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Spermiogram Characteristics in Epididymal Washings of Bucks During Winter and Summer Season

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ABSTRACT

The present investigation was designed to study certain morphological characteristics of epididymal washings/plasma during winter and summer season in the bucks. The study was undertaken with the view to find out the effect of different season on the epididymal physiology of bucks. The epididymis was collected from sixteen apparently healthy bucks, immediately after their slaughter. In the laboratory, separation and washings of caput, corpus and cauda epididymis were carried out separately. The spermatozoa of the cauda epididymis showed higher ($P < 0.05$) mass motility as compared to corpus epididymis in winter as compared to summer season and caput epididymis in which they were non-motile in both the seasons. The result of present study showed high ($P < 0.01$) concentration of spermatozoa in different part of epididymis during winter as compared to summer season. During winter season, spermatozoa concentration in the cauda was significantly higher ($P < 0.01$) as compared to corpus and caput. During summer, live per cent values of spermatozoa were significantly lower ($P < 0.01$) in caput, corpus and cauda epididymis as compared to winter value of caput, corpus and cauda epididymis. Most of the spermatozoa from the caput epididymis showed the higher cytoplasmic droplet at their neck portion. However, the spermatozoa of the corpus and cauda epididymis revealed a non-significant reduction in the cytoplasmic droplet at the neck portion. All the above parameters indicate that summer stress severely affect epididymal physiology and semen quality of buck.

Keywords: Buck, spermiogram, summer, winter

Goat is considered as the great future in changing livestock scenario. The poor man's cow- goat has tremendous potential to be projected as the future animal for rural areas under the changing agro-climatic conditions and lack of forages (Jindal *et al*., 2011). Sperm cells have capacity of fertilization linked, not only to physical and morphological aspects but also to biochemical aspect of the semen. The seminal plasma is a complex mixture from epididymal and accessory glands fluid (Muino-Blanco *et al.*, 2008). Oyeyemi and Ubiogoro (2005) observed progressive motile spermatozoa first in the testes (6 – 10%) in the caput (20 – 30%), corpus (40 – 50%) and highest at the caudal epididymis (85%) in large white boar. This motility increases from caput to caudal epididymis. Good motility is an important factor associated with sperm quality, a property often regarded as being of the utmost importance for fertility. It is perhaps even the most important parameter for assessing fertilization *in vivo* and *in vitro*, although the necessary technology for assessing the motility characteristic in not yet standardized (Feichtinger, 1989). The motility of the spermatozoa adversely affected during summer season as compared to winter season. Therefore the present study was undertaken to evaluate the extent of changes in the epididymal physiology certain morphological and biochemical characteristics of epididymal washings/plasma/ spermiogram of the buck spermatozoa during winter and summer season. The research investigation was conducted in during winter (November to January 2014) and summer season (April to June 2014).

MATERIALS AND METHODS

The epididymis was collected from sixteen apparently

healthy bucks, immediately after their slaughter. In the laboratory, separation and washing of caput, corpus and cauda epididymis were carried out separately. The following semen parameters were studied in the washings. Motility of the spermatozoa was evaluated immediately after collection of epididymal fluid, on a clean and dry glass slide. Motility of the spermatozoa (Zemjanis, 1970), concentration of the spermatozoa (Salisbury and Van Demark, 1978) and live/dead spermatozoa count (Hancock, 1952) were recorded. The data was statistically analyzed by t-test. Mean and standard errors were calculated as per procedure by Steel *et al.* (1980).

RESULTS AND DISCUSSION

Concentration of the spermatozoa was worked out and subsequently the epididymal content and spermatozoa concentration per gram of epididymal weight was calculated. The result obtained showed a highly significant ($P < 0.01$) difference in spermatozoa concentration between caput corpus and cauda epididymis, out of which cauda having maximum concentration (1097.12 ± 81.25 millions/ml), and least in corpus (140.37 ± 7.41 millions/ml) though similar, but low concentration of the spermatozoa was recorded during summer season (Table 01).

The eosin-nigrosin stained spermatozoa were studied for their live percentage and presence of cytoplasmic droplets (per cent). The live per cent values of the spermatozoa were significantly ($P < 0.05$) lower in corpus (82.13 ± 1.99 %) epididymis as compared to the caput (86.04 ± 2.20 %) and cauda (86.72 ± 1.62 %) epididymis (Table 1). In summer season, there was low per cent of live spermatozoa found out in the epididymis.

Most of the spermatozoa from the caput epididymis showed the cytoplasmic droplet at their neck (43.53 ± 17.40 per cent) portion (Table 2). However, the spermatozoa of the corpus and cauda epididymis revealed a significant ($P < 0.05$) reduction in the cytoplasmic droplet in the neck portion. In summer season midpiece was having highest concentration of cytoplasmic droplet.

The statistical analysis revealed no significant effect of summer season on either portions of epididymis. The findings in the present investigation are very much in agreement with that of Chandrapal and Bharadwaj (1988), who reported considerably low weight of corpus epididymis than those of the other portions of epididymis. Similar observations were also recorded by Ansari *et al.* (1972) and Hafez (1993).

