



Effect of heterogenous cattle bull seminal plasma on seminal parameters of mithun

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ABSTRACT

The present study was conducted to assess the effect of replacement of cattle bull seminal plasma with that of mithun bull on seminal parameters of mithun bulls. After collection, the semen samples were divided into group 1 (control): without addition of heterogenous seminal plasma; group 2 to group 5: mithun semen with half and full replacement of good quality cattle seminal plasma and mithun semen with half and full replacement of poor quality cattle seminal plasma, respectively. Individual motility, livability, total sperm abnormality, acrosomal, plasma membrane and nuclear integrity were assessed. The result revealed that there was no significant difference between full and half seminal plasma replacement in both good and poor quality seminal plasma, but the seminal traits of half seminal plasma replacement has nonsignificantly higher value than full seminal plasma replacement. Comparison between the control, half and full seminal plasma replacement of good and half and full seminal plasma replacement of poor quality semen revealed a significant difference.

Key words: Heterogenous cattle seminal plasma, Mithun, Seminal parameters

Recent livestock survey indicated that population of the mithun (*Bos frontalis*), a semi-wild, free-range bovine species in the North-Eastern Hill (NEH) region of India, is decreasing due to lack of suitable breeding management. Use of AI for improvement of its pedigree is utmost essential. Among various factors, composition and concentration of seminal plasma is considered as the most important because the high concentration affects the semen quality as in bovine species (Baas *et al.* 1983). Composition and inhibitory factors varied between the species as buffalo bull semen contains higher content of inhibitory factors compared to cattle semen (Sahni 1990). Replacement of buffalo seminal plasma with that of cattle seminal plasma showed some beneficial effects on livability and preservation of buffalo bull spermatozoa (Ibrahim *et al.* 1981, Sahni and Mohan 1990). However, there was no study on effect of addition and replacement of cattle/ buffalo heterogenous seminal plasma on mithun semen and seminal parameters. Therefore, the objective of the present study was to investigate the effect of substitution of cattle (Jersey

crossbred) seminal plasma with that of mithun on seminal parameters.

MATERIALS AND METHODS

Apparently healthy mithun bulls (10) of approximately 4 to 6 years of age were selected from the herd derived from various hilly tracts of the NEH region of India. The average body weight of the bulls was 501 kg (493 to 507 kg) at 4–6 yr of age with good body condition (score 5–6) and were maintained under uniform feeding and managerial conditions. Each experimental animal was fed in this experiment as per the farm schedule. Semen was collected from the animals through rectal massage method. Ejaculates (25) were evaluated and accepted for evaluation if the following criteria were met: concentration: >500 million D ml; mass activity >3+, individual motility: >70% and total abnormality: <10%. During the study, all the experimental protocols met the Institutional Animal Care and Use Committee regulations.

Cattle bull seminal plasma was collected from Jersey crossbred bulls, maintained under uniform conditions of feeding and management at Frozen Semen Bank, Cuttack, Odisha. The seminal plasma was divided into good and poor quality based on the certain seminal parameters, viz. mass activity more than 3+ and above and individual motility 70% and above was considered as good quality ejaculates, whereas poor quality ejaculates had mass activity and individual motility less than 3+ and 70%, respectively. Semen from each fresh ejaculate was aliquoted immediately

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after collection and the sperm pellet and seminal plasma were separated by centrifugation at 4,000 rpm for 20 min at 4°C. The seminal plasma was stored at -80°C for further utilization.

After semen collection from mithun, semen ejaculate was divided into 5 groups, viz. group 1 (control): without addition of heterogenous seminal plasma; group 2 to group 5: mithun semen with half and full replacement of good quality cattle seminal plasma and mithun semen with half and full replacement of poor quality cattle seminal plasma, respectively. For replacement, seminal plasma from the 2 groups was removed by centrifuging the ejaculates at 3,000 rpm for 15 min (Ahmad *et al.* 1994). All the semen groups were diluted with standard tris egg yolk citrate glycerol extender at the rate of 1: 10 and stored at 37°C. The percentage of sperm motility, viability (eosine-nigrosine stain), total sperm abnormality, acrosomal integrity (giemsa stain: Watson 1975), the plasma membrane integrity (hypo-osmotic swelling test: Jayendran *et al.* 1984), nuclear integrity (Feulgen staining technique, Barth and Oko 1989) and vanguard distance travelled by sperm in cervical mucus (CMPT) (Matouseket *et al.* 1989) were determined as per standard procedures.

The results were analysed statistically and expressed as the mean $\pm$ SEM. Means were analyzed by student "t" test between the half and full seminal plasma replacement in both good and poor quality seminal plasma and one way ANOVA between the 5 groups, followed by the Tukey's post hoc test to determine significant differences between the 5 experimental groups on the sperm parameters using the SPSS/PC computer program (version 15.0; SPSS.). Differences with values of $P < 0.05$ were considered to be statistically significant after arcsine transformation of percentage data by using SPSS 15.

