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Effect of Genetic and Non-genetic Factors on Semen Quality Traits in Bovine Breeding Bulls

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ABSTRACT

Background: A breeding bull is used to serve many females; therefore, for overall herd fertility, the bull's fertility is more important than female fertility.

Methods: The present study was carried out to compare the effects of species, season, period, and interaction between species and season on the semen quality traits of Murrah buffalo and crossbred cattle bulls. For the 25-year study period, a total of 26205 ejaculation records of 389 bovine breeding bulls, including Murrah buffalo and crossbred cattle bulls, were analysed that were maintained at an organised farm.

Result: Species and period of semen collection had a highly significant effect on all the traits (P<0.01). Season of semen collection and interaction between season and species had a highly significant effect (P<0.01) on semen volume (SV), consistency, mass activity (MA), total sperm count (TSC), initial motility (IM) and post-thaw motility (PTM), a significant effect (P<0.05) on age at first semen collection (AFSC) and non-significant effect on the semen colour and age at first semen freezing (AFSF). Overall, semen quality was good at the organised farm during all seasons. In bovine breeding bulls, PTM during the rainy and summer seasons was comparatively low. An overall increase in semen quality in the later periods showed progressive improvement in management at the organised farm over the period.

Key words: Bovine, Period, Season, Semen, Species.

INTRODUCTION

Livestock is an essential part of agriculture in India. Through the 'green revolution', India was able to produce enough food grains on its own, while the 'white revolution' produced similarly impressive advancements in milk production. With a total of 302.79 million bovines, including 192.49 million cattle and 109.85 million buffaloes, India is the country with the biggest bovine population (Livestock Census, 2019). In terms of total milk production, India ranks first in the world (BAHS, 2022), which could only be possible due to the availability of high genetic merit cattle and buffaloes.

The best buffalo breeds in the world are found in India, like Murrah (*Bubalus bubalis*), which is used as an improver breed and the breed average is about 2200 kg (2000-2500 kg) per lactation (Taraphder, 2002). Elite animals of this breed have demonstrated lactation averages of up to 4,000 kg or more. Germplasm and bulls of Murrah are extensively used for upgrading native buffalo stock in many nations whose buffalo breed needs improvement (Shivahre *et al*, 2017). Exotic crossbred cattle make up 16.93% (51.36 million) of the total bovine population and 26.54% of the total cattle population in India, while they produce around 31.83% of total milk production and buffaloes produce 45.07% of total milk production (BAHS, 2022). For exotic/ crossbred cattle, the average yield per animal per day is 8.39 kg/day during 2020-21 and for indigenous/non-descript it is 3.20 kg/day. That means India contains exotic/ crossbred cattle of high genetic merit.

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The male factor is very important since a single bull is used to serve numerous females; therefore, the bull's breeding capacity is more important to the productivity of the herd than issues with female infertility. To evaluate the breeding soundness of bovine breeding bulls, seminal profile and high fertilizing ability are crucial parameters. The basic parameters for determining the quality of semen include sperm volume, sperm concentration, sperm motility, mass activity, sperm morphology and seminal pH (Tohura, 2018).

The season is one of the key factors that affect the reproductive performance of dairy animals by influencing both macro- and microclimatic factors like temperature, humidity, rainfall, and photoperiod (Mandal *et al*, 2000). Adverse climatic and disease conditions may result in the disposal of those buffalo and crossbred bulls that show poor semen quality (those who consistently produce less than 60% sperm initial motility), poor freezability (those who consistently produce less than 35% post-thaw motility), and poor libido (Bhakat *et al*, 2009). A better understanding of the effects of the season of semen collection can help the AI industry with proper management and increase semen output. Differences in puberty ages can be related to the environment, management, and breed of the bulls that were reared (Brito *et al*, 2004). Therefore, to gain a better understanding of semen production and quality traits, 25 years of data on bovine breeding bulls were analysed.

