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Testicular Biometry for Breeding Soundness Evaluation of Sahiwal Males Reared under Tropical Conditions

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ABSTRACT

Background: The testicular biometry such as scrotal circumference (SC) and scrotal volume, affect semen quality hence bull fertility and thus testicular biometry can be used for breeding soundness of bulls.

Methods: The present study was carried out to study the testicular parameters of 77 Sahiwal males of different ages and to establish its importance for selection of breeding bulls during 2015-2016 at ICAR-NDRI, Karnal. The testicular biometry of each male was done at monthly intervals using Vernier calliper, cloth tap and ultrasonography.

Result: The semen quality parameters were significantly (P<0.05) more in the group having higher scrotal circumference. The scrotal circumference follows the curvilinear manner of growth from 12 months to >72 months (correlation value of 0.92 and R^2 0.84). The paired testicular volume (PTV) differed significantly (P<0.05) between different age group males. Scrotal circumference and paired testicular volume are directly related to semen quality hence bull fertility. Thus, testicular biometry is an important criterion for breeding soundness for selecting Sahiwal males.

Key words: Breeding soundness, Paired testicular volume, Sahiwal bulls, Scrotal circumference, Testicular biometry.

INTRODUCTION

The Sahiwal is one of the best native milch breed of India and Indian sub-continent. Sahiwal is adaptable to a high degree of heat tolerance, resistant to tick and ability to survive on poor quality feed and fodder. Selection of quality bulls is the foundation of profit from a herd. According to Sprott *et al*. (2003) both high fertility and high genetic breeding value for one or more economically important characteristics, such as growth, calving ease, milk production and milk quality is offered by a good bull. Lots of changes have come into the picture from that time to make breeding soundness examination more scientific and predictive. But in India there is no proper protocol for breeding soundness evaluation of Sahiwal bulls. Nowadays, male is gaining popularity due to their contribution towards genetic improvement as we all are aware of the fact that the concept “bull is half of herd” has changed with the truth that “bull is more than half of herd”. Proper emphasis on monitoring; starting from young age, introduction at the right time, training and semen collection along with their functional andrological examination may help us in a better selection of Sahiwal breeding bulls. Testis size and its function are critical for bull fertility (Kastelic, 2014) and SC is an integral component of a breeding soundness evaluation (Kastelic *et al*., 2012). It is well known that scrotal circumference (SC) and semen quality are highly correlated with fertility (Parkinson, 2004). SC varies according to the age of the bull (Kastelic *et al*., 2012). Testicular biometry (SC) could be used to predict the onset of puberty, whereas age, weight and SC are equally good predictors of sexual maturity in B. *indices* bulls (Kastelic, 2014). So, the current study was taken to develop protocol for selection of indigenous dairy bulls for breeding soundness.

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MATERIALS AND METHODS

The present investigation was conducted on 77 Sahiwal males of different ages from January to December, 2015 (1 year). Adult Males >3 year semen donating (N=20), Adult Males >3 year, non-semen donating (N=23), Young males <3 year (N=8) and Calves <0.5 year (N=26). The adult bulls were maintained at Artificial Breeding Research Centre, ICAR-NDRI, Karnal, Haryana. The male calves were maintained at Livestock Research Centre, ICAR-NDRI - Karnal, Haryana for six months and then shifted to ABRC, ICAR-NDRI. The work was carried out at these two farms of the institute. The bulls were reared at Artificial Breeding Research Centre, ICAR-NDRI, Karnal under loose housing system. Bulls were kept in concrete floored individual pens (30×10 feet) with corrugated asbestos roofed shed. Concentrate ration with 21 percent CP and 70 percent TDN

@ 2.0 to 2.5 kg per bull. Green fodder was supplied throughout the experimental period. Water was provided ad lib. Herd-health programme was followed as per the farm schedule to ensure good health. Bulls were exercised once a week, the day before semen collection in the rotary exerciser, so as to maintain the sexual vigour of bulls and ensure quality semen production. The semen was collected by artificial vagina technique during morning hours.

