Traits of Sheep and Effects of Protein Supplements on Semen Profile in Indigenous Sheep of Bangladesh

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ABSTRAK

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Penelitian ini dilakukan di distrik Chittagong di Bangladesh dengan kuesioner yang dirancang dengan baik dan terstruktur untuk mengetahui informasi awal tentang domba asli dan efek suplemen protein pada kesuburan. Tiga macam ransum yang mengandung kadar iso-kalori tetapi berbeda dipasok ke tiga kelompok domba yang berbeda di tiga lokasi. Sifat-sifat morfometrik domba seperti panjang rambut, panjang telinga, panjang ekor, panjang badan dan sifat kuantitatif, berat badan di lokasi 3 lebih tinggi daripada dua lokasi lainnya. Panjang rambut jantan (1,91 ± 0,01 cm) lebih besar dari betina sedangkan panjang tubuh rata-rata, panjang ekor dan berat badan betina lebih tinggi daripada jantan. Semua nilai korelasi positif, dimana nilai tertinggi diamati di antara berat badan, panjang tubuh dan tinggi pada pundak (r = 0,73) dan nilai terendah diamati diantara ketebalan dada dan panjang telinga (r = 0,25). Mempertimbangkan persentase sifat-sifat kualitatif dari warna bulu polos, warna kulit tidak berpigmen, warna bulu coklat dan telinga semi-terjumbai ditemukan maksimum daripada yang lain dan nilainya masing-masing 54,21%, 69,16%, 45,79%, 45,79%, 57,01%. Volume semen, jumlah sperma, persentase sperma normal dan viable lebih tinggi dalam pengobatan 2 daripada dua kelompok lainnya. Penelitian ini menyimpulkan bahwa ada pengaruh suplementasi protein pada kinerja reproduksi terutama profil semen dalam ramand studi ini akan menciptakan cakrawala baru produksi domba di Bangladesh.

Kata Kunci: Domba, Sifat, Suplemen protein, Kualitas semen

ABSTRACT

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The study was carried out at Chittagong district of Bangladesh with a predesigned well-structured questionnaire to know the baseline information of indigenous sheep and effects of protein supplementations on fertility. Three iso-caloric but different graded levels of protein containing rations were supplied to the three different groups of sheep in three locations. The morphometric traits of sheep such as hair length, ear length, tail length, body length and quantitative trait, body weight in the location 3were higher than the other two locations. Hair length of male (1.91±0.01cm) was longer than female whereas the average body length, tail length and body weight of females were higher than the males. All the correlation values was positive, where the highest value was observed among the body weight, body length and withers height (r=0.73) and the lowest value was observed in between chest girth and ear length (r=0.25). Considering the qualitative traits percentage of plain coat color, non-pigmented skin color, brown coat color and semi-pendulous ear found maximum than others and the values were 54.21%, 69.16%, 45.79%, 57.01%, respectively. The semen volume, sperm counts, percentages of normal and viable sperm were higher in treatment 2 than the other two groups. The present study concluded that there is an influence of protein supplementation on reproductive performance especially semen profile in ram and this outcome will create a new horizon of sheep production in Bangladesh.

Key Words: **S**heep, Traits, Protein supplements, Semen quality

INTRODUCTION

In Bangladesh, the sheep are mainly non-descriptive type with their average body weight (15 to 25 kg) and probably originated from south-eastern to sub-tropical regions, having adaptive capacity to hot and humid climate. The common characteristics of indigenous sheep of this country are: grey coat with black or white patches and the face, ear and feet are mostly light black with coarse wool. Sheep are generally sparsely distributed throughout the country and most commonly found in the Rajshahi, Dinajpur, Bogra, Rangpur,

Tangail districts and in the delta regions of Noakhali areas (Bindon 1984) managed under traditional production system. Male fertility is the capability to produce good quality semen and the ability to cause pregnancy in a fertile female which can be measured by the serving capacity test. Semen is the combined secretion of male reproductive glands, which plays a major role in determining the fertility and reproductive efficiency. Rekik et al. (2007) reported that protein has a positive effect on reproductive parameters in case of ram. In ruminants low planes of nutrition during the pre-pubertal period delay testicular growth and the onset of puberty by inhibiting the development of a mature reproductive endocrine system (Brown 1994). The previous study (Zohara et al. 2014) has reported the importance of protein supplementation on growth and productivity of small ruminants. On the other hand, it has long been established that rams reared on higher levels of feeding grow faster and attain puberty at younger ages than rams on lower planes of nutrition (Fernández et al. 2005). Spermatogenesis and production of total number of spermatozoa per ejaculate in rams are responsive to appropriate nutritional management (Fernández et al. 2005). This effect has been related to an increase in testicular size results an increase in the volume of the seminiferous epithelium and in the diameter of seminiferous tubules (Al-kawmani et al. 2014). Undeniable, protein supplements using the concept in sheep ration will help to open a new horizon in the countable improvement of the semen quality and reproductive efficiency. This exploration will be subsidiary for realizing the effects of feed supplements (protein) on semen quality in breeding and selection programme. For this purpose, the study was carried out to evaluate different traits of indigenous sheep of Bangladesh, and to know the effects of protein supplements on semen quantity and quality.

