Characteristics of semen collection floors and effect on sexual behaviour and semen quality of Murrah bulls

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ABSTRACT

The present study was carried out on Murrah breeding bulls maintained at artificial breeding complex of NDRI, Karnal and JV Degree College, Baraut, Baghpat. The investigation was aimed to assess the reproductive activity of Murrah bulls on the basis of sexual behaviour characters and seminal attributes as a result of introduction of management interventions. To study the effect of collection floor, three types of semen collection floors viz., concrete floor, brick clay floor and rubber mattress were provided. Data recorded on sexual behaviour and seminal attributes were statistically analyzed. The results indicated that bulls were very active and had higher sexual stimulation before semen collection on rubber mattress followed by brick clay floor and concrete collection floor as reflected by reaction time, dismounting time, total time taken in mounts and other behavioural manifestations. Erection score, protrusion score, intensity of thrust, temperament score and libido score were highly influenced by the type of floor. Analysis of seminal attributes revealed highly significant variation in ejaculate volume between bulls exposed to different types of collection floor. Rubber mattress showed significantly higher value of semen volume which confirmed its superiority in semen collection over other types of floors. However, physical appearance, mass activity, initial progressive motility, non eosinophilic spermatozoa count, post thaw motility, hypo osmotic swelling test were not significantly affected by the type of floor. Thus the rubber mattress collection flooring helps in improving the semen output without having any detrimental effect on semen quality.

Key Words: Murrah buffalo bull, collection floor, sexual behaviour, semen quality.

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INTRODUCTION

Murrah, considered as one of the best buffalo milch breed of world, has its breeding tract in Haryana, India. Murrah buffaloes are known for their remarkable power of endurance for hot climate of tropics, resistance to tropical diseases, low cost of maintenance and having higher percentage of milk constituents as compared to other buffalo breeds. Due to these attributes, Murrah buffaloes have been exported to many countries globally from India for replacing or upgrading their local buffalo population. Technological advances in artificial insemination (AI) have facilitated tremendous genetic progress in productivity of buffaloes at a faster pace. However, quality semen production in buffaloes is beset with many constraints viz., poor libido, low sperm count, deterioration in libido and semen quality in inclement weather conditions. Recent study (Khate, 2005) revealed that a number of high genetic merit Murrah bulls are culled due to diverse reasons viz., poor libido (20.45%), poor semen quality (3.40%) and poor semen freezability. Though a few studies have been conducted on sexual behaviour and semen production performance of Murrah buffaloes, but effect of the management interventions especially type of semen collection floor on augmenting libido, semen quality and quantity has not been studied. The present study was, therefore, planned with the specific objective of studying the effect of semen collection floor on sexual behaviour and semen quality of Murrah buffalo bulls.

MATERIALS AND METHODS

The present investigation was conducted on Murrah buffalo breeding bulls maintained at NDRI, Karnal. A total of 12 bulls (more than two years of age) were used in the experiment. All the bulls were housed individually under loose housing system. The bulls were fed ad libitum. All bulls were tied in the washing shed and were washed thoroughly half an hour before semen collection. The semen was collected at 8.30 - 9.30 AM during winter and from 7.30 - 8.30 AM during

Treatment	No. of animals	Period of experiment
Rough cemented floor	12	First 2 weeks
Rubber mattress	12	Next two weeks
Burnt brick clay floor	12	further two weeks

summer. All the bulls selected were randomly divided into two groups for the experiment. This experiment was conducted by switch over design for the three different types of floor treatments as shown below:

Observations were recorded on sexual behavioural and seminal characteristics. The sexual behaviour was recorded at the time of semen collection /training and various sexual behavioural expressions using different score cards. Observations were recorded on temperament using temperament score (0-5). Libido score (1-9) card developed by Chenoweth et al. (1977) was used for recording libido score. Erection and protrusion scores during the experiment were observed and recorded as described by Joshi and Kharche (1992). The release of thrust (intensity of thrust score) was observed and recorded as described by Joshi and Kharche (1992). Reaction time, dismounting time and total time taken in mounts was also recorded. Reaction time is the time lapse between the appearance of bull to the dummy bull and its first mount or mounting attempt. Dismounting time is the time lapse between immediately after release of thrust (ejaculated) and stepping down of the front legs on the ground. Total time taken in mounts is the duration of time taken by a non-stimulus male from the appearance to a mount with successful ejaculation when it was brought to a stimulus bull. All these times were recorded by using stop watch. The frequency of Flehman's response was also recorded. Flehman's response is a typical male behaviour observed when a bull is brought in contact with another herd mate.