Biometry of scrotum and testes

The scrotal circumference of each bull was measured at monthly intervals while bulls were restrained in standing position in a metallic crush. At the time of testicular biometry in the morning 10 O'clock and animal handling personnel were kept constant in order to maintain monotony. The testes were brought into the distal part of the scrotum until the ventral scrotal skin folds, if any, were eliminated. Then testes were held firmly in place by grasping the scrotum with left hand above the head of the epididymis. The right hand was then used to guide the loop of a flexible measuring tape around the scrotum (Pant *et al*., 2003). Scrotal circumference was measured with sufficient manual pressure on the measuring tape to cause slight skin indication (Ahmad, 2005). The length (proximal-distal) and width (mediolateral) of each testis, scrotal skin fold thickness (SSFT) was measured with the help of Vernier caliper, keeping in mind not to modify the normal shape of the organ (Pant *et al*., 2003), the averages for two testes of each bull were calculated.

Diagnostic Ultrasonography with Linear probe (KAIXIN KX 2600) B-mode ultrasound scanner was used to measure scrotal and testicular dimensions to increase accuracy. The bull was restrained properly and the scrotum was clean shaved and the linear probe was palpated over the scrotal surface to obtained scans of testes and scrotum. Each testis was examined by placing the transducer vertically (parallel to the long axis of the testes) and on the caudal aspect of the scrotum.

The testicular biometry measurements done in all the males were scrotal circumference (SC), Average testicular length (TL), Average testicular width (TW), Paired Testicular volume (PTV) and Scrotal skin thickness (SST). The PTV was calculated as per Lunstra and Cundiff (2003), PTV= 0.0396 (Avg. TL) $(SC)^2$.

Statistical analysis

The calculations of means and standard deviations (SD) of semen traits were calculated with SAS® 9.2, SAS Institute Inc., Cary, NC, USA. For all analysis, differences were considered significant when P values were ≤ 0.05. Data was analyzed by one-way ANOVA, followed by means comparisons for all pairs using Tukey-Kramer HSD test.

RESULTS AND DISCUSSION

It was found that during measurement of scrotal skin thickness, Vernier callipers were painful, due to that the animals were getting disturbed which lead into errors in measurements whereas USG was painless and measurements were more accurate. The overall testicular biometry of Sahiwal bulls *i.e*. SC, average testicular length, average testicular breadth and paired testicular volume (PTV) are given in Table 1. There were no significant differences in testicular biometry between good and poor semen quality bulls (Table 1).

Bulls were grouped into three groups based on the scrotal circumference (<34 cm, 34-36 cm and >36 cm). The mass activity (MA), live sperm%, total sperm abnormalities and sperm concentration (billion /ml), were significantly (P<0.01) more in the bulls having higher scrotal circumference (>36 cm) as compared lower SC group (<34 cm) Table 2. Similarly, the HOST% were significant (P<0.05) between lower and higher SC group. The total sperm abnormality was significantly (P<0.05) less in the higher SC group (>36cm) than the lower SC group (<34cm). The semen volume and intact sperm% were more in higher scrotal circumference group of bulls but statistically non-significant (Table 2).

The scrotal circumference showed an increasing trend with the advancement of age. The scrotal circumference differed significantly (P<0.05) between 12 month age group and 24 months, 48 month age group (6.70±0.44, 21.93±1.98 and 33.52±0.89 cm). After 48 months there was no significant difference in scrotal circumference with the advancement of age. The SC follows the curvilinear manner of growth from 12 months to >72 months. The correlation value was 0.92 and R^2 was 0.84 (Fig 1).

The testicular biometry significantly (P<0.05) changed between the different age groups. The SC, average testicular length, average testicular breadth and PTV differed significantly between adult male (>3year), young male <3 years and calf <0.5 years Table 3. The testicular biometry significantly (P<0.05) changes during calf hood to 0-6 months. The skin fold thickness (SFT) does not differ significantly during 0-6 months.

