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# Scrotal Circumference Variation on Semen Characteristics of Artificial Insemination (AI) Bulls

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**Abstract:** The main objective of the present investigation was to evaluate the importance of scrotal circumference variation on semen characteristics of bulls used for Artificial Insemination (AI) programme, in Bangladesh. For this purpose, a total of 3720 semen samples were collected from 71 bulls at three AI centres/stations (CCBSDF, Savar, Dhaka, RDCIF, Rajabarihat, Rajshahi and DAIC, Rajshahi) from 1995 to 2002; inclusive. These bulls were classified into 4 groups according to their scrotal circumference (SC) such as Group-I: <34 cm, Group-2: 34-36 cm, Group-III: 36-38 cm and Group-IV: >38 cm. The semen characteristics of the average values with standard deviation of volume, colour, density, mass activity, sperm concentration, sperm motility, total number of sperm cells/ejaculate, total number motile sperm cells /ejaculate, number of semen doses /ejaculate, post-freezing motility of sperm and % of abnormal spermatozoa were 7.14±2.1ml, 3.23±0.9(scale:1-4), 3.56(scale:1-5), 2.91±0.81(scale:1-5), 1314.5±12.6 million/ml, 63.95±4.61%, 9375.7±3431.5million, 6023±2299million, 312.0±115.3, 55.09% and 16.38±3.2 respectively. Scrotal circumference groups had significant effect (P<0.05) on semen characteristics except on post-freezing motility of sperm. The volume, total sperm cells/ejaculate, total number of motile sperm cells/ejaculate and number of semen doses per collection were gradually increasing with the sizes of SC groups of bulls. The largest volume (8.53±2.07ml) was found in >38 cm with smallest (5.15±1.70ml) in <34 cm of SC groups. The variations of SC on % of total sperm abnormalities were found to be significant. The highest average percentage of total sperm abnormalities was recorded (17.00%) in >38cm followed by <34 cm, 34-36 cm and 36-38 cm of SC groups (16.76%, 16.39% and 15.73%, respectively). Again, Analysis of variance indicated that breeds of bull had significant (P<0.05) effect on scrotal circumference. Significant (P<0.05) highest SC (38.0+0.7 cm) in 100% Friesian and lowest (33.9=1.2 cm) in 100% Local bulls. Result suggested that SC of >38 cm group of bulls were suitable for better performance of semen characteristics as well as 100% Friesian bulls ware sound for top scrotal circumference of AI bulls.

**Key words**: Artificial insemination, Bull, Scrotal circumference, Semen characteristics and Spermatozoa

## Introduction

Determination of scrotal circumference is an essential aspect of breeding soundness examination of bulls because of its significance as indicators of puberty, total sperm production, semen quality and pathological conditions of the testes (Ott, 1991). Testicular size of the sire lines is an important traits in the selection of males practically those used in AI progarmmes (Ying and Johnson, 1996). A relationship between the size of testes and the sperm motility and total spermatozoa per ejaculate of bulls (Neely *et al.*, 1982; Bruner *et al.*, 1995). In many studies testicular measurements have been evaluated and related to some seminal parameters, usually sperm number, sperm concentration and sperm motility (Achnelt *et al.*, 1964; Gipson *et al.*, 1985; Ruttle *et al.*, 1982). Bull with small testicles was shown to have semen with a higher percentage of sperm defects (Achnelt *et al.*, 1964). The average scrotal circumference was larger in the Friesian than Ayrsires but the difference was not significant (32.6 cm Vs 32.2 cm). Testicular dimensions were correlated with semen traits and scrotal circumference and testicular consistency provided a good fertility index (Jakubiec, 1983). Accuracy of scrotal circumference as a predictor of specific pathologic changes within testicular parenchyma was assessed by scoring 121 Hereford and Simmental bulls for scrotal circumference and semen quality and thirty-two centimeters was the minimal acceptable scrotal circumference to ensure both a low prevalence of tubules with irreversible loss of germinal epithelium and acceptable semen quality (Thompson *et al.*, 1992). The success of Artificial Insemination Programme mainly depends on the successful selection of properly fertile bulls. So, a thorough knowledge of scrotal circumference in semen characteristics of bulls used for AI programme in Bangladesh is very important. Information is not available of the semen characteristics as influenced by scrotal circumference of bulls in CCBSDF, Savar, Dhaka, RDCIF, Rajabarihat, Rajshahi and District Artificial Insemination Centre (DAIC), Rajshahi. The present study was undertaken with following objectives: i) To study the effect of scrotal circumference (SC) on semen characteristics of AI bulls and ii) To find out the effect of breed on scrotal circumferences of bulls.

