# EFFECT OF SEASON ON SPERM MORPHOMETRY AND DEAD SPERM PERCENTAGE IN DIFFERENT AGE GROUPS OF *BUBALUS BUBALIS*

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## ABSTRACT

The morphometrical values of buffalo bull spermatozoa studied for one year revealed 7.91 $\pm$0.86 $\mu$ head length, 4.84 $\pm$0.57 $\mu$ maximum head width, 12.35 $\pm$ 1.60 $\mu$ mid piece length and 54.40 $\pm$ 6.38 $\mu$ total tail length. The values for these components were significantly ($P<0.05$) higher in summer season whereas among age groups no difference was observed. The dead sperm percentage was 16.25 $\pm$ 13.28 and it was also significantly ($P<0.05$) higher in summer season, and in group 3 (bulls of 8-9 years of age).

## INTRODUCTION

The variation in morphology of spermatozoa has been reported by many workers in different species. The difference in shape of spermatozoa in cow and buffalo has also been reported (Malik *et al.*, 1974). Similarly, the size of various components of spermatozoa varied among cow and buffalo bulls (Malik *et al.*, 1974). Yet in literature, there seems to be sparse information on these components including sperm head length, maximum head width, mid piece length and total tail length in *Bubalus bubalis* in relation to age and season. The present investigations were thus carried out to see the effect of age and season on the sperm morphometeric values in buffalo bulls.

## MATERIALS AND METHODS

This study was carried out for one year on 16 buffalo bulls (*Bubalus bubalis*) being used for semen collection at Semen Production Unit, Qadirabad, Punjab. These bulls were randomly selected and divided into four age groups, each group having four bulls as under:

Group A : 3-4 years of age
Group B : 6-7 years of age
Group C : 8-9 years of age
Group D : 12-15 years of age

The study period was divided into following seasons:

Hot Dry Summer: May 1 to June 30
Hot Humid Summer: July 1 to September 15
Autumn: September 16 to November 14
Winter: November 15 to February 15
Spring: February 16 to April 30

Semen from all the bulls was collected at fortnightly interval with artificial vagina. The smears on glass slides from freshly collected semen were prepared taking simultaneously one drop of Eosin-Nigrosin stain (modified method of Bloom and Anderson, 1965) and a relatively small drop of semen. The prepared smears after drying were seen under the light microscope using oil immersion at 1000 X. A total of 200 sperms were counted to determine dead sperm percentage and another 100 sperms to study the sperm morphometry by using ocular micrometer to measure the head length, maximum head width, middle piece length, and total tail length (including mid piece).

The results obtained were compared in different seasons and in different age groups using oneway analysis of variance or DMR for comparing means (Anonymous, 1989).

## RESULTS AND DISCUSSION

The study was conducted on 16 buffalo bulls of different ages for one year. The data on head length (HL), maximum head width (MHW), middle piece length (MPL), and total tail length (TTL) of buffalo bull sperms showed statistically no difference between four age groups compared in any season or at the end of the year. However, HL, MHW, MPL and TTL showed significantly ($P < 0.05$) higher values in summer (hot dry and hot humid) season (Table 1).

The mean head length was 7.91 $\pm$0.86 $\mu$, but it was also measured 9.22 $\pm$0.38 $\mu$ in dry summer, where it

Table 1: Comparison of (means $\mu \pm$ SD) different components of buffalo spermatozoa at 1000X, using oneway analysis of variance.