In our study it was presumed that testicular biometry has relationship with fertility as Mahmood *et al*. (2014) found average length (L), average width (W) and overall PTV of testes of Cholistani bulls as 16.91±0.70 cm, 7.05±0.35 cm and 874.37±137.07 cm^3, respectively, which indicates that length in our case was lower, but the PTV is more than our results. It is similar to Lunstra and Cundiff (2003) for Angus bulls who found mean PTV of 618±24 cm^3 but our results

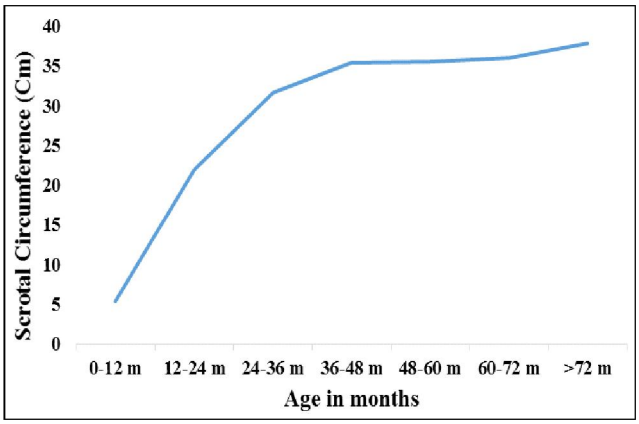

Fig 1: Relationship of age with scrotal circumference (P<0.05 N=77).

Table 1: Relationship of testicular biometry (ME±SE) of sahiwal bull with semen quality.

Parameter	Good semen quality bulls (N=9)	Poor semen quality bulls (N=11)	Overall of bulls (N= 20)
SC (cm)	36.39±1.11	35.69±0.85	36.01±0.67
Length (cm)	12.74±0.50	12.67±0.24	12.82±0.28
Breadth (cm)	12.74±6.62	6.38±0.38	9.44±6.38
PTV (cm^3)	680.97±64.88	644.15±37.03	666.48±34.78

SC scrotal circumference, PTV paired testicular volume.

Table 2: Relationship of scrotum circumference (ME±SE) with semen quality.

Parameters	Scrotal circumference		
	<34 cm (N=6)	34-36 cm (N=6)	>36 cm (N=6)
Volume	4.14±0.54	4.60±0.26	4.76±0.5
Mass activity	1.44^{a}±0.27	2.55^{ab}±0.19	2.92^{bc}±0.17
Live sperm%	54.83^{a}±3.16	66.19^{ab} ±3.01	71.31^{bc}±1.87
HOST%	53.56^{a}±1.44	61.23^{ab}±2.05	65.75^{bc}±2.62
Acrosome integrity%	92.18±0.41	93.05±0.55	93.91±0.44
Total sperm abnormality%	11.66^{a}±1.32	7.78^{ab}±0.5	7.50^{bc}±0.61
Sperm concentration (10^6/ml)	916.45^{a}±36.03	1054.88^{ab}±62.52	1177.77^{bc}±51.34

Superscript significant at P<0.05.

Table 3: Relationship of age with testicular biometry (ME±SE) of sahiwal bulls.

Group	SC (cm)	Length (cm)	Breadth (cm)	PTV (cm^3)
Adult males >3year (N=20)	36.01^{a}±0.67	12.82^{a}±0.28	9.44^{a}±2.95	666.48^{a}±34.78
Young males <3- year (N=8)	31.43^{b}±0.92	11.23^{b}±0.44	5.92^{b}±0.22	446.53^{b}±39.62
Calves <0.5 year (N=26)	4.79^{c}±0.04	4.61^{c}±0.03	1.89^{c}±0.08	45.12^{c}±0.82

SC: scrotal circumference, PTV: paired testicular volume, Superscript significant at P<0.05.

were higher 550±18 cm^3 than Brahman bulls, as were reported by Lunstra and Cundiff (2003). These differences in results may be due to breed and age differences. In contrary Rajak *et al.* (2013) reported that in good bulls, the testicular parameters were higher than poor bulls. The non-significant difference between the two groups of bulls suggests that level of exercise and genetic may have also their effect on semen quality. Madrid *et al.* (1990) found a significant correlation between scrotal circumference with age, body weight, ejaculate volume, sperm concentration, sperm motility, normal percentage and with abnormal spermatozoa.