## Materials and Methods

The present study has been dealing with the scrotal circumference variation on semen characteristics of AI bulls. The study was conducted in three Artificial Insemination (AI) centres /stations and at the Department of Genetics and Breeding, Rajshahi University during the period from 1995 to 2002. The AI centres /stations were Central Cattle Breeding Station and Dairy Farm (CCBSDF), Savar, Dhaka, Rajshahi Dairy and Cattle Improvement Farm (RDCIF), Rajabarihat, Rajshahi and District Artificial Insemination Centre (DAIC), Rajshahi. The basic materials for the present study comprised seventy-one (71) bulls belonged to six (6) genetic groups. Out of 71 bulls, 50 bulls from CCBSDF, Savar, Dhaka, 7 from RDCIF, Rajabarihat, Rajshahi and 14 bulls from DAIC, Rajshahi. A total of 3720 ejaculates (626 from 100% Friesian, 620 from 100% Sahiwal, 784 from 75%Fx25%L, 572 from 50%SLx50%F, 769 from 50%Fx50%L and 349 from 100% L) were obtained from 71 AI bulls in three AI centres.

The scrotal circumference of the bulls were classified keeping with in four (4) groups as follows 1st group = < 34 cm, 2nd group = 34 to 36 cm, 3rd group = 36 to 38 cm and 4th group = > 38 cm.

Measurements of scrotal circumference (in centimeters) were made fortnightly at the area of the largest diameter of the scrotum using a cloth tape as described by Ott (1991).

The testes were pulled firmly into the lower part of the scrotum by encircling its base with the hand and pulling down on the testes. The

scrotal tape was formed into a loop and slipped over the scrotum and pulled up snugly around the greatest diameter of the scrotal contents. The thumbs and fingers were located on the side of the scrotum rather than between the testes to prevent separation of the testes and inaccurate measurement.

The experimental AI bulls were divided into 6 (six) groups along with their genetic composition viz; Group-1 = 100% Friesian (100% F), Group-2 = 100% Sahiwal (100% SL), Group-3 = ≥75% Friesian ×≤25 %Local (≥75% F × ≤25 % L), Group-4 = ≥50% Sahiwal × ≤50% Friesian (50% SL × 50% F), Group-5 = 50% Friesian×50% Local (50% F × 50% L) and Group-6= 100% Local (100%L).

All the bulls were kept under identical conditions of management, feeding (seasonal fodder) and watering. The bulls were housed individually in pens with sufficient cross ventilation and protection against summer heat and in an open space for sunbathing in winter. The management system at 3 (three) AI centres was more or less same.

The information of semen characteristics among 6 (six) breeds of AI bulls was collected on the basis of normal semen collection routine between January 1995 to December 2002, inclusive. Before introduction in collection schedule the bulls were clinically examined. The examination consisted of physical examination and examination of integrity of testes and reproductive tracts as described by Settergren (1983).

The data on semen characteristics were analyzed to see the effect of the scrotal circumference as well as breed effect on scrotal circumference of AI bulls. Semen was collected early in the morning every 4 to 7 days intervals with the aid of an artificial vagina (AV). Total one or two ejaculates were collected from each bull of three AI centres for the preparation of semen dose used for AI programmes. The semen was brought to the laboratory immediately after collection and was placed in water bath at 37°C for evaluation. The characteristics of semen were recorded were as follows: volume of ejaculate (ml), colour (scale: 1-4), density (scale: 1-5), mass activity (scale: 1-5), sperm concentration (million/ml), sperm motility (%), total number of sperm cells/ejaculate (million), total number of motile sperm cells/ejaculate (million), total number of semen doses /ejaculate, post-freezing sperm motility (%) and percentages of total sperm abnormalities.

