



## Quantification of Sexual Behaviour Traits in Presence of Teaser Male in Relation to Libido and Semen Quality in Buffalo Bulls

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### ABSTRACT

Screening and selecting bulls for desirable reproductive traits and high libido is known to improve the reproductive performance of the herd. Evaluation of sexual behaviour and semiogram is very useful in demarcating the high fertility bulls. Therefore, the purpose of the present study was to quantify the sexual behaviour in presence of male teaser in relation to semen quality in buffalo bulls. Bulls were categorized into two groups (high and low libido) on the basis of reaction time. The average duration of first licking of prepuccial area was significantly ( $p < 0.05$ ) lower in high libido bulls as compared to low libido bulls. The average frequency and overall duration of licking of prepuccial area until ejaculation in high libido buffalo bulls was significantly ( $p < 0.05$ ) lower as compared to low libido bulls. The average time lag to show flehmen response following sniffing and licking the prepuccial area and urine of teaser male was similar ( $p > 0.05$ ) in high and low libido buffalo bulls. The average chin resting time were similar ( $p > 0.05$ ) in high and low libido buffalo bulls. Individual sperm motility (%) and viability (%) was found to be significantly higher ( $P < 0.05$ ) in high than the low libido bulls. Sperm abnormality (%) was significantly higher ( $P < 0.05$ ) in low libido bulls. Therefore, it can be concluded that breeding buffalo bulls can be demarcated for high libido based on quantification of sexual behaviour traits in relation to semen picture.

**Keywords:** Buffalo bull, Libido, Sexual behaviour, Semen quality

Evaluation of sexual behaviour as well as semiogram is very useful in identifying high fertility bulls. Breeding soundness evaluation based analysis of sexual behaviour traits has been proved in economizing the dairy industry. Further, various sexual behaviour traits and semen quality parameters has been found to be helpful in predicting reproductive performance of breeding bulls (Kumar *et al.*, 2008). It has been observed that almost one fourth of breeding bulls maintained at bull stations are affected with poor libido (Kumar *et al.*, 2008). Hence, characterization and quantification of sexual behaviour traits of buffalo bulls in relation to semen quality warrants attention and investigation. The behavioural characteristics of bulls while detecting and serving cows in estrus is poorly understood (Galina *et al.*, 2007). Bulls having high sexual pursuits show better penile erection, penile protrusion, ejaculatory thrust, libido and mating ability. Presently,

little information is available on quantified sexual behavior traits in breeding buffalo bulls. Therefore, present study was conducted to quantify the sexual behaviour traits of buffalo bulls in presence of male teaser in relation to libido and semen quality.

### MATERIALS AND METHODS

The present study was conducted on buffalo bulls (Murrah, N=32) between 5–10 years of age, being maintained loosely in half walled concrete sheds in individual pens (covered area of 12 × 10 ft and uncovered area of 25 × 10 ft) at bull station, Guru Angad Dev Veterinary and Animal Sciences University (GADVASU), Ludhiana, India (Latitude/Longitude, 30.55°N, 75.54° E) and Sperm station (MilkFed), Bhattian, Khanna, Punjab (30.7071° N, 76.2170° E). All the animals were being fed according

to standard feeding schedule along with *ad libitum* green fodder. The bulls were given an exercise for half an hour on alternate days. On the day of semen collection, breeding buffalo bulls was allowed to sexually excite in presence of a male teaser within fenced collection area. Three false mounts were given before semen collection and later semen was collected using artificial vaginal method. Videography (Sony Handycam, HDR CX-240) of semen collection procedure was conducted right from first exposure to sexual stimuli in semen collection area until ejaculation. The video signals were transferred to a computer and video streamings were analysed (Windows 10, Movie maker software) for different sexual behaviour traits with respect to time elapsed. Reaction time was calculated by analysing the video streamings as time from first exposure of sexual stimuli until ejaculation. Bulls were categorized as high libido (Group 1, N=22, reaction time < 5 minutes) and low libido (Group 2, N=10, reaction time > 5 minutes). For the comparative quantification of sexual behaviour traits in both the groups, 10 buffalo bulls from Group 1 (random selection) and all the bulls from Group 2 were selected. Sexual behaviour traits of bulls viz. reaction time, time taken to lick prepucial area after sniffing / licking perineal area, duration of licking of prepucial area, frequency of licking of prepucial area until ejaculation, Flehmen response after sniffing/licking urine and prepucial area were analyzed from recorded video streaming .