Semen was collected from each bull once a week with two consecutive ejaculates by using artificial vagina technique, using Danish model artificial vagina (AV). The temperature of AV was maintained at 45-47 °C with sufficient pressure and lubrication. The semen was kept in water bath at 35-37 °C and evaluated for various physical attributes and semen quality parameters within one hour after collection. Immediately after collection the ejaculates were assessed for the ejaculate volume (ml), mass activity (0-5), initial progressive motility percent, percent live/dead and abnormal and total sperm account. Two ejaculates from the same bull, which were normal and having comparative consistency and mass activity, were pooled for subsequent use in experiment. Semen was extended in egg yolk tris glycerol diluents and cryo-preserved in liquid nitrogen. Semen cryo-preserved in French mini straws (0.25 ml capacity) were kept in storage containers. Frozen semen was assessed for post thaw motility percent. Data were analysed by least squares technique.

RESULTS AND DISCUSSION

Data were analyzed for assessing the effect of different types of semen collection floor on sexual behaviour and seminal attributes of breeding bulls. Reaction time (RT, seconds), dismounting time (DMT, Seconds), total time taken in mounts (TTTM, Seconds), and Flehmen's response (FR): Least square means and analysis of variance are presented in Tables 1 and 2, respectively.

The overall mean RT was 39.66±2.35 seconds. The bulls were more active and had higher sexual stimulation before semen collection on rubber mattress followed by brick clay floor and least on

Table 1: Least square means of sexual behaviour characters of bulls for different types of semen collection floor

Type of collection floor	Reaction time (s)	Dismounting time (s)	Total time taken in a mount (sec)	Flehmen's response
Concrete	40.0±1.89[a]	7.32±0.28[a]	47.32±1.8[a]	7.18±1.33
Brick clay	36.24±1.78[b]	8.24±0.306[b]	44.48±1.83[b]	6.32±1.89
Rubber mattresses	42.75±3.80[a]	7.79±0.18[b]	50.54±3.83[a]	6.54±0.06
Overall mean	39.66±2.35	7.78±0.27	47.38±2.63	--

Table 2: Analysis of variance (MS) of sexual behaviour characters of bulls expose to different types of semen collection floor

Source	df	Reaction time	Dismounting time	Total time take in amount	Flehmen's response
Between bulls	5	**1676.78	3.68	**1755.38	*0.12
Between floors	2	456.26	1.76	560.72	0.01
SE		364.79	2.64	357.27	0.05

*Significant (p<0.05); **Significant (p<0.01)

Table 3. Least square means for sexual behaviour characters of bulls for different types of semen collection floor

Collection floor type	Erection score (0-4 scale)	Protrusion score (0-4 scale)	Intensity of thrust (0-4 scale)	Temperament score (0-5 scale)	Libido score (0-9 scale)
Concrete	3.16±0.187 ^{a}	2.53±0.164 ^{a}	2.43 ±0.08 ^{a}	0.38±0.05	7.15±0.20 ^{a}
Brick clay	3.41±0.175 ^{b}	2.91±0.19 ^{b}	2.80±0.19 ^{b}	0.41±0.14	7.25±0.133 ^{b}
Rubber mattress	3.03±0.10 ^{b}	2.43±0.10 ^{b}	2.03 ±0.10 ^{b}	0.37±0.10 ^{a}	7.09±0.10 ^{a}
Overall mean	3.26±0.165	2.62±0.152	2.49±0.08	0.38±0.16	7.17±0.148

concrete floor. The variation between bulls was highly significant (p<0.01). Similar values were reported by Tomar and Singh (1998) in Murrah bulls. Lower reaction time was found by Kumar (1995) and by Mandal and Tyagi (2004).

Perusal of tables 1 and 2 revealed that there was no significant effect of type of semen collection floor on DMT. The overall mean of DMT for different types of semen collection floor was 7.78±0.27s, which indicated that bulls quickly dismounted after ejaculation. Estimates lower than present values were reported in Sahiwal bulls (Kumar 1993; Rao et al., 1996; Mandal and Tyagi 2004). Slightly lower DMT values were also found in Ongole, Jersey x Ongole bulls and Jersey as reported by Rao et al. (1996) and in Surti buffalo bulls, by Purohit et al. (2000)

The overall mean for TTTM was 55.68±2.23 seconds which indicated that all the bulls took less time in ejaculation. TTTM was highest for concrete followed by brick clay and least for rubber mattress type of floors, so it could be preferable than others. However, analysis of variance revealed that the differences were not statistically significant between different types of collection floor. Similar values for TTMM were reported in Murrah and Sahiwal bulls (Kumar, 1995) and in Sahiwal bulls (Mandal and Tyagi, 2004). Higher values, on the other hand, were recorded in Tharparkar bulls (Mishra et al. 1972) and in crossbred bulls (Pathkar et al. 1990).