| Parameters/ Group | Summer | | Autumn | Winter | Spring | Total |
|---|---|---|---|---|---|---|
| | Dry | Humid | | | | |
| **Head Length (HL)** | | | | | | |
| A | 9.45 ± 0.38c | 8.52 ± 0.84b | 7.49 ± 0.25a | 7.33 ± 0.11a | 7.28 ± 0.12a | 8.01 ± 0.95 |
| B | 9.18 ± 0.37b | 8.63 ± 0.88b | 7.44 ± 0.21a | 7.38 ± 0.17a | 7.38 ± 0.47a | 8.00 ± 0.89 |
| C | 9.23 ± 0.41b | 7.71 ± 0.22a | 7.38 ± 0.36a | 7.33 ± 0.21a | 7.49 ± 0.38a | 7.83 ± 0.78 |
| D | 9.01 ± 0.38c | 8.04 ± 0.68b | 7.55 ± 0.27ab | 7.28 ± 0.28ab | 7.11 ± 0.21a | 7.80 ± 0.78 |
| Total | 9.22 ± 0.38c | 8.22 ± 0.74b | 7.46 ± 0.26a | 7.33 ± 0.18a | 7.32 ± 0.31a | 7.91 ± 0.86 |
| **Maximum Head Width (MHW)** | | | | | | |
| A | 5.81 ± 0.21b | 5.48 ± 0.88ab | 4.78 ± 0.36a | 4.40 ± 0.21a | 4.51 ± 0.21a | 4.99 ± 0.69 |
| B | 5.65 ± 0.30b | 5.05 ± 0.62ab | 4.72 ± 0.27a | 4.51 ± 0.11a | 4.40 ± 0.11a | 4.86 ± 0.55 |
| C | 5.59 ± 0.32b | 4.61 ± 0.11a | 4.56 ± 0.47a | 4.45 ± 0.22a | 4.51 ± 0.21a | 4.74 ± 0.51 |
| D | 5.65 ± 0.39b | 4.72 ± 0.27a | 4.72 ± 0.32a | 4.34 ± 0.00a | 4.40 ± 0.11a | 4.77 ± 0.53 |
| Total | 5.67 ± 0.29c | 4.96 ± 0.61b | 4.69 ± 0.34ab | 4.42 ± 0.15a | 4.45 ± 0.16a | 4.84 ± 0.57 |
| **Mid Piece Length (MPL)** | | | | | | |
| A | 14.93 ± 0.84c | 12.71 ± 1.26b | 10.97 ± 0.57a | 11.78 ± 0.60ab | 11.18 ± 0.28ab | 12.31 ± 1.63 |
| B | 14.61 ± 0.44c | 13.47 ± 1.80bc | 11.08 ± 0.64a | 11.78 ± 0.37a | 11.46 ± 0.11a | 12.48 ± 1.58 |
| C | 15.15 ± 0.57b | 11.89 ± 0.62a | 11.29 ± 1.06a | 11.40 ± 0.38a | 11.35 ± 0.21a | 12.22 ± 1.62 |
| D | 14.93 ± 1.30c | 12.92 ± 1.33b | 11.51 ± 0.75ab | 11.51 ± 0.39a | 11.08 ± 0.36a | 12.39 ± 1.67 |
| Total | 14.90 ± 0.79c | 12.75 ± 1.31b | 11.21 ± 0.73a | 11.62 ± 0.43a | 11.26 ± 0.27a | 12.35 ± 1.60 |
| **Total Tail Length (TTL)** | | | | | | |
| A | 65.81 ± 1.40b | 57.12 ± 7.94a | 50.28 ± 0.80a | 50.99 ± 0.92a | 53.10 ± 1.20a | 55.46 ± 6.70 |
| B | 64.62 ± 1.64b | 57.45 ± 7.22ab | 50.50 ± 1.73a | 51.69 ± 1.70a | 51.15 ± 1.49a | 55.08 ± 6.34 |
| C | 59.78 ± 8.96b | 51.20 ± 2.57ab | 48.27 ± 5.24a | 52.61 ± 1.96ab | 50.93 ± 0.91ab | 52.56 ± 5.89 |
| D | 65.10 ± 1.15b | 55.33 ± 8.08ab | 50.72 ± 1.70a | 50.39 ± 1.05a | 50.82 ± 1.11a | 54.47 ± 6.68 |
| Total | 63.82 ± 4.82c | 55.28 ± 6.63b | 49.94 ± 2.79a | 51.42 ± 1.57ab | 51.50 ± 1.44ab | 54.40 ± 6.38 |

Values with different letters in a row are different ($P < 0.05$).