**Semen Evaluation:** A total 3720 ejaculates (2474 from Central Cattle Breeding Station, Savar, Dhaka and 401 from Rajshahi Dairy and Cattle improvement farm, Rajabarihat, Rajshahi and 855 from District AI centre, and) obtained over the period of 1995 to 2002 to study the semen characters. Immediately after collection semen was placed in a beaker containing lukewarm water and was examined for semen characteristics.

**Semen Characteristics Studied**

**Volume:** Volume of the ejaculate was measured directly from graduated collecting tube and recorded as ml.

**Colour:** The colour of semen was recorded as watery/opalescent, milky white, yellowish and creamy depending on the thickness and pigment the semen and was assigned a numerical weight from 1 to 4 for statistical analysis. A numerical weight of 1 was assigned to opalescent /watery, 2 to milky white, 3 to yellowish white and 4 to creamy white which were determined by eye estimation.

**Density:** The density of semen was scored into 1- 5 scales:

1= watery to cloudy
2= milky
3= thin creamy
4= creamy and
5= creamy grainy were determined also by eye estimation.

**Mass activity/ Wave motion:** The mass activity was evaluated in a drop of fresh undiluted semen placed on a pre-warmed slide without cover-slip at low magnification (100×). The mass activity was scored into 1- 4 scales:

1= weak motion without forming any wave
2= small, slow moving wave
3= vigorous movement with moderate rapid waves and eddies and
4= dense, very rapidly moving waves and eddies.

**Sperm Concentration:** The concentration of spermatozoa (million/ml) in fresh semen was determined by using an improved Neubaur Haemocytometer (Salisbury *et al.*, 1978) or Kara's scale at District AI centre, Rajshahi.

The heamocytometer using red blood count was used. Semen was drawn into the capillary tube of the dilution pipette with the mark of 1. The pipette tip was carefully wiped without drawing from the capillary. The semen was pulled into the bulb and pipette was filled with diluents, usually 0.9% chloride solution (to mark of 101). The mixing pipette was then properly stippled and shaken gently by hand with a quick wrist motion while holding it between the thumb and forefinger. The first 3/4 drops of fluid withdrawn from the pipette was discarded after mixing. The cover glass was pressed firmly down on the slide and a drop of the diluted semen was allowed to flow under the cover glass by capillary action. The examination was delayed for a few minutes to let the cells settled. By convention, spermatozoa lying across the top and the left–hand–side grids of the squares were counted and those on the bottom and right-hand – side grids were ignored. The following formula was used for calculating total number of spermatozoa / ml of fresh semen.

$$N = C \times \frac{400}{S} \times d/ml$$

Where,

N= number of spermatozoa counted per ml of semen.

C= number of spermatozoa counted a given number of small squares.
S= number of small square counted
d= dilution ratio.
The sperm concentration/ml was directly recorded by using a Photometer at Central AI laboratory, CCBSDF, Savar, Dhaka and Deep frozen semen production laboratory, RDCIF, Rajabarihat, Rajshahi.

**Sperm Motility /Initial Sperm Motility:** The motility of sperm was evaluated in small drop of diluted semen placed on a clean, prewarmed slide, covered with a cover slip and examined at a magnification of 400× under light microscope at District AI centre, Rajshahi and Video microscope was used at Dairy and Cattle Improvement Farm, Rajabarihat, Rajshahi and Central cattle breeding centre, Savar, Dhaka. Sperm motility was scored on the basis of the percentage of spermatozoa with normal forward progressive movement while those showing circling movements or oscillating at one place were regarded as immotile (Ahmad, 1994). Two examiners separately follow blind techniques for the evaluation. The average score of sperm motility given by the two examiners were recorded.

**Total number of Spermatozoa / Ejaculate:** Total number of sperm cell per ejaculation was calculated by multiplying the spermatozoon concentration per ml by volume (Bane, 1952).

**Total Number of Motile Sperm Cells /Ejaculate:** From the total number of sperm cells per ejaculates and motility, the number of motile sperm cells/ejaculate was calculated automatically.