MATERIALS AND METHODS

Experimental design and data collection

To study the effects of species, season of semen collection, period of semen collection, and species and season interaction on semen quality traits, information on Murrah buffalo and crossbred cattle (*Bos taurus* × *Bos indicus*) bulls over a period of 25 years (1995-2019) was gathered from records kept at the Directorate of Livestock Farms, Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana. During the study period, a total of 26205 ejaculation records from 389 bulls were included in the study, with 11434 ejaculation records of 207 Murrah buffalo bulls (sired by 84 bulls) and 14771 ejaculation records of 182 crossbred cattle bulls (sired by 102 bulls). The various semen quality traits like semen volume, colour, mass activity (MA), consistency, total sperm count (TSC) (million/ml), initial motility% (IM%) and post-thaw motility% (PTM%), age at first semen collection (AFSC) (days), and age at first semen freezing (AFSF) (days) were recorded during the study. The present experiment was approved by the Institutional Animal Ethics Committee (Protocol No. GADVASU/2022/IAEC/65/10).

Using an artificial vagina, the semen from all the bulls was collected during the morning hours (37°C). The semen was immediately brought to the lab and placed at 37°C in a water bath. Test tubes were used to directly measure the amount of semen in each ejaculate. The cream colour and cream with a tinge of yellow were considered normal for Murrah buffalo and crossbred cattle bulls, respectively (Code given 1). While milky (Code given 2), watery (Code given 3), bloody, pink and other (Code given 4) were considered abnormal. The MA of spermatozoa was recorded immediately after semen collection by putting a drop of semen on a grease-free, clean and dry slide maintained at 30-35°C under a low-power microscope with an attached stage warmer at 37°C. The score was calculated on a 0-5 numerical scale. The TSC was measured using a Neubauer haemocytometer. IM was calculated as a percentage of motile sperm cells by placing a drop of diluted fresh sperm on a dry slide and by covering it with a clean cover slip and examining it under a microscope. The PTM was measured after 24 hours of freezing.

The following traits were generated for statistical analysis:

i. Age at first semen collection (AFSC)

AFSC= Date of first semen collection (DFSC)-
Date of birth (DOB)

ii. Age at first semen freezing (AFSF)

AFSF= Date of first semen freezing (DFSF)-
Date of birth (DOB)

To study semen quality parameters of Murrah buffalo and crossbred cattle bulls reserved for breeding, data were classified into five seasons and five periods. Based on the prevailing climatic conditions in Punjab, the months of the year were grouped into 5 seasons (Table 1).

The data for the span of 25 years were classified into 5 periods, with each period covering 5 years of duration, so that an acceptable number could be achieved (Table 2).

Statistical analysis

The statistical model used for least square analysis to study the effect of species, the season of semen collection, the period of semen collection and the interaction between species and season was as follows:

$$Y_{ijklm} = \mu + SP_i + S_j + P_k + (SP * S)_{ij} + e_{ijkLm}$$

Where,

Y_{ijklm} = Character variable of m^th^ observation of l^th^ bull under i^th^ species, j^th^ season of semen collection and k^th^ period of semen collection.

μ = Overall population mean.

SP_i = Fixed effect of i^th^ species (I= 1, 2).

S_j = Fixed effect of season of semen collection (j= 1, 2...5).

P_k = Fixed effect of k^th^ period of semen collection (k= 1, 2...5).

$(SP * S)_{ij}$ = Effect of interaction between species and season of semen collection.

e_{ijkLm} = Random error for m^th^ observation of l^th^ bull under i^th^ species, j^th^ season of semen collection and k^th^ period of semen collection ~NID (0, σ^2e).

p value less than 0.01 were considered statistically highly significant and p value less than 0.05 were considered statistically significant.

Table 1: Classification of the season of semen collection.