Soon after the semen collection, ejaculate was placed in a water bath at 35°C. Semen was evaluated for volume and mass activity, individual motility, progressive motility, viability and total abnormalities of sperm. Semen was diluted in Tris Egg yolk extender and straws were filled, sealed and kept for equilibration at 4°C for 4 hours. Then, the straws were frozen at ultralow temperature using vapour freezing method. Retrospective analysis of data was done to calculate the discard rate of frozen semen as the proportion of frozen semen below the acceptable post thaw motility (>50%) subjected to cryopreservation.

### Statistical analysis

Data were analysed using students 't' test. The data was presented as mean ± SE. The significant interaction was considered at P<0.05.

## RESULTS AND DISCUSSION

In the present study, quantification of the sexual behaviour traits exhibited by breeding buffalo bulls in presence of a teaser male was carried out. The data on sexual behaviour traits were analysed and presented in Table 1.

**Table 1:** Sexual behaviour traits exhibited in high and low libido bulls

Sexual behaviour traits	High Libido Bulls	Low Libido Bulls
	N=10	N=10
Duration of first licking of prepucial area (seconds)	6.9 ± 0.57*	11.9 ± 0.53
Frequency of licking of prepucial area until ejaculation.	1.5±0.17*	2.8±0.89
Overall average licking duration of prepucial area (seconds)	7.1±0.28*	16.5±0.71
Time gap to show flehmen response after sniffing of prepucial area (seconds)	3.2±0.25	3.9±0.23
Resting of the chin on the back of teaser bull after ejaculation (seconds)	3.2±0.32	3.1±0.27

Our study revealed that sniffing and licking of perineal area of teaser male was observed as the first trait in both high and low libido buffalo bulls (Fig. 1A), followed by nudging and protrusion of penis (Fig. 1B). The protrusion of penis was less frequent in low libido (30%) bulls as compared to high libido (70%) buffalo bulls. After that, buffalo bulls sniffed and licked prepucial area of teaser bull a number of times (Fig. 1C). The average duration of first licking of prepucial area in high libido bulls was significantly (p<0.05) lower (6.9 ± 0.57 seconds) as compared to 11.9 ± 0.53 seconds in low libido bulls. The average frequency of licking of prepucial area until ejaculation in high libido buffalo bulls was significantly (p<0.05) lower (1.5±0.17) as compared to low libido bulls (2.8±0.89). The overall average duration of sniffing and licking of prepucial area in high libido bulls was significantly (p<0.05) lower (7.1±0.28 seconds) as compared to low libido buffalo bulls (16.5±0.71 seconds). The sniffing and licking of urine voided by teaser bull was the specific sexual trait in breeding buffalo bulls (Fig. 1D). The average time lag to show flehmen response following sniffing and licking the prepucial area and urine of teaser