**Total Number of Semen Doses per Ejaculate:** From the volume, motility and sperm concentration, total number of semen doses /ejaculate was also calculated automatically. The total number of insemination doses per ejaculate was calculated using the following formula for chilled semen at District AI centre, Rajshahi.
Total No. of insemination dose/ejaculate =
Again, The total number of insemination doses per ejaculate for deep frozen semen was calculated the following formula used at Frozen semen production AI Laboratory.

$$\text{Total insemination dose/ejaculate} = \left\{ \left[ \frac{\text{Volume} \times \text{Sperm conc.}}{240} - \frac{\text{volume}}{2} \right] \times 2 + \text{volume} \times 4 \right\}$$

standards for the concentration, giving different numbers of sperm cells per semen dose (1 ml /0.25 ml). The standards were 20 million sperm cells/AI dose for chilled semen at District AI centre, Rajshahi and 30 million sperm cells/AI dose for frozen semen at CAIL, CCBSDF, Savar, Dhaka and DFSPL, RDCIF, Rajabarihat.

**Post-freezing Motility:** The frozen semen was thawed in a water-bath at 37-39°C for 10 to 12 seconds. The sperm motility was always evaluated at +37°C using phase contrast optics (400x) by two investigators. The motility scores given by the investigators were averaged.

**Percentage of Sperm Abnormalities:** The proportion of abnormal spermatozoa at William's stain and Formol-Saline preparation included those, only had different abnormalities in head, mid-piece and tail defects.

**Morphological Examination of Spermatozoa:** Morphological examination was done by staining the smear of semen with William's method (Williams, 1920; Lagerlof, 1934). A thin smear of fresh semen was prepared on a grease free slide for the study of morphological abnormalities of sperm head after staining with William's stain.

**Staining Procedure:** The smear was air dried and fixed in flame and some information like bull ID and date of semen collection marking on slide at any end with the help of permanent marker pen. Then the smear was treated with absolute alcohol for 3-4 minutes and washed in treated with 0.5% chloramine for 1-2 min. until appeared fairly clear and then washed in distilled water followed by rinsing in 95% alcohol and finally stained with Carbol- fucshin eosin (8-10 min.). After staining, the slides were washed in running tap water, dried off and examined under light microscope at 1000× (oil emersion). The proportion of sperm with normal head morphology included only was free from any detectable abnormalities. The head abnormalities in spermatozoa were classified according to William's (1920) such as pear shape head, narrow at the base head, narrow head, broad, big and little short head, abaxial position of the mid piece, undeveloped head, abnormal contour and others abnormal head. At least 500 spermatozoa from individual smears were examined. The proportion of spermatozoa with abnormal head morphology included only these, which were any detectable abnormalities.
The morphology of sperm mid-piece and tail was studied after fixed with buffered Formol–saline at the same temperature.

**Procedures:** 1 ml of formol-saline was taken in sample glass tube with plastic cork and a very small drop or 10 µml of fresh semen was mixed, shacked and marked the sample with the information like bull ID, breed and date of semen collection for later examination and sample was preserved for a long time in refrigerator (+4°C). The abnormalities of formol-saline fixed spermatozoa were observed under Phase contrast microscopy. The following abnormalities were found in the freshly collected and preserved semen viz. free loose head, abnormal mid-piece, abnormal tail (simple bent tail and coil tail), proximal and distal cytoplasomic droplets and double folded tail, broken neck, abnormal acrosome and others. At least 200 spermatozoa from individual replicates were examined at 1000× magnification. The proportion of abnormal spermatozoa at formol–saline preparation included only those, which had abnormalities in the mid-piece and tail. The spermatozoa were considered as normal having no mid-piece and tail defects.

**Statisical Analysis:** The descriptive statistics on the data on volume, colour, density, mass activity, sperm concentration, sperm motility, total number of sperm cells / ejaculation, total motile sperm cells/ejaculate, no. of semen doses per ejaculates, post-freezing motility of sperm and % of abnormal spermatozoa with regards to the scrotal circumference was performed using SSPS/PC +version –10.01 (SSPS 1999, Microsoft Corp; 1988). Data were used to Compare Mean One-Way ANOVA: Post Hoc Multiple Comparisons by using the

'exclude cases analysis by analysis' for the missing values. Duncan's multiple range was used to detect significant differences between means (Steel and Torrie, 1980). Distributions were expressed as Mean±SD. To evaluate the scrotal circumferences variation on semen traits in AI bull the following model was fitted using mix procedure of SPSS/PC.