Month	Season
March to Mid-April	Spring
Mid-April to June	Summer
July to September	Rainy
October to November	Prewinter
December to February	Winter

RESULTS AND DISCUSSION

Species

The findings of the present study revealed that species had a highly significant effect (P<0.01) on all the traits. In crossbred cattle bulls, semen volume, colour, MA (mass activity), and PTM% (post thaw motility%) were higher than Murrah buffalo bulls (Table 3). While consistency, TSC (total sperm count), IM% (initial motility%), AFSC (age at first semen collection), and AFSF (age at first semen freezing) were higher in Murrah buffalo bulls (Table 3). In comparison to our study, in Murrah buffalo and crossbred cattle bulls, higher estimations of semen volume (5.5.176 ml and 5.8.176 ml, respectively) and same estimates of IM% were reported by Khatun *et al.* (2013). On the other hand, in Murrah buffalo bulls a lower estimates of semen volume (2.52 ±0.009 ml) and TSC (880.10±22.71 million/ml) and higher estimates of IM% (78.02 ± 0.84%) and PTM (50.59±0.45%) were reported by Rajashri (2019). Our findings on the mean AFSC and AFSF in Murrah buffalo and crossbred cattle bulls were lesser (Table 3) than what was reported for Murrah and crossbred cattle bulls by Khatun *et al* (2013). The mean AFSC and AFSF were lower in cattle bulls since these are crossbred, hence attain maturity earlier than Murrah buffalo bulls.

Effect of season and interaction between species and season on semen quality traits

It was observed that in bovine breeding bulls, the season of semen collection had a highly significant effect (P<0.01) on overall semen volume, consistency, MA, TSC, IM and PTM; a significant effect (P<0.05) on AFSC; and a non-significant effect on colour and AFSF. In bovine breeding bulls, the least square means for semen quality traits are shown in Table 4. To determine the semen quality traits in Murrah buffalo and crossbred cattle bulls during various seasons, the interaction between species and season was assessed in the analysis. It was observed that in bovine breeding bulls, species and season interaction had a highly significant effect (P<0.01) on the semen volume, consistency, MA, TSC, IM%, and PTM%; a significant effect on AFSC and a non-significant effect on colour and AFSF.

In Murrah buffalo bulls, the least square means for semen volume were found to be highest during the summer, rainy and prewinter seasons, followed by the spring and winter seasons (Table 5). Ravimurgan *et al* (2003) observed the highest semen ejaculate volume in Murrah buffalo bulls during the rainy season. In crossbred cattle bulls, semen volume was found to be highest during the spring and winter seasons, followed by the summer, prewinter, and rainy seasons (Table 5). Small increases in scrotal temperature in *Bos taurus* AI bulls can have a significant impact on spermatogenesis (Januskauskas *et al.*, 1995).

In Murrah buffalo and crossbred cattle bulls, the least square means for semen colour during the different seasons are shown in Table 5. In Murrah buffalo bulls during the prewinter and rainy seasons, the consistency of semen was found to be highest, followed by the summer, spring, and winter seasons (Table 5). In crossbred cattle bulls, the consistency of semen was higher during spring, prewinter, winter, and summer and lowest during the rainy season (Table 5).

In our study, the least square means for the MA of semen in Murrah buffalo bulls were highest during prewinter, followed by rainy, spring, summer and winter season (Table 5). In crossbred cattle bulls, the least square means for sperm MA were found to be highest during spring, followed by winter, summer, prewinter and rainy seasons (Table 5). In Murrah buffalo bulls, significantly higher total TSC/ml were observed during the spring, summer and rainy

Table 2: Classification of the period of semen collection.

Duration	Period code
January 1995 - December 1999	1
January 2000 - December 2004	2
January 2005 - December 2009	3
January 2010 - December 2014	4
January 2015 - December 2019	5

Table 3: Least squares mean±S.E. for the semen volume (SV), colour, consistency, MA, TSC, IM%, PTM%, AFSC and AFSF for Murrah buffalo and crossbred cattle bulls.