MATERIALS AND METHODS

The study was carried out at the two upazilla (sub-district) in the Chittagong district of Bangladesh during the month of January 2017 to December 2017.

Experimental animal selection, ration formulation and study design

A baseline survey was conducted in the studied area with a predesigned questionnaire. A total of 216 sheep, from four different sub-districts (Sandwip 108, Modonhat 16, Hathazarisadar 72 and Bakolia 16) were surveyed directly. The phenotypic and morphological features of the sheep were recorded. Thirty-six indigenous sheep were selected from three different areas (Modonhat (location 1), Hathazari sadar (location 2) and Bakolia (location 3)) from these sub-district on the basis of their body condition score (BCS), health status and normal clinical conditions. Then the sheep were randomly divided into three treatment groups according to location which was heterogeneous in nature and ages were ranged from one to five years. The three groups were managed under the semi-intensive system, kept in separate pens and fed individually according to the group pattern. They were acclimatized and observed regularly for two weeks with a view to screening haemoparasites and helminthes by the farmers and the researcher. Three iso-caloric rations (12 MJ/kg DM) containing graded level of protein were formulated (11.68% CP for control/To, 12.95% CP for T1 and 13.96% CP for T2) by using of conventional feedstuffs (Table 1) and concentrate mixtures (PRO-PAK) were supplied (0.5 kg/day/sheep) to all the groups of sheep for 9 month (March 2018 to November 2018) . Protein-concentrate was provided in both T1 and T2 groups, but there was no protein concentrate in the formulated ration of the control group. All of the treatment groups' sheep allowed for grazing about 8 to 9 hours per day in the grazing land.

Semen collection and evaluation

At the mid of the trial, all rams under different groups were subjected to a fertility test. Scrotal circumference (SC) was recorded by spermatic cord grasping in cm for find out the correlation with semen volume. The rams were trained for semen collection by artificial vagina (AV) method using receptive restrained ewes. During the trial period, semen collection was performed several times in order to assess semen quality (physical, biochemical and microscopic test). Collected semen samples were evaluated by following the procedure of Zemjanis (1970). The volume of semen was obtained directly from the calibrated tube and recorded. Microscopic examination for wave pattern (gross sperm motility) was determined by placing a drop of raw undiluted semen on a pre-warmed slide and viewed using a field microscope at 40X magnification (Jibril et al. 2011). Sperm concentration was measured by Neubauer-haemocytometer according to the method of Organization (2010). Live and dead ratio of the sperm cells was determined as described by Esteso et al. (2006). A thin smear of the semen sample was made on clean grease free glass slide and stained with eosin-nigrosin stain for enumeration of live dead percentage. Sperm abnormalities were determined by making a thin smear of the semen sample on clean grease free glass slide and fixed with buffered normal saline. In both cases, three hundred thirty-three sperm cells were counted per slide using light microscopy at 40X magnification (Jibril et al. 2011).

Table 1. Ingredients And Chemical Composition Of The Experimental Ration

	Treatment groups		
	Control(T0)	Treatment 1(T1)	Treatment 2(T2)
Ingredients			
Animals number	09	16	11
Maize	59	56.5	54.5
Rice Polish	22	22	22
Wheat bran	10	10	10
Soybean oil	0.75	0.75	0.75
Soybean meal	5	5	5
Protein concentrate	0	2.5	4.5
Dicalcium phosphate	1.15	1.15	1.15
DL-Methionine	0.6	0.6	0.6
Vitamin B premix	1	1	1
Common salt	0.5		0.5
Calculated chemicals composition			
Total amount	100	100	100
Total Crude Protein (%)	11.68	12.94	13.96
Total Crude Fiber (%)	4.44	4.46	4.48
Ether Extract (%)	6.03	6.19	6.32
Total Energy, ME (MJ)	12.00	12.00	12.00

Statistical analysis

Data was recorded for the phenotypic traits, semen motility, and live dead ratio of sperm cell. The collected data were corrected and analyzed using PROC GLM of SAS (SAS 2008) followed by completely randomized design (CRD). The mean differences were compared using the least significant difference (LSD) at 5% level of significance.