$Y_{ijk} = \mu + \alpha_i + \beta_j + e_{ijk}$

Where,
$Y_{ijk}$ - observed semen characteristics and SC measurement
$\mu$ - overall meaná$_i$ – fixed effect of jth scrotal circumference group of the AI bulls (i=1-4)
$\beta j$ – fixed effect of ith genetic group of the bull at collection (j=1-6)
$e_{ijk}$= is a random residual effect
Mean effects were systematically included in the model. Random effects were assumed independently and identically distributed.

## Results and Discussions

A total 3720 samples from 71 bulls belongs to six (6) genetic groups of three (3) AI centres or stations were used. The experiment to evaluate the semen characteristics of AI bulls relation to scrotal circumferences as well as the effect of breed on SC. The average values with standard deviation of volume of ejaculate, colour, density, mass activity, sperm concentration, sperm motility, total number of sperm cells /ejaculate, total number of motile sperm cells /ejaculate, number of semen doses per collection, post-freezing motility and percentages of abnormal spermatozoa were 7.14±2.11ml, 3.23±0.97 (scale: 1-4), 3.56±0.78 (scale: 1-5), 2.91±0.81 (scale: 1-4), 1314.5±312.6 million/ml, 63.95±4.61%, 9375.7±3431.5 million/ejaculate, 6023±2299 million/ejaculate 312.0±115.3, 55.09±5.63% and 16.38±3.2, respectively according to scrotal circumference factors. The results of the semen characteristics and the effect of breed on SC analyzed are presented in Table 1 and 3. The analysis of variance (ANOVA) on the semen characteristics of dependable factor of scrotal circumference is presented in Table 2.

Table 1: Scrotal circumference variation on semen characteristics of Artificial Insemination (AI) bulls

| Semen characteristics | Scrotal circumference groups | | | | |
|---|---|---|---|---|---|
| | <34 cm | 34 to ≤ 36 cm | 36 to ≤ 38 cm | > 38 cm | Overall |
| Volume (ml) | 5.15±1.70 [d] | 6.89±1.86 [c] | 7.45±1.71 b | 8.53±2.07 [a] | 7.14±2.11 |
| | n=563 | n=1171 | n=1226 | n=760 | n=3720 |
| Colour(1-4)* | 3.26±0.96 | [a]3.28±0.79 [a] | 3.15±0.98 [b] | 3.26±0.98 [ab] | 3.23±0.97 |
| | n=563 | n=1171 | n=1226 | n=760 | n=3720 |
| Density | 3.38±0.87 [b] | 3.59±0.82 [a] | 3.57±0.73 [a] | 3.64±0.73 [a] | 3.56±0.78 |
| (D; 1-5 scale)** | n=563 | n=1171 | n=1226 | n=760 | n=3720 |
| Mass activity | 2.83±1.21 [b] | 3.02±0.81 [a] | 2.89±0.66 [b] | 2.82±0.62 [b] | 2.91±0.81 |
| (1-5 scale)*** | n=563 | n=1171 | n=1226 | n=760 | n=3720 |
| Sperm concen- | 1238.8± 335.6 [b] | 1341.1± 340.0 [a] | 1316.8±287.2 | 1325.7± 278.7 [a] | 1314.5± 312.6 |
| tration (×10$^6$) /ml | n=563 | n=1171 | n=1226 | n=3720 | n=760 |
| Sperm motility (%) | 63.60±5.37 | [b]364.51±4.27 | [a]63.47±4.22 [b] | 64.11±5.00 [a] | 63.95±4.61 |
| | n=56 | n=1171 | n=1226 | n=760 | n=3720 |
| Total sperm cells | 6418.8±2818.2 [d] | 9142.8±3089.0 [c] | 9796.7±3060.4 [b] | 1125 ± 3410.6 [a] | 9375.7±3431.5 |
| / ejaculate (×10$^6$) | n=563 | n=1171 | n=1226 | n=760 | n=3720 |
| Total motile sperm | 4150±1995 | [d]5914±2078 [c] | 6236±2048 [b] | 7235±2313 [a] | 6023±2299 |
| cells / ejaculate (×10$^6$) | n=563 | n=1171 | n=1226 | n=760 | n=3720 |
| No. of semen doses | 212.5±74.9 [d] | 304.2±103.9 [c] | 326.3±102.8 [b] | 374.8±114.5 | [a]312.0±115.3per |
| collection | n=563 | n=1171 | n=1226 | n=760 | n=3720 |
| Post-freezing | 55.46±5.96 | 54.94±5.87 | 54.87±5.34 | 55.46±5.45 | 55.09±5.63 |
| motility (%) | n=737 | n=760 | n=452 | n=309 | n=2258 |
| % of sperm | 16.76±3.1 [a] | 16.39±3.9 [a] | 15.73±2.8 [b] | 17.00±2.3 [a] | 16.38±3.2 |
| abnormalities | n=101 | n=156 | n=150 | n=497 | n=90 |