	Murrah buffalo	Crossbred cattle
SV (ml)	3.344±0.014^{a} (13,968)	5.240±0.016^{b} (11,112)
Colour	1.015±0.005^{a} (13,996)	1.228±0.006^{b} (10,747)
Consistency	2.541±0.005^{a} (13,600)	2.431±0.005^{b} (10,721)
MA	2.402±0.011^{a} (13,092)	2.686±0.013^{b} (10,267)
TSC (million/ ml)	1,325.623±15.612^{a} (3,037)	908.414±16.812^{b} (2,555)
IM (%)	66.404±0.001^{a} (12,550)	64.562±0.001^{b} (9,836)
PTM (%)	42.121±0.001^{a} (6,600)	44.994±0.001^{b} (4,112)
AFSC (days)	802.614±1.333^{a}	655.070±1.464^{b}
AFSF (days)	917.997±1.657^{a}	719.777±1.894^{b}

Data in parentheses indicate the number of observations.

^{a,b}Different superscripts indicate significant differences between two sub-effects (P<0.05).

seasons as compared to the prewinter and winter seasons (Table 5). In Crossbred cattle bulls, TSC/ml during the spring and summer seasons were significantly higher as compared to the rainy, prewinter, and winter seasons (Table 5).

In Murrah buffalo and crossbred cattle bulls, the least square means for IM% during different seasons are shown in Table 5. In Murrah buffalo bulls, IM% was highest during the prewinter season, and in crossbred cattle bulls, it was highest during the spring season. The least square means for PTM in Murrah buffalo and crossbred cattle bulls during different seasons are shown in Table 5 and it was observed that in both Murrah buffalo and crossbred cattle bulls, PTM% was found to be comparatively lower during the summer and rainy seasons when climatic conditions are hot. The least square means for AFSC and AFSF during different seasons are shown in Table 5, which indicates that the season had no effect on AFSC and AFSF in both Murrah buffalo and crossbred cattle bulls. Without affecting the semen's quality or freezeability, early bull training could reduce the average age of the first semen collection (Sethi *et al*, 1989).

Period

A highly significant effect (P<0.01) of the period of semen collection on all traits was found to be observed. It has been revealed that, as compared to the first 3 periods, the per ejaculate semen volume during the last 2 periods was comparatively high (Table 6). Laing *et al*, (1988) suggested that a bull with higher fertility produced more semen than a bull with lower fertility. The period had shown a decreasing pattern in overall semen colour abnormalities except for a slight increase in the third period, which indicates the decreasing trend of semen abnormalities and increasing normal semen colour over periods.

In bovine breeding bulls, consistency and mass activity of semen during different 5 periods of semen collection follow the increasing trend from period 1 to period 5, from 2.397±0.007 to 2.575±0.008 and 1.981±0.016 to 3.117±0.021, respectively (Table 6). For overall TSC during 5 periods of semen collection, the least square means ranged from 966.208±17.020 to 1,271.630±59.956 million/ml, respectively (Table 6). The highest TSC was observed during the first and third periods. Rajashri (2019) reported that a higher semen concentration was recorded in the low-fertility Murrah buffalo bull group (897.44 million/ml) as compared to the high-fertility Murrah buffalo bull group (864.06 million/ ml).

For IM% in bovine breeding bulls, least square means during different periods ranged from 58.934±0.001 to 76.148±0.001% (Table 6). During the last 3 periods, IM% was found to be higher as compared to the first 2 periods, which shows an increasing trend over the periods (Table 6). Khatun *et al.* (2013) also reported an increasing trend in the IM% over periods. During different periods, the least square means for PTM% ranged from 38.561±0.001 to 50.186±0.001%. During the fifth period (50.186±0.001%), significantly highest PTM% was observed as compared

Table 4: Least squares mean ± S.E. for overall semen volume (SV), colour, consistency, MA, TSC, IM%, PTM%, AFSC and AFSF for bovine breeding bulls during different seasons.