The statistical analysis revealed no significant effect of winter and summer season on either portion of epididymis. The findings in the present investigation are very much in agreement with that of Oyeyemi and Ubiogoro (2005), who reported that progressive motile spermatozoa was the highest in the caudal epididymis in large white boar. This motility increased from caput to caudal epididymis. Souza *et al.* (2010) also observed that mass motility and vigour did not vary between seasons, throughout the year in tropical environment. Datta *et al.* (1990) revealed that the percentage of motile spermatozoa was found to be maximum in the cauda, followed by corpus and in caput the motility was absent.

Table 1: Spermiogram of the epididymis of bucks

Parameter	Season	Caput	Corpus	Cauda
Weight (g)	Winter	5.39±0.33	1.48±0.08	3.94±0.29
	Summer	5.11±0.18	1.39±0.06	3.15±0.22
t-value		0.7304	0.8721	2.174
Mass motility (%)	Winter	Non-motile	19.38±2.90	78.13±3.27
	Summer	Non-motile	15.00±3.78	64.37±5.78
t-value		0	0.92	2.07
Concentration (10^6/ml)	Winter	142.75±8.1	140.37±7.41	1097.12±81.25
	Summer	47.62±7.96	43.87±6.25	228.87±21.53
t-value		8.38**	9.95**	10.33**
Live spermatozoa (%)	Winter	86.04±2.20	82.13±1.99	86.72±1.62
	Summer	65.35±4.76	67.69±2.53	73.98±2.74
t-value		3.94**	5.17**	4.00**
Dead spermatozoa (%)	Winter	13.96±2.2	17.87±1.19	13.25±1.62
	Summer	34.65±4.74	32.31±2.53	26.01±2.74
t-value		3.93**	5.18**	4.01**

*Significant (P<0.05), ** Significant (P<0.01)

In the present study our findings differ considerably to values reported by Jindal and Panda (1980) and Bhatt and Chauhan (1982). Distribution of the spermatozoa between caput, corpus and cauda epididymis in the present study are very much in correspondence with those reported for Black Bengal buck by Datta *et al.* (1990). These

Table 2: Cytoplasmic droplet count in epididymis of buck

Particulars	Season	Caput	Corpus	Cauda
Cytoplasmic droplet in neck of sperm	Winter	21.19±8.95	0.90±.53	0.24±0.24
	Summer	7.92±4.25	1.57±0.78	1.39±0.83
t-value		0.72	0.72	1.34
Cytoplasmic droplet in mid-piece of spermatozoa	Winter	20.76±7.67	12.19±3.22	6.85±1.23
	Summer	8.97±2.45	20.76±7.67	10.81±4.05
t-value		1.03	1.03	0.94
Cytoplasmic droplet in tail of spermatozoa	Winter	1.57±0.78	1.91±0.95	1.30±0.85
	Summer	0.44±0.29	1.986±1.06	5.39±2.36
t-value		0.05	0.05	1.63
Total	Winter	43.53±17.40	15.00±4.7	8.39±2.32
	Summer	17.33±6.99	24.32±9.51	17.61±7.24

differences in values appear to be due to the difference in age, breed, species, variable environmental conditions, managemental practices and spermatozoa counting methodology. Abnormal climatic conditions are, thus, likely to affect adversely the spermatozoa function and male fertility, though still not a tenable fact.

The findings of the present investigation are in confirmation with Datta *et al.* (1990) in Black Bengal buck and El-Darawany (1999) in rams who reported that dead spermatozoa are selectively removed during their transit from the caput to cauda epididymis being the store house of spermatozoa and provides conducive environment for their survival which might be responsible for the presence of greater number of live spermatozoa in the cauda epididymis (Hafez, 1993). Similar finding were also observed in the present research investigation the lower values of live spermatozoa in the corpus epididymis may be possibly due to it is elongated, tubular, reduced luminal structure acting as a conduit. Partly, it may be due to continuous transfer of spermatozoa from the corpus to cauda epididymis. Similar trends for the live spermatozoa content of three regions of the epididymis were also reported by Oyeyemi and Ubiogoro (2005) in large white boar.

The possible physiological significance of cytoplsmic droplet, the un-sequestered cytoplasm, in relation to spermatozoa metabolism and maturation are still to be investigated. However, the movement, alterations in the fine structure and position of the cytoplasmic droplet during spermatozoan passage through the epididymis plays a key role for maturation of the male gametes. It is assumed that the droplets have a nutritive role in the economy of the spermatozoan and may be the source of metabolizable endogenous substrate. The lysosomal enzymes of the droplets perhaps prepare the spermatozoon for the final stage of its maturation (Cunningham and Hafez, 1980).

Presence of cytoplasmic droplet, on the spermatozoa observed in indigenous breed, their migration from proximal region to distal region of the spermatozoa, moreover, decrease in their number and/or absence during passage through the male tracts as a sequence of spermatozoa maturation corroborated with the findings obtained from different mammalian species by Jindal and Panda (1980) in goat, Rao *et al.* (1980) in cattle, Olugbeng and Babalola (2006) in bulls and Datta *et al.* (2014) in Black Bengal buck.

CONCLUSION

Most reliable parameter of semen evaluation i.e., motility of the spermatozoa was adversely affected during summer season in bucks. The concentration of spermatozoa, live percentage and percentage of cytoplasmic droplet were highest during winter season as compared to summer season. All the above parameters indicate that summer stress severely affect epididymal physiology and semen quality of buck.

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