                     | 27.00 <sup>a</sup>  | 47.07 <sup>a</sup> | 58.17 <sup>a</sup> | 64.47 <sup>a</sup>  | 100.43 <sup>a</sup> | 101.72 <sup>a</sup>  |
| R70                     | 24.10 <sup>b</sup>  | 42.88 <sup>b</sup> | 57.20 <sup>a</sup> | 64.17 <sup>a</sup>  | 99.99 <sup>a</sup>  | 101.79 <sup>a</sup>  |
| Pooled SE for treatment | ±.74                | ±1.45              | ±.76               | ±0.84               | ±0.21               | $\pm 0.06$           |
| Days                    | **                  | **                 | NS                 | **                  | **                  | NS                   |
| 0                       | 27.47 <sup>ab</sup> | 55.10 <sup>a</sup> | 58.27 <sup>a</sup> | 60.53 <sup>c</sup>  | 100.03 <sup>a</sup> | 101.93 <sup>a</sup>  |
| 7                       | 27.60 <sup>ab</sup> | 52.53 <sup>a</sup> | 56.67 <sup>a</sup> | 62.13 <sup>bc</sup> | 100.63 <sup>a</sup> | 101.85 <sup>ab</sup> |
| 14                      | 24.67 <sup>bc</sup> | 57.73 <sup>a</sup> | 58.80 <sup>a</sup> | 64.93 <sup>ab</sup> | 100.65 <sup>a</sup> | 101.75 <sup>ab</sup> |
| 21                      | 29.07 <sup>a</sup>  | 36.93 <sup>b</sup> | 56.73 <sup>a</sup> | 65.60 <sup>ab</sup> | 100.05 <sup>a</sup> | 101.68 <sup>ab</sup> |
| 28                      | 22.47 <sup>c</sup>  | 36.20 <sup>b</sup> | 56.80 <sup>a</sup> | 68.00 <sup>a</sup>  | 99.13 <sup>b</sup>  | 101.63 <sup>b</sup>  |
| 35                      | 27.33 <sup>ab</sup> | 41.67 <sup>b</sup> | 59.40 <sup>a</sup> | 66.60 <sup>a</sup>  | 100.22 <sup>a</sup> | 101.66 <sup>ab</sup> |
| Pooled SE for day       | ±1.04               | ±2.05              | $\pm 1.07$         | ±1.19               | ±0.29               | ±0.09                |
| Treatment* Day          | NS                  | NS                 | NS                 | NS                  | *                   | NS                   |

Table 3 Effect of different roughage to concentrate ratio on RR (breaths/min), PR (beats/min), and RT (°F) of Malpura ewes

 $\mu$  indicates the overall mean for the parameter. Means with similar superscripts along the column do not differ significantly (*P*>0.05) from each other. Group I, R to C 50:50. Group II, R to C 60:40. Group III, R to C 70:30

RR respiration rate, PR pulse rate, RT rectal temperature, NS non-significant

\*P<0.05; \*\*P<0.01

The lower feed and water intake observed in this study showed that as the level of concentrate intake decreased, the animal consumed less feed and water. Furthermore, both BW and BCS were significantly lower in R70. Maternal nutritional status is one of the extrinsic factors programming nutrient partitioning and ultimately affecting growth and reproduction in farm animals (Funston et al. 2010; Sejian et al. 2014b). The reduced BW and BCS in R70 could be attributed to the low level of concentrate in the present study. Koyuncu and Canbolat (2009) indicated that restriction of dietary energy

 $\begin{array}{l} \textbf{Table 4} \quad \text{Effect of different} \\ \text{roughage to concentrate ratio on} \\ \text{T}_3 \ (nmol/l), \ \text{T}_4 \ (nmol/l), \ \text{estradiol} \\ (pg/ml), \ \text{progesterone} \ (ng/ml), \\ \text{and GH} \ (mIU/l) \ \text{concentrations of} \\ \text{Malpura ewes} \end{array}$ 