Season	SV (ml)	Colour	Consistency	MA	TSC (million/ml)	IM (%)	PTM (%)	AFSC (days)	AFSF (days)
Spring	4.337 ± 0.030^{a} (2,829)	1.131 ± 0.011 (2,749)	2.486 ± 0.010^{b} (2,723)	2.609 ± 0.024^{a} (2,554)	$1{,}158.398\pm27.480^{b}$ (460)	66.486 ± 0.001^{a} (2,547)	44.629 ± 0.001^{b} (1,304)	730.122 ± 2.792^{ab} (2,586)	818.817 ± 3.552 (2,420)
Summer	4.326 ± 0.023^{a} (4,882)	1.116 ± 0.008 (4,815)	2.481 ± 0.008^{b} (4,728)	2.519 ± 0.018^{b} (4,567)	$1{,}215.656\pm19.979^{a}$ (1,164)	64.829 ± 0.001^{c} (4,380)	41.605 ± 0.001^{c} (2,015)	726.336 ± 2.060^{cb} (4,806)	819.136 ± 2.622 (4,491)
Rainy	4.262 ± 0.020^{b} (6,294)	1.113 ± 0.007 (6,276)	2.472 ± 0.007^{b} (6,172)	2.470 ± 0.016^{c} (5,929)	$1{,}109.637\pm17.767^{b}$ (1,186)	63.574 ± 0.001^{d} (5,656)	41.691 ± 0.001^{c} (2,309)	729.259 ± 1.840^{ac} (5,964)	818.013 ± 2.348 (5,528)
Prewinter	4.297 ± 0.024^{ab} (4,177)	1.131 ± 0.009 (4,066)	2.513 ± 0.008^{a} (4,009)	2.641 ± 0.019^{a} (3,920)	$1{,}044.404\pm20.107^{c}$ (1,071)	66.963 ± 0.001^{a} (3,658)	44.196 ± 0.001^{b} (1,793)	725.248 ± 2.288^{b} (3,817)	818.395 ± 2.913 (3,554)
Winter	4.238 ± 0.019^{b} (6,898)	1.116 ± 0.007 (6,837)	2.479 ± 0.007^{b} (6,689)	2.480 ± 0.015^{bc} (6,389)	$1{,}057.615\pm18.098^{c}$ (1,711)	65.560 ± 0.001^{b} (6,145)	45.672 ± 0.001^{a} (3,291)	733.246 ± 1.802^{a} (6,214)	820.072 ± 2.276 (5,858)

Data in parentheses indicate the number of observations.

^{a,b} Different superscripts indicate significant differences between two sub-effects (P<0.05).

to other periods (Table 6), but they didn't show any particular trend.

The least square means for AFSC during different periods ranged from 661.896±2.016 to 777.599±2.425 days, and for AFSF, they ranged from 728.391±2.510 to 892.874±2.600 days (Table 6). Both follow almost the same trend over time. Khatun *et al.* (2013) observed that the least square means of AFSC and AFSF showed a decreasing trend over the periods. Bertram *et al* (2003) reported that scrotal size is closely related to daily sperm production and small scrotal size indicates less sperm production. From around 8 to 10 months of age to 18 to 24 months of age, young bulls go through a testicular development spurt when they should be on a rising plane of nutrition (Bertram *et al*, 2003). Nearly all of the traits in the present context have shown improvement over time. The majority of reproductive traits are significantly influenced by environmental conditions. Bulls that donate more neat semen and have higher mass activity are supposed to produce freezable semen (Sethi *et al.*,1989).

Table 5: Least squares mean±S.E. for the semen volume (SV), colour, consistency, MA, TSC, IM%, PTM%, AFSC and AFSF for Murrah buffalo and Crossbred cattle bulls during different seasons.