It was observed that that majority of bulls didn't show FR at the time of semen collection and no variation between different types of floor. The overall mean of FR for different types of collection floors was 6.65±0.826. However significant variation in FR between bulls was observed (P< 0.05), this might be due to the individual variation in FR. Similar finding was reported by Salvador et al. (2003). Higher FR values were obtained by Panwar (1989), Kumar (1993), Panwar and Nagpaul (1994) and Kumar (1995) in Sahiwal and Murrah bulls.

Erection score (ES), Protrusion score (PS) and Intensity of thrust (ITS): Least square means and analysis of variance are depicted in Tables 3 and 4, respectively. ES was found significantly (p<0.01) influenced by the type of collection floor. Erection was obtained between fair to good with an overall mean as 2.69±0.047. Concrete floor showed significantly higher values of ES (2.92 ±0.06) than Rubber mattresses (2.63±0.10) and Brick

Table 4. Analysis of variance (MS) for sexual behaviour characters of bulls for different types of semen collection floor

Source	DF	Erection score	Protrusion Score	Intensity of thrust	Temperament Score	Libido score
Within bulls	5	0.82	0.52	0.72	0.28	4.15
Between floors	2	0.73	0.73	0.83	1.26	2.85
SE		0.16	0.16	0.16	0.22	0.49

* Significant ($p<0.05$); * * Significant ($p<0.01$) , SE= Standard error

clay (2.54±0.10), which might be due to adaptation of bulls to the concrete floor. The findings obtained in the present study were similar to the observations in Sahiwal and Murrah bulls (Kumar 1993). However, slightly higher score for erection in Sahiwal bulls was reported by Joshi and Kharche (1993) and lower value by Mandal and Tyagi (2004).

PS was significantly ($p<0.01$) influenced by the type of collection floor. The PS varied between fair to good among bulls on a 0-4 scale with an overall mean of 2.69±0.05 on different floors. Concrete floor showed significantly higher values of PS than rubber mattresses and brick clay, which might be due to adaptation of bulls to the concrete floor and preference of animals to mount on rubber mattresses than brick clay floor. The findings of this study are in agreement to the observation reported by Mandal and Tyagi (2004) in Sahiwal bulls. Lower values in Sahiwal cattle and Murrah buffalo bulls were reported by Kumar (1993). Higher erection score (3.950 ± 0.28) was reported by Joshi and Kharche (1992) in cross breed cattle.

The Intensity of thrust was significantly ($p<0.01$) influenced by the type of collection floor. The overall mean of erection score was 2.69±0.05, which was in the range of fair to good on different types of collection floors. Concrete floor showed significantly higher values of intensity of thrust than rubber mattresses and brick clay, which might be due to adaptation of animal to the concrete floor and preference of animals to mount on rubber mattresses than brick clay floor. The present score was higher than the scores reported by Kumar (1993) in Sahiwal cattle and Murrah buffalo bulls and by Mandal and Tyagi (2004) in Sahiwal cattle bulls. However, higher scores were reported by Mathur and Vyas (1969) in Nagori cattle bulls and by Joshi and Kharche (1992) in crossbreed cattle bulls.

The effect of semen collection floor type on TS was highly significant ($p<0.01$). Rubber mattress showed significantly higher value of TS than concrete and brick clay floor. The overall mean was 0.40±0.06, which ranged from docile to very aggressive during semen collection. The results indicated that bulls preferred rubber mattress than other floor types, which might be due to better comfort during semen collection. Different values of behavioural temperament were observed in Murrah bulls by Panwar (1989) and Kumar (1993).

Libido score was found to be significantly ($p<0.01$) influenced by type of collection floor. Rubber mattress revealed significantly higher values than brick clay and concrete floors indicating that rubber mattress was the choice of floor followed by brick clay. The variation between bulls was significant ($p<0.05$). The overall means of LS (0-9 scale) of bulls on different types of collection floor was 7.07±0.08, which indicated that all bulls were active to very active at collection time and exhibited pronounced libido as evidenced from their libido ratings. The results of the present study were in agreement with the values reported in literature for Sahiwal cattle and Murrah buffalo bulls (Panwar, 1989; Kumar, 1993). Kumar (1993) also found significant ($p<0.05$) variation between the bulls.