RESULTS AND DISCUSION

Quantitative traits of sheep

The average of phenotypic characteristics (quantitative traits) of sheep under the three different locations is shown in Table 2. Among the locations, the averages of hair length, ear length, tail length, body length and body weight were found comparatively higher in location 3 than the other two locations.

Considering sex, significant differences were observed between sexes for all the traits except the hair length. Hair length was longer in male than female whereas the average body length, tail length and body weights were superior in females than the males.

The Pearson's correlations among the phenotypic traits (quantitative traits)

Correlations coefficients (r) of different quantitative traits of the indigenous sheep are shown in Table 3. All the traits are positively correlated with each other ($P < 0.001$). The highest positive correlation was found among the body weight, body length and withers height ($r=0.73$) whereas lowest (positive) correlation was observed in between chest girth and ear length ($r=0.25$) of the studied indigenous sheep population.

Qualitative traits of sheep

The qualitative traits of studied indigenous sheep of Bangladesh are shown in Table 4. Irrespective to age and sex, the number of plain coat color sheep was superior (54.21%) than patchy coat color sheep. In the case of skin color, the numbers of non-pigmented sheep were comparatively higher than the pigmented, which was 69.16%. Among the coat color types, brown coat color was found to be highest (45.79%) than the other coat color individuals. Semi pendulous ear was maximum (57.01%) within three areas among the ear patterns. Most of the sheep were polled type, which was 86.92%. Among the tail type patterns, thin tail type nearly 72.90% which was more than fat and fat rump tail percentage.

Seminal traits

The mean and standard error of mean (SEM) values of different seminal traits are shown in Table 5. The semen volume was statistically significant ($P<0.05$) among the three treatment groups of ram. The semen volume was higher in T2 which was 0.94 ml than the other two groups. In the case of sperm count T2 contains 4.25×10^6sperm, which was higher than the other two groups and SEM value was 0.018 that's consoled those groups were significantly different. Among the three treatment groups, scrotal diameter was found to be larger in T2 than the other two groups which were not significantly different ($P>0.05$) among treatments.

Table 2. Mean ± standard error of quantitative traits of sheep of Bangladesh

Categories		Traits (average)				
		Body weight (kg)	Body length (cm)	Ear length (cm)	Tail length (cm)	Hair length (cm)
Location	Location 1	$16.07^{ab} \pm 0.13$	79.47 ± 0.35	$9.21^{b} \pm 0.05$	$10.77^{b} \pm 0.06$	1.88 ± 0.01
	Location 2	$13.27^{b} \pm 0.10$	77.18 ± 0.27	$12.51^{a} \pm 0.04$	$12.63^{a} \pm 0.04$	1.85 ± 0.28
	Location 3	$19.1^{a} \pm 0.74$	86.99 ± 1.91	$12.92^{a} \pm 0.29$	$12.79^{a} \pm 0.33$	2.03 ± 0.04
	P-Value	0.02	0.24	0.001	0.01	0.49
Sex	Male	$10.42^{b} \pm 0.16$	$72.54^{b} \pm 0.43$	$10.12^{b} \pm 0.06$	$11.07^{b} \pm 0.07$	1.91 ± 0.01
	Female	$17.01^{a} \pm 0.08$	$81.93^{a} \pm 0.10$	$11.7^{a} \pm 0.08$	$12.28^{a} \pm 0.03$	1.86 ± 0.01
	P-Value	0.001	0.003	0.05	0.01	0.43

Means with different superscripts i n the same column differ significantly ($p<0.05$)

Table 3. Correlations among the phenotypic traits (quantitative traits) of indigenous sheep of Bangladesh

	Body weight	Body length	Withers height	Chest girth	Ear length	Tail length
Body weight	1	0.73 <0.001	0.73 <0.001	0.71 <0.001	0.35 <0.001	0.36 <0.001
Body length		1	0.61 <0.001	0.47 <0.001	0.38 <0.001	0.33 <0.001
Withers height			1	0.53 <0.001	0.48 <0.001	0.49 <0.001
Chest girth				1	0.25 <0.001	0.26 <0.001
Ear length					1	0.42 <0.001
Tail length						1

Sperm morphology and viability

Percentages of sperm morphology are shown in Figure 1. Microscopic view of normal and abnormal sperm in Picture 1 whereas live and dead sperm in Picture 2. In Figure 1 percentage of normal sperm was highly assiduous in case of treatment groups (T1, 14.50% sperm was abnormal in nature in case of control, which was higher than the other two groups (Picture 1). Among the three groups, these values have no significantly difference (P>0.05). Sperm viability was not statistically significant among the three groups where T2 contain 87.33% viable sperm, which was higher than the other two groups.

