

Research Article

Influence of Semen Collector on Semen Characteristics of Murrah Buffalo and Crossbred Bulls

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Abstract | A total of 156 ejaculates each from 8 Murrah buffalo and 8 Crossbred (Karan Fries) bulls were collected during one year period at Artificial Breeding Research Centre, NDRI, Karnal, India and analysed using least squares analysis to quantify the effect of collector on various semen quality parameters. The overall least squares mean values of Murrah buffalo and Crossbred bulls for ejaculate volume (ml), mass activity, Initial motility (%), sperm concentration ($10^6/\text{ml}$), total sperm output (10^6), non-eosinophilic count (%), HOST (%), acrosome integrity (%), head abnormality (%), mid-piece abnormality (%), tail abnormality (%), total abnormality (%), pH and osmolality (mOsmol/kg) were 2.66 ± 0.10 & 4.45 ± 0.10 ; 2.54 ± 0.70 & 1.79 ± 0.78 ; 60.64 ± 0.02 & 46.73 ± 0.03 ; 1016.68 ± 21.25 & 839.25 ± 27.38 ; 2748.67 ± 122.86 & 3689.55 ± 146.17 ; 67.20 ± 0.03 & 53.38 ± 0.04 ; 52.72 ± 0.01 & 40.88 ± 0.03 ; 70.10 ± 0.02 & 55.73 ± 0.04 ; 2.30 ± 0.001 & 3.16 ± 0.02 ; 1.62 ± 0.01 & 2.23 ± 0.007 ; 5.50 ± 0.002 & 7.53 ± 0.001 ; 9.47 ± 0.002 & 12.97 ± 0.001 ; 6.78 ± 0.20 & 6.91 ± 0.19 and 277.78 ± 2.40 & 287.30 ± 2.34 , respectively. Collector was found to have significant ($p < 0.01$) influence on all semen quality traits of Murrah buffalo bulls, but there were no significant differences ($p < 0.05$) between collector 1 and collector 2. In case of Karan Fries bulls, the collector had significant ($P < 0.05$) effect on ejaculate volume (4.84 vs. 4.06 ml) and total sperm output (4032.19 vs. 3346.19 10^6), but did not have significant ($P < 0.05$) effect on other semen quality parameters. It was concluded that semen collection is highly skilled and scientific job and need efficient collector to achieve collection of optimum quantity and quality of semen, which is the major aim of semen stations.

Keywords | Collector, Motility, Semen quality, Karan Fries and Murrah buffalo bull

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INTRODUCTION

Presently India is producing about 66 million straws, which is covering only 20-25% breedable bovine population. To achieve the national target of covering 50% breedable bovine population there is need to produce 140 million of frozen semen straws. Therefore, semen stations throughout the India are

giving emphasis on production of required number of frozen semen doses, but there are various factors which influence the quality and required quantity of semen. The central monitoring unit of government of India is also periodically evaluating the performance for producing good quality germplasm. To achieve the quality criteria we need to intervene at the time of sexual preparation of bulls by bull handlers and semen

collector, which is one of the important aspects for quality semen production. Researchers have depicted significant effect of collection team on semen quality (Waltl et al., 2006). Sexual preparation, sexual stimulation and appropriate technique of semen collection are the contributing factors influencing the semen quality. The amount of motile sperm collected may be influenced by the bull handler's skill and behaviour (Younis, 1996) since both the motility (Foster et al., 1970) and number of sperm obtained (Almquist, 1973) are enhanced, only if the sexual stimulation is appropriate. In general semen collection is practiced in the early morning to avoid any disturbance in exhibiting proper sexual behaviour and vigour of the breeding bulls due to presence of only the persons involved in semen collection, absence of unwanted people, phone calls and gossip. Attendants and nearby structures may also influence the sexual behaviour and semen quality (Kerruish, 1955). Motivation of collector and handler for their work is necessary to optimize semen production and greater incentives need to be given to workers to improve their techniques (Amann and Almquist, 1976; Larson et al., 1982). Therefore, the experiment was designed to study the effect of collector on semen quality parameters of Murrah buffalo and crossbred bulls.