		Spring (March-Mid-April)	Summer (Mid-April-June)	Rainy (July-September)	Prewinter (October-November)	Winter (December-February)
SV (ml)	Murrah	3.294±0.039^{ab}	3.415±0.030^{c}	3.414±0.027^{c}	3.380±0.033^{ac}	3.214±0.025^{b}
		(1,599)	(2,812)	(3,460)	(2,257)	(3,840)
	Crossbred	5.379±0.044^{d}	5.237±0.034^{f}	5.109±0.029^{h}	5.213±0.036^{g}	5.263±0.028^{e}
		(1,230)	(2,070)	(2,834)	(1,920)	(3,058)
Colour	Murrah	1.021±0.014	1.023±0.010	0.999±0.009	1.015±0.012	1.016±0.009
		(1,589)	(2,813)	(3,476)	(2,258)	(3,860)
	Crossbred	1.241±0.016	1.209±0.012	1.227±0.010	1.246±0.013	1.216±0.010
		(1,160)	(2,002)	(2,800)	(1,808)	(2,977)
Consistency	Murrah	2.515±0.013^{b}	2.530±0.010^{b}	2.573±0.009^{a}	2.578±0.011^{a}	2.512±0.009^{b}
		(1,539)	(2,725)	(3,400)	(2,174)	(3,762)
	Crossbred	2.456±0.015^{c}	2.432±0.012^{c}	2.371±0.010^{d}	2.449±0.012^{c}	2.447±0.010^{c}
		(1,184)	(2,003)	(2,772)	(1,835)	(2,927)
MA	Murrah	2.358±0.031^{f}	2.357±0.023^{f}	2.437±0.021^{e}	2.607±0.026^{c}	2.251±0.020^{g}
		(1,439)	(2,623)	(3,288)	(2,150)	(3,592)
	Crossbred	2.859±0.035^{a}	2.681±0.027^{b}	2.503±0.023^{d}	2.675±0.028^{bc}	2.710±0.022^{b}
		(1,115)	(1,944)	(2,641)	(1,770)	(2,797)
TSC	Murrah	1,352.429±35.277^{a}	1,424.662±22.409^{a}	1,373.752±22.186^{a}	1,241.491±26.151^{b}	1,235.782±21.680^{b}
(million/ml)		(247)	(739)	(626)	(516)	(909)
	Crossbred	964.367±37.884^{c}	1,005.414±28.207^{c}	845.522±24.905^{d}	847.318±25.059^{d}	879.448±22.318^{d}
		(213)	(425)	(560)	(555)	(802)
IM (%)	Murrah	66.024±0.002^{bc}	65.710±0.001^{bc}	65.908±0.001^{bc}	68.982±0.002^{a}	65.328±0.001^{c}
		(1,429)	(2,532)	(3,160)	(1,993)	(3,436)
	Crossbred	66.947±0.003^{b}	63.943±0.002^{d}	61.207±0.001^{e}	64.912±0.002^{cd}	65.792±0.001^{bc}
		(1,118)	(1,848)	(2,496)	(1,665)	(2,709)
PTM (%)	Murrah	42.604±0.002^{de}	40.472±0.001^{h}	41.484±0.001^{g}	42.535±0.001^{ef}	43.555±0.001^{d}
		(768)	(1,277)	(1,529)	(1,102)	(1,924)
	Crossbred	46.681±0.003^{ab}	42.742±0.002^{cdf}	41.915±0.002^{ceg}	45.863±0.002^{b}	47.813±0.001^{a}
		(536)	(738)	(780)	(691)	(1,367)
AFSC (days)	Murrah	801.781±3.673^{a}	805.556±2.675^{a}	803.412±2.479^{a}	798.177±3.122^{a}	804.145±2.421^{a}
		(1,466)	(2,780)	(3,242)	(2,026)	(3,408)
	Crossbred	658.463±4.188^{bd}	647.116±3.118^{c}	655.106±2.693^{bc}	652.319±3.321^{cd}	662.348±2.650^{b}
		(1,120)	(2,026)	(2,722)	(1,791)	(2,806)
AFSF (days)	Murrah	914.809±4.558	922.522±3.326	915.774±3.075	920.270±3.855	916.607±3.006
		(1,423)	(2,691)	(3,151)	(1,985)	(3,303)
	Crossbred	722.825±5.424	715.750±4.037	720.253±3.518	716.520±4.335	723.537±3.390
		(997)	(1,800)	(2,377)	(1,569)	(2,555)

Data in parentheses indicate the number of observations.