Table 4. Qualitative traits of indigenous sheep of Bangladesh

Traits	Types	Percentage(number)
Coat color	Plain	54.21%(58)
	Patchy	45.79% (49)
Skin color	Non pigmented	69.16% (74)
	Pigmented	30.84% (33)
Presence of horn	Present	13.08% (14)
	Absent	86.92% (93)
Coat color type	White	28.97% (31)
	Black	1.86% (2)
	Fawn	14.95% (16)
	Brown	45.79% (49)
	Others	8.41% (9)
Ear pattern	Erect	27.10% (29)
	Semi pendulous	57.01% (61)
	Pendulous	15.89% (17)
Tail type	Thin	72.90% (78)
	Fat rump	9.35(10)
	Fat	17.76% (19)

Table 5. Various seminal traits of ram

Traits	T0/Control	Treatment 1 (T1)	Treatment 2 (T2)	SEM	P-value
Semen Volume (ml)	0.81^{b}	0.88^{ab}	0.94^{a}	0.013	0.031
Sperm Count (10^9)	4.09^{b}	4.15^{ab}	4.25^{a}	0.018	0.04
pH	7.43^{a}	7.07^{b}	6.95^{b}	0.028	0.002
Scrotal diameter(cm)	13.10	17.86	20.82	2.248	0.225

Means with different superscripts in the same row differ significantly among the treatment groups (P<0.05)

Phenotypic characteristics (Quantitative traits) of Sheep

Phenotypic characteristics of indigenous sheep population showed the presence of clear morphological variations according to production systems diversity persisted in indigenous sheep. A similar finding was reported by Yakubu et al. (2011) with the current study.

The average body weight of sheep under this investigation was found similar to the results of Nsoso

et al. (2004) and Pervage et al. (2012). The female sheep attained more live weight which is also matched with Nsoso et al. (2004) who reported the body weight of indigenous sheep was higher in the female than the male. Differences of body length were observed between male and female sheep which was higher than the findings of Hassan & Talukder (2012) and conform to the study of Pervage et al. (2012) in the case of indigenous sheep.

The body length of indigenous sheep was varied, and this variation in body length occurs due to the differences in genetic make-up, feeding management, housing management, health and other management factors and same factors was described by Hassan & Talukder (2012) for the body length variation of sheep.Ear length of indigenous sheep firmly coincided with the findings of Nsoso et al. (2004) and larger than the findings of Pervage et al. (2012) and Hassan & Talukder (2012). This variation in the measurements might be due to the variation in different managerial system and ages of sheep. Average tail length of sheep was similar to the measurement of Hassan & Talukder (2012) and Nsoso et al. (2004),they found that the tail length was 12 cm in indigenous sheep. Tail length could be varied due to differences in production systems among the studied indigenous sheep.