MATERIALS AND METHODS

A total of 156 ejaculates each of 8 Murrah buffalo bulls (nearly 30 to 58 months of age and body weight of 518.58-782.50 kg) and 8 Karan Fries (Crossbred) bulls (nearly 24 to 37 months of age and body weight of 423.17 to 570.33 kg) maintained at Artificial Breeding Research Centre, NDRI, Karnal, India were evaluated for one year duration. The bulls were maintained under uniform managerial conditions and as per bull feeding standard at NDRI, Karnal. The bulls were sexually mature with good libido, healthy, clinically normal and free from diseases. All the bulls were vaccinated against FMD twice a year and HS & BQ annually. There were three collectors assigned to collect semen from Murrah buffalo bulls and two collectors to collect semen from crossbred bulls during the experimental period randomly. Collector 1 and Collector 2 are same for semen collection for both the breeds except Collector 3. Collector 1 and Collector 2 have more than fifteen years of experience and taken scientific training from different experts of andrology from time to time. However, Collector 3 started the

semen collection after 45 years of age whereas, other two collectors learned the semen collection in their twenties. Collector 3 has only 4-5 years of experience in semen collection. Semen was collected in the morning once a week two ejaculate within 30 minutes from the bulls using sterilized bovine artificial vagina (IMV model-005417) maintained at (42-45°C), over a male dummy bull. Soon after collection, each ejaculate was placed in a water bath at 30-32°C and standard laboratory evaluation for semen was recorded. Quality of semen was assessed for volume and microscopic tests such as mass activity, individual motility, concentration, non-eosinophilic count, HOST, acrosomal integrity, sperm abnormalities using DIC phase contrast microscope (Nikon Eclipse E600, Tokyo, Japan) with Tokoiheat thermal stage as per standard method. The pH of fresh semen was determined immediately after collection with Cyberscan 510 pH meter (Eutech Instrument, Singapore) and osmolality by WESCOR vapour pressure Osmometer (WESCOR model 5500, INC, USA). The data were subjected to statistical analysis using LSML-91 software package, Walter Harvey (Snedecor and Cochran, 1994).

RESULTS AND DISCUSSION

Efficient semen collection from breeding bulls is an art and an important activity of frozen semen production centre. The management of entire process from bull training to semen collection is critical, for which semen collector and the bull handler play a vital role. All the collectors were well trained and were collecting semen from the bull for last minimum of three to five years regularly. Least squares means values with significant levels for the effect of collector on various seminal attributes of Murrah buffalo and crossbred bulls are presented in Table 1 and 2.

SEMINAL ATTRIBUTES OF MURRAH BUFFALO AND CROSSBRED BULL

The overall least squares mean values of Murrah buffalo for ejaculate volume (ml), mass activity, Initial motility (%), sperm concentration (10^6 /ml), total sperm output (10^6), non-eosinophilic count (%), HOST (%), acrosome integrity (%), head abnormality (%), mid-piece abnormality (%), tail abnormality (%), total abnormality (%), pH and osmolality (mOsmol/kg) were 2.66 ± 0.10 ; 2.54 ± 0.70 ; 60.64 ± 0.02 ; 1016.68 ± 21.25 ; 2748.67 ± 122.86 ; 67.20 ± 0.03 ; 52.72 ± 0.01 ; 70.10 ± 0.02 ; 2.30 ± 0.001 ; 1.62 ± 0.01 ; 5.50 ± 0.002 ;

Table 1: Least squares means along with standard error for the effect of collector on semen quality parameters of Murrah buffalo bulls

Parameters	Collector 1 (N=32)		Collector 2 (N=77)		Collector 3 (N=47)		Overall (N=156)	
	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.
Ejaculate volume (ml)	2.97 ^A	0.21	2.86 ^A	0.14	2.14 ^B	0.18	2.66	0.10
Mass activity (0-5 Scale)	2.76 ^a	0.14	2.59 ^a	0.09	2.28 ^b	0.12	2.54	0.70
Initial motility (%)	65.40 ^a	0.08	61.93 ^a	0.04	54.43 ^b	0.06	60.64	0.02
Sperm concentration (10 ⁶ /ml)	1089.46 ^a	43.17	1041.46 ^a	28.69	919.11 ^b	36.96	1016.68	21.25
Sperm concentration per ejaculate (10 ⁶)	3176.92 ^A	249.58	2991.31 ^A	165.90	2077.79 ^B	213.70	2748.67	122.86
Non-eosinophilic count (%)	72.50 ^A	0.11	69.04 ^A	0.05	59.76 ^B	0.08	67.20	0.03
HOST (%)	56.99 ^A	0.06	54.13 ^A	0.03	47.03 ^B	0.05	52.72	0.01
Acrosome integrity (%)	75.07 ^A	0.10	71.64 ^A	0.04	63.25 ^B	0.07	70.10	0.02
Head (%)	2.14 ^a	0.001	2.23 ^a	0.001	2.53 ^b	0.001	2.30	0.001
Mid piece (%)	1.54 ^A	0.001	1.58 ^A	0.001	1.74 ^B	0.001	1.62	0.01
Tail (%)	4.95 ^A	0.006	5.23 ^A	0.003	6.37 ^B	0.005	5.50	0.002
Total (%)	8.68 ^A	0.008	9.10 ^A	0.004	10.68 ^B	0.006	9.47	0.002
pH	6.70 ^A	0.04	6.75 ^A	0.03	6.88 ^B	0.04	6.78	0.20
Osmolality	270.02 ^A	4.87	275.08 ^A	3.23	288.24 ^B	4.17	277.78	2.40