(1-4)*; 1=Opalescent, 2=Milky white, 3= yellowish white and 4=creamy white. (1-5)**; 1=watery, 2=milky, 3=mild creamy, 4=creamy and 5=creamy grainy. (1-5)***; 1= weak motion without forming any wave, 2=small, slow moving wave, 3=vigorous movement with moderate rapid waves and eddis and 5= dense, very rapidly moving wave and eddis. N= Number of observation, The values are Mean±SD, a,b,c d Mean±SD with different superscript letters in the same row differs significantly with each others (P<0.05).

Mean±SD with comparisons of semen characteristics according to sizes of scrotal circumferences (SC) is shown in Table 1. Scrotal circumference of the AI bulls had significant (P<0.05) effect on all the semen characteristics except post-freezing motility of sperm (Tabe-1). The highest volume of ejaculate (8.53±2.07 ml) was found in SC groups of >38 cm. On the other hand, the volume of ejaculate was lowest (5.15±1.70 ml) of the SC groups of <34 cm. Volume, total sperm cells/ejaculate, total number of motile sperm cells/ejaculate and number of semen doses per collection were gradually increased with increasing sizes of SC groups of bulls. Colour and density, sperm concentration, sperm motility and post-freezing motility varied from 3.15-3.28 (scale: 1-4), 3.38-3.64 (scale: 1-5), 1238.8-1341.1 million/ml, 63.47-64.51% and 54.84-55.46%, respectively depending on the sizes of testes of AI bulls. The data on percentage of abnormalities of spermatozoa for different SC groups of bull obtained in the present study is also presented in Table 1. Analysis of variance showed that SC had significant effect on total sperm abnormalities (Table 2). The variations on the % of total sperm

abnormalities in different SC groups of bulls were found to be significant. The highest average percentage of total sperm abnormalities was recorded (17.00%) in >38cm in SC groups by followed by <34 cm, 34-36 cm and 36-38 cm of SC groups (16.76%, 16.39% and 15.73%, respectively).

The Mean+SD with comparison of scrotal circumference in 6 (six) genetic groups of AI bulls is shown in Table 3. Analysis of variance indicated that genetic groups had significant (P<0.05) effect on scrotal circumference (Table 4).

Table 2: Analysis of variance for the semen characteristics of different scrotal circumference (SC) groups of bulls used for AI in Bangladesh

| Semen characteristics | Sources of variation | D.F | Sum of square | Mean of square | F-value |
|---|---|---|---|---|---|
| Volume (ml) | Between SC groups | 3 | 3897.2 | 1299.0 | 380.5 |
| | Within SC groups | 3716 | 12683.5 | 3.4 | |
| Colour(1-4)* | Between SC groups | 3 | 10.6 | 3.5 | 3.7 |
| | Within SC groups | 3716 | 3507.7 | 0.9 | |
| Density(1-5 scale)** | Between SC groups | 3 | 23.2 | 7.7 | 12.7 |
| | Within SC groups | 3716 | 2260.6 | 0.6 | |
| Mass activity | Between SC groups | 3 | 23.9 | 7.9 | 12.1 |
| (1-5 scale)*** | Within SC groups | 3716 | 2436.2 | 0.6 | |
| Sperm concentration | Between SC groups | 3 | 4156345.1 | 1385448.35 | 14.31 |
| ($\times10^6$) /ml | Within SC groups | 3716 | 359256352.6 | 96678.2 | |
| Sperm motility (%) | Between SC groups | 3 | 739.2 | 246.4 | 11.70 |
| | Within SC groups | 3716 | 78263.7 | 21.0 | |
| Total sperm cells | Between SC groups | 3 | 7861420479.6 | 2620473493.3 | 271.0 |
| / ejaculate ($\times10^6$) | Within SC groups | 3716 | 35930880235.2 | 9669235.8 | |
| Total motile sperm | Between SC groups | 3 | 3160175958.6 | 1053391986.2 | 237.2 |
| cells / ejaculate ($\times10^6$) | Within SC groups | 3716 | 16496447438 | 4439302.3 | |
| No. of semen doses | Between SC groups | 3 | 8884127.3 | 2961375.7 | 271.0 |
| per collection | Within SC groups | 3716 | 40595396.1 | 10924.4 | |
| Post-freezing motility | Between SC groups | 3 | 157.835 | 52.612 | 1.663 |
| (%) | Within SC groups | 2254 | 71328.914 | 31.645 | |
| No. of total sperm | Between SC Groups | 3 | 44.994 | 14.998 | 3.293 |
| abnormalities | Within SC Group | 493 | 5199.038 | 10.546 | |