Seminal attributes: Ejaculate Volume (ml), Physical appearance, Mass activity, Initial progressive motility (IPM) (%), Non eosinophilic spermatozoa count (NESC)(%), Hypo Osmotic Swelling Reactivity Test (HOST) and Post thaw motility (PTM) (%): Least square means, analysis of variance and graph of volume are presented in Tables 5 and 6, respectively. Ejaculate volume showed highly significant ($p<0.01$) variation

Table 5. Least square means for semen quality parameters of bulls for different semen collection floors

Types of collection floor	Ejaculate Volume(ml)	Semen colour	Mass activity (0-4) grade	Initial progressive motility	Non eosinophilic Sperm (%)	Post Thaw Motility (%)	HOST (%)
Concrete	3.40±0.16b	2.92±0.17	3.00±0.07	66.67±7.02	82.08±8.59	47.18±4.92	71.3±9.3
Brick clay	3.79±0.22b	2.92±0.10	3.02±0.09	64.16±6.74	78.75±8.22	49.16±5.17	73.7±7.7
Rubber mattresses	4.25±0.31a	2.92±0.12	2.18±0.09	59.16±6.23	78.75±8.32	47.18±4.92	67.6±5.6
Overall mean	3.81±0.11	2.92±0.08	2.94±0.048	63.33±4.00	80.14±5.02	47.78±3.00	75.8±4.3

Table 6. Analysis of variance (MS) of sexual behaviour characteristics of bulls for different types of semen collection floor

Source	DF	Ejaculate Volume	Semen colour	Mass activity	Initial progressive motility	Non eosinophilic Sperm (%)	Post Thaw Motility	HOST (%)
Between bulls	5	7.66	0.60	0.21	35.00	9.51	3.06	41.6
Between floors	2	4.39	0.00	0.34	87.50	18.06	8.68	92.5
SE		0.89	0.41	0.17	1149.22	1813.92	648.10	12.5

* Significant ($p<0.05$); ** Significant ($p<0.01$)

between bulls exposed to different types of collection floors. The overall mean of volume (ml) for different types of floors and between bulls was 3.81±0.11. Rubber mattress showed significantly highest semen volume (4.25±0.31) which confirmed its superiority in semen collection than other two types of semen collection floors. The mean semen volume was similar to values in Murrah buffalo bulls varying from 3.36±0.14 to 4.57±0.32 with the range of 3-7 ml reported by various workers (Tomar, 1964; Bhosrekar and Nagpaul, 1972; Kumar, 1993; and Keshava, 1996). Slightly higher values were reported by Gupta et al. (1990) and lower values reported by Panwar and Nagpaul (1994).

No significant effect was found on semen colour due to collection floor type. Slight differences were observed between bulls which were not significant. The colour of semen ranged from milky to creamy. The semen colour observed was similar as reported by Panwar (1989) and Kumar (1993).

Similarly, non significant effect of semen collection floor was found on mass activity. The overall mass activity was 2.94±0.048, which was in close agreement with the values (2.80 ± 0.06) reported by Singh and Pangawkar (1990) and Keshava (1996) (2.70). Slightly higher mass activity (3.12 ± 0.20) was reported by Bhosrekar and Nagpaul (1972) and (3.57) Gupta et al. (1990). Lower value (1.6 – 2.8) was reported by Panwar (1989) and (2.18 ± 0.266) Kumar (1993) in Sahiwal bulls.

No significant effect of semen collection floor was found on IPM. The overall mean of IPM was 63.33±4.00. Similarly, non-significant effect of collection floor types on NESC and between bulls was observed. The overall mean of NESC was 80.14±5.02, which assured good quality of semen that might be due to breed inheritance, nutrition and management factors. The values of present study were similar to those reported by Rao et al. (1996) in Ongole cattle bulls.

Collection floor type did not significantly influenced HOST. The overall means of HOST was 75.8±4.3%, which indicated acceptable quality of semen for freezing and artificial insemination. The estimated semen HOST values of present study were in close agreement with the values reported by Chauhan et al. , 1992. Identical results were obtained for PTM. The overall means of PTM was 47.78±3.00, which indicated acceptable quality of semen for freezing and artificial insemination.

Conclusions: The results of this study indicated that bulls were very active and had higher sexual stimulation before semen collection on rubber mattress followed by brick clay floor and concrete collection floor as reflected by reaction time, dismounting time, total time taken in mounts and other sexual behavioural manifestations. Erection score, protrusion score, intensity of thrust, temperament score and libido score were highly influenced by collection floor, being superior on rubber mattress. Rubber mattress showed significantly highest value of semen volume which confirmed its superiority in semen collection than other types of semen collection floor. An overview of the results indicated that the rubber mattress collection flooring helps in improving the semen output without having any detrimental effect on semen quality.

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