RESULTS AND DISCUSSION

The results revealed that there was no significant difference between full and half seminal plasma replacement in both good and poor quality seminal plasma, but the seminal traits of half seminal plasma replacement has nonsignificantly higher value than full seminal plasma replacement. Comparison between the control, half and full seminal plasma replacement of good and half and full

seminal plasma replacement of poor quality semen revealed that there was a significant ($P < 0.05$) difference (Table 1).

High concentration of seminal plasma in semen has adverse effect on semen preservation (Sahni 1990, Sahni and Mohan 1990, Ahmad *et al.* 1994). Centrifugation was used for reducing concentration of seminal plasma (Clay *et al.* 1984). In the present study, half replacement has nonsignificantly higher value of seminal parameters than full replacement indicating that half replacement is more beneficial than full replacement of seminal plasma in both good and poor quality seminal plasma. But comparison between the good and poor quality seminal plasma replacement, revealed that good seminal plasma has significantly ($P < 0.05$) higher seminal value than poor quality indicating that poor quality seminal plasma may contain some adverse or deleterious factors or lack of protective factors, which affected the seminal parameters. Similarly between the control and good and poor seminal plasma replacement, good quality seminal plasma revealed significantly ($P < 0.05$) higher value than other 2 groups. These findings are in agreement with Albright *et al.* (1958) and Ibrahim *et al.* (1981), who reported that preserving ability of buffalo bull spermatozoa improved with the replacement of half of the volume of seminal plasma of cow bull. Sengupta *et al.* (1977) observed similar results in buffalo seminal plasma replacement with cattle seminal plasma; and the seminal parameters improved as the dilution of the inhibitory factors believed to be present in higher concentration in the seminal plasma of buffalo bulls. The observed improvement in seminal parameters following the replacement of 50% of the volume of seminal plasma in mithun semen in the study was may be due to changes in molarity in seminal plasma. However, the possibility of presence of some other unknown factor(s) cannot be excluded. The improvement of seminal parameters in half seminal plasma replacement semen samples suggested that the molarity of the mixed seminal plasma is more suitable than complete and un-substituted seminal plasma semen samples. However, complete replacement of seminal plasma resulted in nonsignificant reduction in seminal parameters over half replacement of seminal plasma, indicating that permissible change in molarity was achieved when substitution was done to only half of the volume. These findings are in agreement with

Table 1. Effect of replacement of cattle bull seminal plasma with that of mithun bull on seminal parameters (mean $\pm$ SE)

Seminal parameters	Control	Good quality seminal plasma		Poor quality seminal plasma	
		Half replacement	Full replacement	Half replacement	Full replacement
Individual motility (%)	66.80 $\pm$ 2.52 ^c	69.32 $\pm$ 2.42 ^b	68.56 $\pm$ 2.54 ^b	50.60 $\pm$ 2.37 ^a	48.12 $\pm$ 1.99 ^a
Livability (%)	71.64 $\pm$ 2.35 ^c	77.04 $\pm$ 2.27 ^b	75.96 $\pm$ 2.13 ^b	55.80 $\pm$ 2.24 ^a	51.84 $\pm$ 2.75 ^a
Acrosomal integrity (%)	74.96 $\pm$ 2.84 ^c	80.20 $\pm$ 2.31 ^b	78.24 $\pm$ 2.35 ^b	57.32 $\pm$ 2.34 ^a	53.80 $\pm$ 2.17 ^a
Total sperm abnormality (%)	12.32 $\pm$ 2.47 ^c	8.46 $\pm$ 1.41 ^b	9.20 $\pm$ 1.69 ^b	18.52 $\pm$ 1.82 ^a	21.08 $\pm$ 1.79 ^a
Plasma membrane integrity (%)	73.68 $\pm$ 2.63 ^c	78.48 $\pm$ 2.16 ^b	77.84 $\pm$ 2.24 ^b	57.92 $\pm$ 2.46 ^a	54.28 $\pm$ 1.53 ^a
Nuclear integrity (%)	77.60 $\pm$ 2.36 ^c	80.88 $\pm$ 2.21 ^b	80.32 $\pm$ 2.32 ^b	56.73 $\pm$ 2.39 ^a	55.28 $\pm$ 1.92 ^a
CMPT (mm/h)	18.44 $\pm$ 1.87 ^c	24.88 $\pm$ 1.99 ^b	22.16 $\pm$ 1.92 ^b	16.83 $\pm$ 1.37 ^a	14.34 $\pm$ 1.09 ^a

Figures with same superscript (a, b, c) do not differ significantly in rows.

those reported by Bora and Rao (1969), since they showed that the half combination of seminal plasma improved significantly the keeping quality, while complete substitution of seminal plasma resulted in significant decrease in seminal parameters in this precious species.

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