| Attributes              | GH                 | T <sub>3</sub>     | T <sub>4</sub>      | Estradiol           | Progesterone       |
|-------------------------|--------------------|--------------------|---------------------|---------------------|--------------------|
| μ±SE                    | $0.08 {\pm} 0.003$ | 3.52±0.07          | 57.99±3.23          | 14.20±0.99          | 1.74±0.11          |
| Treatment               | **                 | **                 | NS                  | *                   | NS                 |
| R50                     | 0.06 <sup>c</sup>  | 3.05 <sup>b</sup>  | 55.51 <sup>a</sup>  | 17.11 <sup>a</sup>  | 1.69 <sup>a</sup>  |
| R60                     | 0.09 <sup>b</sup>  | 3.27 <sup>b</sup>  | 53.59 <sup>a</sup>  | 14.98 <sup>ab</sup> | 1.54 <sup>a</sup>  |
| R70                     | 0.10 <sup>a</sup>  | 4.23 <sup>a</sup>  | 64.86 <sup>a</sup>  | 10.52 <sup>b</sup>  | 1.98 <sup>a</sup>  |
| Pooled SE for treatment | ±0.01              | ±.13               | ±5.60               | $\pm 1.71$          | ±.18               |
| Days                    | **                 | *                  | **                  | **                  | **                 |
| 0                       | 0.05 <sup>c</sup>  | 3.05 <sup>b</sup>  | 65.20 <sup>ab</sup> | 10.30 <sup>b</sup>  | 1.14 <sup>bc</sup> |
| 7                       | 0.05 <sup>c</sup>  | 3.38 <sup>ab</sup> | 25.93 <sup>c</sup>  | 23.76 <sup>a</sup>  | 4.51 <sup>a</sup>  |
| 14                      | 0.08 <sup>b</sup>  | 3.85 <sup>b</sup>  | 82.83 <sup>a</sup>  | 20.09 <sup>a</sup>  | 0.86 <sup>c</sup>  |
| 21                      | $0.10^{\rm a}$     | 3.39 <sup>ab</sup> | 70.10 <sup>ab</sup> | 8.32 <sup>b</sup>   | 1.77 <sup>ь</sup>  |
| 28                      | 0.11 <sup>a</sup>  | 3.78 <sup>b</sup>  | 50.56 <sup>b</sup>  | 10.97 <sup>b</sup>  | 1.74 <sup>b</sup>  |
| 35                      | 0.11 <sup>a</sup>  | 3.64 <sup>b</sup>  | 53.30 <sup>b</sup>  | 11.79 <sup>b</sup>  | 0.39 <sup>c</sup>  |
| Pooled SE for day       | ±0.007             | ±0.18              | ±7.91               | ±2.42               | ±0.26              |
| Treatment *Day          | NS                 | NS                 | NS                  | NS                  | NS                 |
|                         |                    |                    |                     |                     |                    |

 $\mu$  indicates the overall mean for the parameter. Means with similar superscripts along the column do not differ significantly (*P*>0.05) from each other. Group I, R to C 50:50. Group II, R to C 60:40. Group III, R to C 70:30

 $T_3$  tri-iodo-thyronine,  $T_4$  thyroxin, *GH* growth hormone, *NS* non-significant \**P*<0.05; \*\**P*<0.01

| Table 5 | Effect of different roughage to concentrate ratio on hemoglobin, PCV, plasma glucose, total cholesterol, total protein, and albumin of Malpura |
|---------|--|
| ewes    |  |