^{a,b}Different superscripts indicate significant differences between two sub-effects (P<0.05).

Table 6: Least squares mean±S.E. for semen volume (SV), colour, consistency, MA, TSC, IM%, PTM%, AFSC and AFSF for bovine breeding bulls during different periods.

Period	SV (ml)	Colour	Consistency	MA	TSC (million/ml)	IM (%)	PTM (%)	AFSC (Days)	AFSF (Days)
Period 1	4.482±0.021^{c}	1.194±0.008^{a}	2.397±0.007^{d}	1.981±0.016^{d}	1,271.630±59.956^{a}	61.513±0.001^{d}	43.105±0.001^{c}	777.599±2.425^{a}	881.604±3.154^{a}
	(5.491)	(5.511)	(5.412)	(5.415)	(80)	(4.729)	(2.076)	(3.407)	(3.003)
Period 2	3.957±0.019^{d}	1.138±0.007^{b}	2.427±0.006^{c}	2.002±0.014^{d}	966.208±17.020^{d}	58.934±0.001^{e}	38.561±0.001^{e}	711.906±1.688^{c}	793.566±2.162^{b}
	(7.071)	(6.999)	(7.039)	(7.022)	(1.051)	(6.575)	(2.799)	(7.119)	(6.526)
Period 3	3.799±0.023^{e}	1.183±0.008^{a}	2.481±0.008^{b}	2.750±0.017^{c}	1,185.929±26.247^{a}	65.991±0.001^{b}	45.011±0.001^{b}	661.896±2.016^{d}	728.391±2.510^{c}
	(4.750)	(4.749)	(4.753)	(4.720)	(412)	(4.533)	(2,474)	(5.009)	(4.844)
Period 4	4.663±0.028^{a}	1.052±0.010^{c}	2.551±0.010^{a}	2.870±0.022^{b}	1,059.311±15.631^{c}	64.261±0.001^{c}	41.021±0.002^{d}	719.146±2.488^{b}	797.998±3.125^{b}
	(3.132)	(3.100)	(3.034)	(2.998)	(1.140)	(2.858)	(963)	(3.216)	(3.050)
Period 5	4.558±0.023^{b}	1.040±0.008^{c}	2.575±0.008^{a}	3.117±0.021^{a}	1,102.015±10.058^{b}	76.148±0.001^{a}	50.186±0.001^{a}	773.664±2.081^{a}	892.874±2.600^{a}
	(4.636)	(4.384)	(4.083)	(3.204)	(2.909)	(3.691)	(2.400)	(4.636)	(4.428)

Data in parentheses indicate the number of observations.

^{a,b} Different superscripts indicate significant differences between two sub-effects (P<0.05).

CONCLUSION

In crossbred cattle bulls, semen volume, colour, MA and PTM% were higher than Murrah buffalo bulls. While consistency, TSC, IM%, AFSC and AFSF were higher in Murrah buffalo bulls. In bovine breeding bulls, overall semen quality was found to be good during all seasons. But in Murrah buffalo bulls, per ejaculate semen volume, consistency, MA and TSC were comparatively lower during the winter, whereas in crossbred cattle bulls, they were comparatively lower during the rainy season. In bovine breeding bulls, IM% and PTM% during the rainy and summer seasons were comparatively low. Overall semen quality traits revealed an increase in semen quality in the later periods, showing progressive improvement in management at the organised farm over the period.

Conflict of interest: None.

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