A: Sniffing of perineal area



B: Nudging and protrusion of penis



C: Licking of prepucial area



D: Sniffing and licking urine



E: Flehmen response



F: Chin resting after ejaculation

male was similar ( $p>0.05$ ) in high ( $3.2\pm 0.25$  seconds) and low libido ( $3.9\pm 0.23$  seconds) buffalo bulls (Fig. 1E). Before mounting, breeding buffalo bulls sniffed the perineal area again followed by resting of chin on rump of teaser male (Fig. 1F). Chin resting on the rump of teaser bull immediately after ejaculation was the peculiar trait found in breeding buffalo bulls. The average chin resting time were similar ( $p>0.05$ ) in high and low libido buffalo bulls ( $3.2\pm 0.32$  vs  $3.1\pm 0.27$  seconds, respectively). It was also observed that low libido bulls showed poor

interest in presence of restrained teaser bull. However, free movement of teaser bull helped in improved sexual excitement in low libido bulls under similar conditions. Whistling by the bull handler and watching other bulls during semen collection led to increased sexual interest and hastened semen collection in low libido bulls. Limited information pertaining to sexual behaviour traits of buffalo bulls is available. Singh *et al.* (2014) reported a significantly high positive correlation between sexual aggressiveness and penile protrusion, libido and mating

ability. Kundhi buffalo bulls after approaching teaser showed tactile stimulations like sniffing (38.19%), licking (36.11%) and flehmen posture (43.5%) (Samo *et al.*, 2005). Chin resting (14.68%) and dismounting time from 5 to 15 sec was reported (Samo *et al.*, 2005). Significant negative correlation of reaction time and total time taken in mounts with libido and sexual behaviour score was found by Singh *et al.* (2015).

Data on semen evaluation parameters were analysed and presented in Table 2. In our study, individual sperm motility (%) was found to be significantly higher ( $P < 0.05$ ) in high ( $73 \pm 0.81$ ) than the low ( $64.5 \pm 2.16$ ) libido bulls. Similar observation was also reported by Kumar *et al.* (2008). So, high individual sperm motility is indicative of high libido bulls and can be used to demarcate low libido buffalo bulls, though libido is a multifactorial trait. Positive correlation was also found between libido and motility of semen (Pineda *et al.*, 2000). Viability (%) of sperms were found to be significantly higher ( $p < 0.05$ ) in high ( $80.2 \pm 0.78$ ) as compared to low ( $70.3 \pm 2.58$ ) libido bulls. It had been found that positive correlation exist between reaction time and viability of sperms (Shukla and Mishra, 2005).

**Table 2:** Fresh semen quality traits in relation to libido in buffalo bulls

Buffalo bulls	Reaction time (Minutes)	Individual sperm motility (%)	Sperm viability (%)	Total sperm abnormalities (%)	Frozen semen discard rate (%)
High libido	$3.83 \pm 0.17$	$73 \pm 0.81^*$	$80.2 \pm 0.78^*$	$20.8 \pm 0.32$	$7.88 \pm 3.12$
Low libido	$7.25 \pm 0.23$	$64.5 \pm 2.16$	$70.3 \pm 2.58$	$26.7 \pm 1.21^*$	$12.21 \pm 3.84$

So, better sperm viability (%) is indicative of high libido in buffalo bulls. Abnormality (%) of sperms was found to be significantly lower ( $p < 0.05$ ) in high ( $20.8 \pm 0.32$ ) than low ( $26.7 \pm 1.21$ ) libido bulls. The frozen semen discard rate in low libido buffalo bulls was non significantly higher ( $p > 0.05$ ) as compared to high libido buffalo bulls ( $12.21 \pm 3.84$  vs  $7.88 \pm 3.12$ , respectively). Similar observations for viability, abnormality and freezability of semen in good and poor libido buffalo bulls were also

reported by Kumar *et al.* (2008). Relationship between libido and seminal characteristics is a useful index for the selection of the males (Moghaddam *et al.*, 2012).

## CONCLUSION

High libido breeding buffalo bulls display almost all the sexual behaviour traits for less duration than the low libido bulls. Fresh semen quality parameters were significantly better in high libido bulls. No significant differences were observed in the discard rate of frozen semen between the high and low libido buffalo bulls. Therefore, it can be concluded that breeding buffalo bulls can be demarcated for high libido based on quantification of sexual behaviour traits in relation to semen picture.

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