Table 2: Comparison of means (± SD) using DMR of Dead Sperm percentage of different age groups in different seasons of the year

| Age Group | Summer | | Autumn | Winter | Spring | Total |
|---|---|---|---|---|---|---|
| | Dry | Humid | | | | |
| A | 21.24 ± 12.09b | 16.84 ± 12.15ab | 9.94 ± 7.37a | 12.80 ± 9.56a | 13.21 ± 9.46$^{a}$ | 14.79 ± 10.79$^{A}$ |
| B | 14.01 ± 6.60ab | 20.32 ± 11.91b | 8.71 ± 6.30a | 15.51 ± 7.90ab | 16.14 ± 7.37$^{ab}$ | 14.91 ± 8.85$^{A}$ |
| C | 26.18 ± 22.94 | 22.03 ± 16.86 | 19.60 ± 20.50 | 20.87 ± 16.74 | 17.48 ± 12.93 | 21.16 ± 18.07$^{B}$ |
| D | 17.23 ± 9.44 | 14.26 ± 9.52 | 14.27 ± 15.55 | 15.88 ± 19.08 | 10.30 ± 4.25 | 14.44 ± 12.95$^{A}$ |
| Total | 19.58 ± 14.50c | 18.10 ± 12.68bc | 13.00 ± 13.97a | 16.34 ± 14.28abc | 14.28 ± 9.33$^{ab}$ | 16.25 ± 13.28 |

The values in last column with different capital letters and in rows with different small letters are statistically different ($P < 0.05$).

was significantly ($P < 0.05$) higher. Similarly, when single age group in different season was studied the HL showed significantly higher ($P < 0.05$) values in summer (group A and B in dry and humid, and group C and D in dry summer) (Table 1). The head length of 8.3 $\mu$ (8.2-8.5) for buffalo bulls has been reported by Malik *et al.* (1974) and a range of 8-10 $\mu$ has been given for cow bulls (Bretschneider, 1948; Salisbury and Dongen, 1964; Bloom and Anderson, 1965).

The maximum head width (MHW) was 4.84 ± 0.57 $\mu$, but in summer it was 5.67 ± 0.29 $\mu$, where it was also significantly ($P < 0.05$) higher. Previously in *Bos bubalis*, MHW was reported to be 4.25 $\mu$ (4.21-4.32) by Malik *et al.* (1974). The ratio of HL to MHW was 1.63:1 where as in *Bos bubalis* it was 1.95:1 (Malik, *et al.*, 1974).

The mid piece length (MPL) was measured to be 12.35 ± 1.60 $\mu$ (Table 1), which was in agreement to the findings of Malik *et al.* (1974) who reported it in *Bos bubalis*. In Murrah buffalo bulls of India, 12.5 $\mu$ MPL has been reported by Guha *et al.* (1959). The total tail length (TTL) was measured 54.40 ± 6.38 $\mu$ (Table 1). Almost similar TL of 54.8 $\mu$ (54.7-55.0) has also been reported in *Bos bubalis* (Malik *et al.*, 1974) and 54.6 $\mu$ in Murrah bulls (Guha *et al.*, 1959) while 47-54 $\mu$ in cow bulls (Bretschneider, 1949). The TTL in our study included mid piece length.

Much difference does not appear in various components of buffalo bulls in regard to previous reports for *Bos bubalis* and Murrah bulls of India. However, the values observed during present study for some of the components, particularly in summer season, were high, indicating possible effect of high temperature on spermatozoa some where in its formation or storage or both, which needs to be investigated.

The four age groups and season did show significant ($P < 0.05$) difference in dead sperm percentage with higher ($P < 0.05$) values in group C, and in summer season (Table 2). When single age group in different seasons was studied, the dead sperm percentage of group A and B showed significantly ($P < 0.05$) higher values in summer (group 1 in dry summer and group B in humid summer). The overall dead sperm percentage was $16.25 \pm 13.28$ (but it was $19.58 \pm 14.50$ in summer which was again significantly ($P < 0.05$) higher. The significant ($P < 0.05$) difference of dead sperm percentage in different seasons recorded was only in bulls of relatively young age groups (3-7 years). The percentage of live spermatozoa in Egyptian buffalo bulls was $58.4 \pm 2.73$ (Osman and El-Azab, 1974) and $72.89 \pm 0.50$ in Murrah buffalo bulls (Guha *et al.*, 1959). Campbell *et al.* (1960) have reported dead sperm percentage of 21.0 in Hereford bulls. Galloway and Norman (1980) proposed, 50 percent live spermatozoa as limits of semen values consistant with normal reproductive functions. Further studies be focused on ultrastrucure of sperm in relation to age and season in *Bos bubalus*.

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