Measurements of the average length (L) and average width (W) are important orchidometer parameters, as the testicular morphology influence testicular thermoregulatory capability and affect semen quality and sperm production in bulls (Brito *et al*., 2004). The PTV has been considered as a better selection marker for reproductive assessment of Zebu cattle as compared to SC (Unanian *et al*., 2000). Positive correlation of PTV with BW, SC and average testicular width shows its role as a reliable parameter for prediction of reproductive efficacy in Cholistani bulls. The bulls in our study were having a narrow age difference, which results in less variation between good and poor semen quality bulls.

Our results regarding scrotal circumference and semen quality are in consonance with Waldner *et al*. (2010) who reported a lower percentage of morphologically normal sperm in bulls with an SC <34 as compared to SC ≥34 cm. Kastelic (2014) reported that bulls with larger SC usually have a higher percentage of morphologically normal sperm as is in our case. Similarly Spitzer *et al*., (1998) found a high correlation of (0.81) between SC and sperm output. It is well known that scrotal circumference (SC) and semen quality are highly correlated with fertility (Parkinson, 2004). The larger volume of testis will result more vasculature of testis, the blood supply will be more to site of sperm production, which ultimately might result more quality sperm production.

Our results regarding relationship of scrotal circumference with age are in agreement with Addass *et al*. (2013), who found significant age variability of the scrotal circumference with the age group of bull ≥ 48 months had highest. In similar line Ahmad *et al*. (2011) reported curvilinear SC growth from 12 months to >72 months of bull. Our values are lower than values reported by Entwistle and Fordyce (2003) in *Bos indicus* bulls at 12-15, 18 and ≥24 months as 24, 28 and 30 cm. SC could be used to predict the onset of puberty, whereas age, weight and SC are equally good predictors of sexual maturity in *B. indicus* bulls (Kastelic, 2014). SC increases with increasing age, the

minimum SC varies according to the age of the bull (Kastelic *et al*., 2012). Both age and BW were highly correlated to the scrotal circumference (r =0.81 and 0.82, respectively) (Sosa *et al*., 2002). Bulls usually reach puberty around 12-14 months of age, weight about 350-450 kg and have a scrotal circumference of 26 cm (Geske, 2003). The testis size is known to be significantly affected by age of the animal, being smaller in young than the adult or old animals (Younis *et al*., 2003). Morphology and motility were ≥50% each in 91% of the bulls between ages 12 and 20 months (Sosa *et al*., 2002). The testis grow rapidly as indicated by the increase in SC up to an age of 2 years and thereafter the growth is rather slow (Ghoneim *et al*., 1993). Moghaddam (2012) found that positive correlation scrotal circumference with reaction time, spermatozoa concentration. These all results are in the similar line to that of our results. Testis size and its function are critical for bull fertility (Kastelic, 2014) and SC is an integral component of a breeding soundness evaluation (Kastelic *et al*., 2012). In weaner and yearling bulls, physical traits such as scrotal size and sheath size are at least moderately to highly correlated with later life values from 10 months onwards (Holroyd *et al*., 2007).

Our results regarding testicular biometry in relation to age are similar to that of Ahmad *et al*. (2011) who found a significant effect of age on testicular biometry, but the values of PTV at different ages were higher than our results. Testicular morphology influences testicular thermoregulatory capability and affects semen quality and sperm production in bulls (Brito *et al*., 2004). Similarly, scrotal circumference, scrotal thickness and testicular length increase linearly with the age from 1 to 24 months in cattle male calves (Suri, 1993). It thus indicated testicular biometry influenced fertility of bulls.

CONCLUSION

The scrotal circumference and pair testicular volume are related with age and semen quality in other word bull's fertility, thus it can be concluded that testicular biometry particularly scrotal circumference is important criteria for selecting Sahiwal males for breeding soundness evaluation.

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Conflict of Interest: None.

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