Table 3: Scrotal circumference differences according to 6 (six) genetic groups of bulls used for AI programme in Bangladesh

| Breeds of Bull | No. of observation | Scrotal circumference (cm)100% |
|---|---|---|
| Friesian | 626 | 38.0±0.7 [a] |
| 100% Sahiwal | 620 | 35.4±0.9 [d] |
| 75% Friesian-25% Local | 784 | 36.4±1.8 [c] |
| 50%Friesian -50% Sahiwal | 572 | 37.3±1.1 [b] |
| 50%Friesian-50% Local | 769 | 35.7±2.2 [d] |
| 100% Local | 349 | 33.9±1.2 [e] |
| Total | 3720 | 36.3±1.9 |

Scale 5 was the best and
1 was the worst. The values are Mean±SD, a,b,c,d,e Mean±SD with different superscript letters in the same column differs significantly with each others (P<0.001).

Table 4: Analysis of variance for the scrotal circumference of genetic groups of different bulls used for AI in Bangladesh.

| Parameters | Sources of variation | D.F | Sum of square | Mean of square | F-value |
|---|---|---|---|---|---|
| Scrotal | Between Genetic groups | 5 | 5308.5 | 1061.7 | 446.0*** |
| circumference (cm) | Within Genetic groups | 3714 | 8839.7 | 2.3 | |

***=P<0.001

## Results and Discussions

From the study of this report, SC of the AI bulls had significant (P<0.05) effect on all the semen characteristics except post-freezing motility of sperm. The highest volume, density, total sperm cells/ejaculate, total number of motile sperm cells/ejaculate and number of semen doses per collection were gradually increased with increasing sizes of the testicular circumference and color, mass activity and sperm motility after 36-38 cm groups of SC groups of bulls. Similar studies were made by Sarder *et al.*, (2001). They observed that the volume of ejaculate for <32 cm, 32-34 cm, 34-36 cm and >36 cm groups were 6.01±0.36, 7.39±0.21, 6.31±0.54 and 9.08±0.30ml respectively. The increase of the semen traits was probably due to the size of bull testes (Almquist, 1978). Bull tended to have small scrotum, less percentage of motile spermatozoa and higher proportion of abnormal spermatozoa is classified as questionable breeders (Spitzer *et al.*, 1988). Bull with small testes, have poor quality semen and after fail to produce no ejaculate in 4 separate Electro-ejaculate attempts, which are classified as unsatisfactory breeders (Spitzer *et al.*, 1988). The differences between SC groups of bulls may be attributed to the variation their scrotal circumference, bred, age body weight and body size and the scretory activity of the sex glands (Leon *et al.*, 1991 and Sharma *et al.*, 1991).

The mean scrotal circumference observed during the present study was varied from 33.9+1.2 to 38.0+0.7cm, which is same from 34.00 to 39.00cm reported by Sarder (2002) for the Sahiwal bull. Silva-Mina *et al.*, (2000) also reported that the SC of Brahman and Nelore

bulls were 36.22±0.90 cm and 32.3±0.38cm respectively. Variation in SC in different reported might be due to effect of breed, nutrition, season and perhaps due to number of bull included in different studies.

Results of my study indicated that scrotal circumference of >38 cm group of bulls were suitable for better performance of semen characteristics. Again, 100% Friesian breed was more appropriate for the best scrotal circumference of AI bulls among other groups of breed.

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