Least squares means bearing different alphabets as superscripts differ significantly (^aP<0.05, ^{AB}P<0.01)

Table 2: Least squares means along with standard error for effect of collector on semen quality parameters of Karan Fries bulls

Parameters	Collector 1 (N=95)		Collector 2 (N=61)		Overall (N=156)	
	Mean	S.E.	Mean	S.E.	Mean	S.E.
Ejaculate volume (ml)	4.84	0.13	4.06	0.15	4.45	0.10
Mass activity (0-5 Scale)	1.82	0.10	1.76	0.12	1.79	0.78
Initial motility (%)	47.43	0.05	46.04	0.08	46.73	0.03
Sperm concentration (10 ⁶ /ml)	840.42	34.79	838.08	42.46	839.25	27.38
sperm concentration per ejaculate (10 ⁶)	4032.19	185.73	3346.91	226.69	3689.55	146.17
Non-eosinophilic count (%)	53.92	0.07	52.86	0.10	53.38	0.04
HOST (%)	41.60	0.04	40.18	0.06	40.88	0.03
Acrosome integrity (%)	56.46	0.06	55.02	0.09	55.73	0.04
Head (%)	3.17	0.001	3.14	0.001	3.16	0.02
Mid piece (%)	2.23	0.001	2.23	0.001	2.23	0.007
Tail (%)	7.56	0.001	7.49	0.002	7.53	0.001
Total (%)	13.02	0.002	12.92	0.003	12.97	0.001
pH	6.9	0.02	6.91	0.03	6.91	0.19
Osmolality	286.50	2.97	288.11	3.63	287.30	2.34

9.47±0.002; 6.78±0.20 and 277.78±2.40; respectively. Similar findings were reported in case of ejaculate volume, mass activity (Ghosh, 2004); initial motility (Kumar, 1993); sperm concentration (Shukla and Mishra, 2005); HOST (Jadhav, 1998) in Murrah buffalo bulls.

The overall least squares mean values of Crossbred bulls for ejaculate volume (ml), mass activity, Initial motility (%), sperm concentration (10⁶/ml), total sperm output (10⁶), non-eosinophilic count (%), HOST (%), acrosome integrity (%), head abnormality (%), mid-piece abnormality (%), tail abnormality (%), total abnormality (%), pH and osmolality (mOsmol/kg) were 4.45±0.10; 1.79 ± 0.78; 46.73 ± 0.03; 839.25± 27.38; 3689.55± 146.17; 53.38 ±0.04; 40.88±0.03; 55.73± 0.04; 3.16 ±0.02; 2.23 ±0.007; 7.53 ±0.001; 12.97±0.001; 6.91±0.19 and 287.30±2.34, respectively. The results are in consonance with the reports in case of ejaculate volume (Sethi et al., 1989; Jain, 2004); mass activity (Jain, 2004); tail abnormalities (Dede et al., 1983) whereas higher values of non-eosinophilic count (Ulfin and Raina, 2002; Fiaz et al., 2010) was reported in dairy bulls.

EFFECT OF COLLECTOR ON SEMEN QUALITY OF MURRAH BUFFALO AND CROSSBRED BULL

Collector was found to have highly significant ($p < 0.01$) influence on all semen quality traits such as ejaculate volume, mass activity, initial motility, sperm concentration, total sperm output, non-eosinophilic count, HOST, acrosome integrity, sperm abnormalities, pH and osmolality. Results of the present study showed that there was no significant difference ($p < 0.05$) in the values of the semen quality parameters between collector 1 and collector 2. The performance of collector 1 and collector 2 was found to be significantly ($p < 0.05$) superior to that of collector 3 in terms of all semen quality parameters studied in this experiment.