| Attributes              | Hb (g/dl)           | PCV (%)             | Glucose (mg/dl)     | Total cholesterol (mg/dl) | Total protein (g/dl) | Albumin (g/dl)    |
|-------------------------|---------------------|---------------------|---------------------|---------------------------|----------------------|-------------------|
| μ±SE                    | 11.17±0.14          | 37.55±0.54          | 57.08±0.62          | 70.23±1.93                | 11.48±.27            | 26.02±21.19       |
| Treatment               | **                  | NS                  | *                   | NS                        | NS                   | NS                |
| R50                     | 10.50 <sup>b</sup>  | 37.43 <sup>ab</sup> | 56.35 <sup>ab</sup> | 73.13 <sup>a</sup>        | 10.88 <sup>a</sup>   | 4.78 <sup>a</sup> |
| R60                     | 10.63 <sup>b</sup>  | 36.08 <sup>b</sup>  | 55.63 <sup>b</sup>  | 68.15 <sup>a</sup>        | 12.10 <sup>a</sup>   | 5.02 <sup>a</sup> |
| R70                     | 12.37 <sup>a</sup>  | 39.15 <sup>a</sup>  | 59.27 <sup>a</sup>  | 69.41 <sup>a</sup>        | 11.47 <sup>a</sup>   | 4.67 <sup>a</sup> |
| Pooled SE for treatment | ±0.25               | ±0.94               | $\pm 1.07$          | ±3.35                     | ±.48                 | ±0.19             |
| Days                    | **                  | NS                  | **                  | **                        | **                   | *                 |
| 0                       | 10.38 <sup>b</sup>  | 37.75 <sup>a</sup>  | 54.85 <sup>b</sup>  | 68.22 <sup>c</sup>        | 13.08 <sup>a</sup>   | 4.99 <sup>a</sup> |
| 7                       | 11.28 <sup>ab</sup> | 36.35 <sup>a</sup>  | 59.58 <sup>a</sup>  | 64.65 <sup>c</sup>        | 12.65 <sup>a</sup>   | 4.28 <sup>a</sup> |
| 14                      | 11.46 <sup>a</sup>  | 37.27 <sup>a</sup>  | 55.33 <sup>b</sup>  | 54.69 <sup>c</sup>        | 14.45 <sup>a</sup>   | 5.00 <sup>a</sup> |
| 21                      | 10.78 <sup>ab</sup> | 37.61 <sup>a</sup>  | 51.51 <sup>b</sup>  | 55.92 <sup>c</sup>        | 9.36 <sup>b</sup>    | 4.45 <sup>a</sup> |
| 28                      | 11.75 <sup>a</sup>  | 36.45 <sup>a</sup>  | 60.32 <sup>a</sup>  | 82.18 <sup>b</sup>        | 9.74 <sup>b</sup>    | 4.73 <sup>a</sup> |
| 35                      | 11.35 <sup>ab</sup> | 39.89 <sup>a</sup>  | 60.92 <sup>a</sup>  | 95.74 <sup>a</sup>        | 9.61 <sup>b</sup>    | 5.50 <sup>a</sup> |
| Pooled SE for day       | ±0.35               | ±1.33               | ±1.52               | ±4.73                     | ±.67                 | ±51.89            |
| Treatment* day          | NS                  | NS                  | *                   | *                         | NS                   | NS                |

 $\mu$  indicates the overall mean for the parameter; means with similar superscripts along the column do not differ significantly (*P*>0.05) from each other. Group I, R to C 50:50. Group II, R to C 60:40. Group III, R to C 70:30

Hb Hemoglobin, PCV Packed cell volume, NS non-significant

\*P<0.05; \*\*P<0.01

intake pre-mating resulted in less BW gain in sheep. However, BW did not differ significantly between R60 and R70. Hence, the additional 10 % concentrate supplementation in R60 as compared to R70 did not benefit the animals in terms of BW gain. This shows that the minimum level of concentrate supplementation could prove sufficient and cost-effective in semi-arid environment. The reduced BCS in R70 could be due to insufficient fat storage as a result of low level of concentrates in their diets.

Among the physiological responses, RR showed significant changes among different groups than PR and RT. The reason for this could be that homoeothermic animals initially reacted to different levels of feeding by enhancing the thermoregulation mechanism, such as RR, to avoid an increase in RT. These findings are in agreement with previous studies (Maurya et al. 2003; Sejian et al. 2010). The increase in

 Table 6
 Effect of different roughage to concentrate ratio on reproductive performance in Malpura ewes

| Parameters     | Group I                     | Group II          | Group III          |
|----------------|-----------------------------|-------------------|--------------------|
| Estrous (%)    | 98.5                        | 100               | 100                |
| Duration (hrs) | $25.9{\pm}3.2^{\mathrm{a}}$ | $24.9{\pm}4.8^a$  | $27.6 \pm 3.8^{b}$ |
| Length (days)  | 15.5±0.3                    | $15.8 {\pm} 0.40$ | 16.0±0.35          |

Values are averages of two estrus cycles. Values bearing different superscripts within a row significantly differ at P < 0.05 ambient temperature in the morning and towards the afternoon may explain the reason for rise in the physiological responses in the afternoon when compared to values recorded in the morning (Maurya et al. 2004).