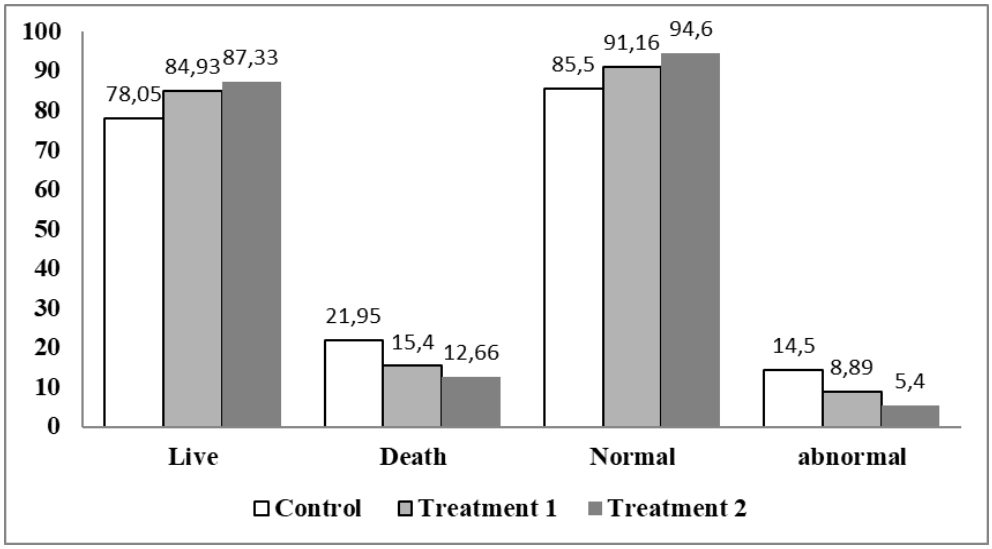
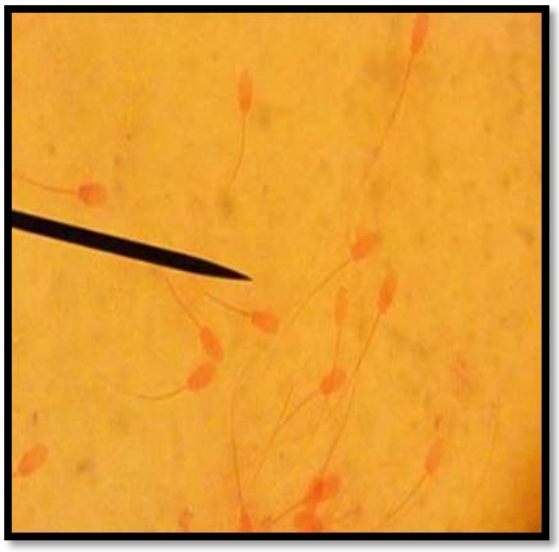


Figure 1. Percentage of sperm morphology



Picture 2 : Normal and abnormal sperm

Picture 3: Live and dead sperm

Seminal traits and sperm morphology

Reproductive performance of livestock is determined by various factors those are genetic merit, physical environment, nutrition and management. Nutritional factors are perhaps the most crucial in terms of their direct effects on the reproductive phenomenon and the potential to moderate the effect of other factors. There was an interrelationship between energy intake and reproductive performance in adult rams (Kheradmand et al. 2006; Jibril et al. 2011).

Semen volume is one of the important factors in semen evaluation and reproduction performance in the males (Ax et al. 2016). The semen volume was0.94 ml in Treatment 2 group (protein-rich feed) which was harmonious with the findings of Fernández et al. (2005), Abera et al. (2014) and Asefa et al. (2017)where they denoted that control group generates less volume semen than the treatment groups. On the other hand, there was in inconsistent with the findings of Jibril et al. (2011) who said that semen volume was not influenced by the level of protein. This variation may occur due to changes in protein percentages in ration and farm management system. Nutrition has effect for the growth and maturation of sertoli cells in newborn lambs, which are strong candidates for future performance because the number of sertoli cells was highly correlated with the maximum rate of sperm production (Bielli et al. 1999) and sperm concentration. In the present experiment sperm count in T2 was higher than the other two groups, strongly coincided with the research outcome of (Jibril et al. 2011) where they said that increase crude protein intake above the minimum requirements resulted in improved sperm concentration and favors for spermatogenesis.

Scrotal diameter, testicular sizes were affected by nutrition, where testicular growth can be affected when animals are fed above their maintenance requirement. The findings of present studies of scrotal diameter were more sizably voluminous in the treatment group 2 than the other two groups which were agreed with the results obtained by Fernández et al. (2005) where they noticed that the improve protein-rich diet was helpful for larger growth of scrotal circumference than the control group, although inconsistent with those obtained by Bielli et al. (1999) who found no significant effect from high dietary protein on testicular dimensions. Percentage of normal sperm was highly diligent in case of treatment groups than the control group. Here percentage of normal sperm are more in the case of protein enriched Treatment 2 group which was consistent with the findings of Kheradmand et al.(2006) and Jibril et al. (2011) and did not matched with the findings of Barth et al. (2008) who found that the medium or high level of nutrition does not have influenced on overall percentage of morphologically normal spermatozoa. Present studies indicated that the sperm viability was not statistically significant among the three groups where Treatment group 2 contains higher viable sperm than the other two groups and strongly coincide with the findings of Jibril et al. (2011).

CONCLUSION

Indigenous sheep from three different areas were varied phenotypically. All the phenotypic traits were positively correlated with each other. The percentage of plain coat color, non-pigmented skin color, brown coat color and semi-pendulous ear were significantly higher than other qualitative traits. The seminal traits were better in treatment 2 (13.96% CP) than the other two groups that suggest that improved dietary intake above maintenance requirements had positive effects on rams and reproductive performances that presage to rear sheep with on supplements may improve the sheep production in Bangladesh.

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