The collector had significant ($p < 0.05$) effect on ejaculate volume and total sperm output in KF bulls. Collector 1 appeared to be more efficient in collecting the semen but, collectors did not have a significant ($p < 0.05$) effect on MA, IM, SPC, SPCE, LIVE, HOST, AI, sperm abnormalities, pH and osmolality of ejaculates.

The findings of the present study are in consonance

with the finding of Walzl et al. (2006), who reported that collection team had significantly ($p < 0.05$) influenced ejaculate volume, sperm concentration, percentage of viable spermatozoa in the ejaculate, total spermatozoa per ejaculate; whereas Mathevon et al. (1998) and Yates et al. (2003) showed significant effect of collection team on ejaculate volume and total number of cells produced. On contrary Mathevon et al. (1998) reported that collection team did not have a significant effect on concentration and motility of the sperm. This may explain why bull handler and semen collector have a significant impact on semen quality and quantity as they are responsible for sexual stimulation and preparation of bull for giving a good ejaculate as it was evident from the finding of Almquist (1978), who also reported that sperm output depends on sexual preparation of the bull by the handler. Komisrud and Berg (1996) emphasized the importance of duration of sexual preparation to improve ejaculate volume, number of doses per ejaculate and post-thaw motility. The amount of motile sperm collected may be influenced by the bull handler's skill and behaviour (Younis, 1996; Walzl et al., 2006). Further, the motility (Foster et al., 1970) and the number of sperm obtained (Almquist, 1973) are enhanced only if the sexual stimulation is appropriate. The proper technique for sexual stimulation depends largely on the sexual behaviour of the bull at the time of semen collection. Dominguez et al. (1994) also reported that the technique used for bull preparation significantly affected the volume of the ejaculate. Buffalo bulls possess some of the peculiar behavioural characteristics, which need to be given due importance by the semen collector on individual basis to optimize the semen production performance. Buffalo bulls are very sluggish in their libido score and takes more time for mount and sometimes a second bull is required to be approached for semen collection behind the bull also stimulates the bull to mount and ejaculate. Another peculiarity, most of the Murrah buffalo bulls are stimulated when the teaser bulls were moved around the collection floor and semen was collected in the collection yard while the teaser bulls were moving. Very few Murrah buffalo bulls prefer to donate semen in stationary teaser (cattle or buffalo) in the serving crate. Therefore, there is need to give emphasis on proper skill development of bull handler and semen collectors as well as they need to be motivated for their work through incentives to optimize semen production (Amann and Almquist, 1976; Larson et

From the present study it was evident that the effect of collector was quite pronounced on various semen quality parameters in Murrah buffalo. This may be due to collector 3, as he collected consistently very poor grades ejaculates from Murrah buffalo bulls. Collector 3 was not suitable for semen collection due to lack of swiftness in approaching the bull for semen collection. Another reason may be more sensitiveness of buffalo bulls in sexual preparation. There is need of more skilled person with knowledge of specific needs of the individual bull during proper sexual stimulation for semen collection, as libido is less prominent and intense in case of Murrah bulls. Most of the bulls are approached for collection by giving stimulus (whistling) to the bull for mounting. In case of buffalo bull's semen collection in moving teaser is a common practice for successful collection of semen, which is emphasizing the need of very active semen collector to provide optimum comfort to buffalo bulls, but on contrary Brockett *et al.* (1994) reported that moving the teaser bull around the collection floor did not stimulate all bulls. The quality of semen collected by Collector 1 was superior as compared to Collector 2 and Collector 3, but there was no significant difference in quality of semen collected by Collector 1 and Collector 2 in both the breeds. It is portraying that Collector 1 and Collector 2 are technically sound and skilled semen collector; therefore their performance was found to be similar in both the breeds. Therefore, it is depicting that the systematic training of the collector from experts is important for learning all the techniques of semen collection in more scientific way to harvest better quality semen. The collector should start the learning from young age with good physical strength and swiftness. Experience of collector along with judging the individual sexual behaviour of bulls will enhance the scope of future improvement in semen production.

CONCLUSION

Collection team significantly affect the performance of bulls, it might be useful for the industry to study in detail the techniques used by different handlers and collectors to maximize production. An improvement in collection techniques will benefit the bull's welfare, although the economic impact will be greater for mature bulls because improved collection techniques will

increase semen production from high genetic merit bulls that are in great demand. Therefore, the collector should be selected on the basis of his ability, enthusiasm and experience to work with bulls.

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CONFLICT OF INTEREST

The authors have no conflict of Interest.

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