The highest level of GH was recorded in R70. Nutrition is one of the environmental cues that affect the somatic growth of ruminants and one of the prime regulators of the levels of GH in livestock (Lee et al. 2005; Sejian et al. 2014b). The underlying mechanisms for this increase could be due to a marked reduction in GH receptors, or a decrease in the binding of GH to its receptor, or post-receptor phenomena (Pulina et al. 2012). Additionally, it has been reported that changes in the secretion of GH due to long-term alterations in nutritional status result from alterations in somatostatin and growth hormone-releasing hormone (GHRH) at the level of the hypothalamus (Henry et al. 2001). The thyroid hormone levels were highest in animals fed 70 % roughage and 30 % concentrate. Appropriate thyroid gland function and activity of T<sub>3</sub> and T<sub>4</sub> are also considered crucial to sustain the productive performance in domestic animals (Todini 2007). Furthermore, energy balance also can play a major role in affecting the plasma level of thyroid hormones in small ruminants (Kong et al. 2004), thus signifying the importance of optimum nutrition for maintaining appropriate thyroid hormone levels in sheep (Sejian et al. 2014b). The highest concentrations of thyroid hormones in R70 suggest that the 70 % roughage and 30 %

concentrate could be the optimum level of nutrition for sheep in semi-arid environment.

The lowest concentration of plasma estradiol was recorded in R70. Such decreased concentration of estrogen may result from diminished ovarian follicular development caused by suppressed peripheral concentration of gonadotrophins (Dwyer 2003). It is generally accepted that nutrition modulates reproductive endocrine function in many species including sheep (Chadio et al. 2007). In our study, the highest plasma progesterone concentration was recorded in R70, and this could be attributed to the fact that low concentrate consumption in this group might have altered the metabolic clearance of progesterone (Aboelmaaty et al. 2008). Similar results, with increase in serum concentrations of progesterone, were also noted in response to low energy fed in sheep (Rabiee et al. 2001). Forcada and Albecia (2006) also demonstrated that the difference in the rate of clearance, rather than differences in secretion levels, is responsible for this apparent inverse relationship between nutrition and peripheral progesterone concentrations in ewes.

Nutrition status of the animal reflects the level of blood cells and blood composition (Sejian et al. 2014b). R70 had significantly a higher level of Hb as compared to R50 and R60. This effect of nutrition on blood Hb may be achieved through a direct effect on erythropoiesis or via hormone action (Umesiobi et al. 2005). The higher level of Hb in R70 demonstrates that sheep fed 70 % roughage and 30 % concentrate could be ideal in maintaining normal blood constituents under semi-arid tropical environment. Furthermore, significantly higher levels of plasma glucose in R70 also support the above statement. However, different nutritional treatment did not affect total cholesterol and total protein levels. This shows that additional concentrate supplementation in R50 and R60 did not bring any beneficial effect in terms of increasing the crucial blood constituents in these animals.

Downing et al. (1995) reported that ovarian responses are influenced by availability of nutrients. However, the different proportion of concentrate and roughage did not influence the estrus% and estrus cycle length. This shows that Malpura ewes were able to adapt to different level of nutrition without affecting the estrus incidences. Sejian et al. (2011) reported a similar result in ewes capable of better adaptation to different levels of nutrition in terms of exhibiting estrus response in the same breed. Further, the estrus duration was significantly higher in R70 as compared to R50 and R60. This shows that sheep fed 70 % roughage and 30 % concentrate is more beneficial in prolonging the estrus duration as the additional concentrate supplementation in R50 and R60 did not improve the reproductive efficiency of Malpura ewes.

# Conclusion

The results from the experiment showed that the feed composition consisting of 70 % roughage and 30 % concentrate is ideal for sheep reared under a semi-arid tropical environment. This is evident from the higher thyroid hormone, Hb, and glucose concentration in R70 as compared to R50 and R60. Further, the BW did not differ significantly between R60 and R70 indicating that the additional 10 % concentrate supplementation in R60 did not benefit the animals in terms of BW gain. Hence, it can be concluded that compared with 50:50 and 60:40, providing 70 % roughage and 30 % concentrate could be the ideal ration composition in semi-arid tropical environment. This ration composition (70:30) will ensure optimal production and reproduction outcomes from these flocks which will in turn ensure economically viable return for the farmers.

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Conflict of interest We declare that we have no conflict of interest.

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