



Effect of season on quality of mithun semen

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Received: 12 August 2014; Accepted: 19 November 2014

Key words: Incubation periods, Mithun, Season, Seminal parameters

Livestock census (2007) indicated that the mithun population is decreasing due to lack of suitable breeding management in this region. Natural breeding is followed in mithun farms with limitations such as cost and disease transmission. Hence, use of artificial insemination for improvement of its pedigree is utmost essential. Season is one of the important factors that influence the variation in semen quality and fertility (Mathevon *et al.* 1998). Heat tolerance and disease resistance capacity of indigenous cattle (*Bos indicus*) are better than exotic cattle (*Bos taurus*) bulls, characterized by lower values of sperm abnormalities (Rajoriya *et al.* 2013). Seasonal effects on semen quality were observed in cattle (Rajoriya *et al.* 2013), buffaloes (Sagdeo *et al.* 1991), and horse (Pickett *et al.* 1975). The objective of this study was to assess effect of seasons (winter and summer) on seminal parameters of mithun to pursue future semen collection and preservation protocols.

Apparently healthy mithun bulls (10), approximately 4 to 6 year-old with good body condition (score 5–6) were selected and maintained under uniform feeding, housing and lighting conditions in institute farm. Semen was collected through rectal massage method.

The collection seasons were split into 2, viz. summer (May to July), and winter (November to February). Ejaculates (30: 15 ejaculates from each season) were collected from the mithun. The average maximum and minimum ambient temperature (°C) and relative humidity (%) were 30.3 and 35.7 ($P < 0.05$), 24.3 and 29.7 ($P < 0.05$), and 77.9 and 75.7, respectively during summer and winter. Immediately after collection, the samples were kept in a water-bath at 37°C and evaluated for volume, mass activity and concentration. After the preliminary evaluations,

samples were subjected to the initial dilution with pre-warmed (37°C) Tris egg yolk citrate extender (TEYC). The partially diluted samples were then brought to the laboratory in an insulated flask containing warm water (37°C) for further processing. Diluted semen samples were kept in glass tubes and cooled from 37° to 5°C @ 0.2–0.3°C/min in a cold cabinet and maintained at 4°C during liquid storage. The percentage of sperm progressive motility and total sperm abnormality (Tomar 1997) were determined as per standard procedure in samples during storage of semen at 4°C in different hours of incubation (0, 12, 24 and 36 h).

The results were analysed statistically and expressed as the mean $\pm$ SEM. Means were analyzed by one-way analysis of variance (ANOVA), followed by the Tukey's post hoc test to determine significant differences between the different incubation periods and student 't' test was done between the summer and winter on the sperm parameters using the SPSS/PC computer program (version 15.0; SPSS, Chicago, IL). Differences with values of $P < 0.05$ were considered to be statistically significant after arcsine transformation of percentage data by using SPSS 15.

Results (Tables 1–2) revealed that fresh seminal parameters and total sperm abnormality and progressive motility of diluted semen nonsignificantly differed between the summer and winter. The progressive motility and total sperm abnormality significantly varied between the incubation period in both winter and summer.

The motive of this present research was to study the physico-morphological characteristics in different seasons in mithun bulls. A nonsignificantly higher semen volume in winter (1.90 ± 0.15 ml) in comparison to summer (1.80 ± 0.10 ml) was observed. A similar finding with higher volume of semen was reported during winter followed by summer in indigenous cattle bulls (Sinha and Prasad 1966). Ejaculate volume varies from breed to breed and within a breed from bull to bull (Rao and Rao 1996). The semen volume may be influenced by various factors like body weight, scrotal size, age, pre-coital stimulation, frequency and season of semen collection (Borgohain 1984).

Mean values of sperm concentration did not show any significant difference between winter and summer. Seasonal variations in the concentration of spermatozoa of *Bos taurus* and zebu bulls were not significant (Kulkarni and Bhosrekar

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1973). In this study, sperm concentration varied nonsignificantly between winter and summer and this variation may be due to frequency of semen collection from different bulls. However, winter have nonsignificantly higher sperm concentration than summer in this study (Rajoriya *et al.* 2013). The mass motility of fresh semen was examined in both seasons immediately after collections. No significant difference in mass activity was recorded in semen ejaculate of winter and summer. Similar reports were observed in indigenous cattle bull (Rajoriya *et al.* 2013).

The initial motility of semen was not found influenced by winter and summer. However, season appears to have some control over the initial motility especially winter and spring (Tomar *et al.* 1985). Humid hot season was not desirable for the production of highly motile spermatozoa. Initial progressive motility in mithun bull semen in present study was in agreement with Rao and Rao (1996). Similar result of mass and initial progressive motility in the present finding in both winter and summer is an indication that season does not have significant effect on these parameters.

In the present study, nonsignificant difference was observed in total abnormal sperm percentage between the seasons. Similar report was observed in zebu and buffalo bulls (Tomar *et al.* 1985). Moreover, Tomar *et al.* (1985) observed that there was no seasonal variation in the abnormal percentage of live sperms in Indian cattle. No seasonal variation in relation to sperm abnormality in mithun bulls indicated its superiority and better adjustment in both the extremes of temperature variation.

Table 1. Physico-morphological seminal attributes of fresh ejaculates during winter and summer in mithun bulls

Seminal parameters	Seasons	
	Winter	Summer
Volume (ml)	1.90± 0.15	1.80 ±0.10
Sperm concentration (×10 ⁶ /ml)	497.50±25.69	469.41±17.62
Mass activity (0–5 point scale)	3.30 ±0.2	3.40 ±0.10

Table 2. Mean (±SE) progressive motility percentage and total sperm abnormality for mithun semen following storage at 4°C for different storage times

Time of preservation	Season	
	Winter	Summer
Progressive motility percentage		
0 h	65.00±2.10 ^d	64.70±1.40 ^d
12 h	52.50±2.90 ^c	55.90±1.90 ^c
24 h	45.00±3.60 ^b	48.50±2.50 ^b
36 h	37.50±3.70 ^a	38.80±2.50 ^a
Total sperm abnormality		
0 h	5.20±0.50 ^a	5.30±0.30 ^a
12 h	7.40±0.80 ^b	7.50±0.60 ^b
24 h	9.60±1.20 ^c	10.60±0.80 ^c
36 h	12.10±1.40 ^d	13.50±0.90 ^d

Figures with same superscript (a, b, c, d) do not differ significantly ($P < 0.05$) in columns.

It was concluded from this study that most of seminal parameters showed nonsignificant difference between the seasons. This indicated no seasonal effect on seminal profiles in mithun semen and semen can be cryopreserved throughout the year in this hilly cattle.

SUMMARY

The present study was undertaken to assess the effect of seasons (summer and winter) on routine seminal parameters such as volume, concentration, mass activity, progressive motility and total sperm abnormality in diluted semen at 4°C for 0, 12, 24 and 36 h of incubation. No significant difference was observed between the seasons for fresh and diluted semen at different hours of incubation. But nonsignificantly higher fresh and diluted seminal parameters in winter than in summer were observed. However, a significant difference was observed between the incubation periods in both progressive motility and total sperm abnormality in both the seasons. It was concluded from the present study that most of seminal parameters showed nonsignificant difference between the seasons indicating that there was no seasonal effect on seminal profiles in mithun bull and mithun semen can be cryopreserved throughout the year.

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