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EFFECT OF BULL AGE AND STORAGE PERIOD ON VIABILITY AND FERTILIZING CAPACITY OF BUFFALO FROZEN SEMEN

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SUMMARY

The present study was carried out on of 198 frozen semen straws taken from 22 buffalo bulls (< 5, 5-10 & >10 years old) at three storage periods (1, 2 & 3 years) with three repetitions. Results revealed more improvement in the sperm viability and fertilizing capacity for bulls aged 5 – 10 years when compared to other age groups. Although there was a non-significant improvement in the quality tests of spermatozoa a non-significant decrease appeared in the conception rate of the buffalo frozen semen stored for three years or more. It was concluded that buffalo bulls aged up to 10 years have more efficiency in artificial insemination purposes based on their high semen quality and fertilizing capacity; storage of the frozen semen seems to depend more on the change in systems of freezing, thawing and handling than the length of the storage period

INTRODUCTION

Since buffalo spermatozoa are more susceptible to hazards during freezing than cattle spermatozoa (Raizada, et al., 1990; Kumar, et al., 1992; Mall, et al., 1997), it is intended to clarify those factors influencing directly the viability and fertilizing capacity of buffalo spermatozoa. Some of these factors are related to the freezing and thawing processes (Pursel, et al., 1985; Watson, 1995; Wielders, 1997), the plasma membrane integrity (Parks, et al., 1987; Crowe, et al., 1989; Lin, et al., 1993). However, individuality due to age of the bull (El-Azab, et al., 1998a) and length of the

storage period of the frozen semen (Lee, et al., 1977; Chinnaiya, et al., 1990) can not be ignored. Judging on these factors, firstly, depended on assessing the change in the physical characteristics of the frozen-thawed semen (Yousef, 1997; El-Azab, et al., 1998b). Now, more recent techniques like the hypo-osmotic swelling test, HOST (Correa & Zavos, 1994; Correa, et al., 1997) and the semen quality test, SQT (Wang, et al., 1998; Reddy & Bordekar, 1999) have been implemented as indicative to the fertilizing capacity. The present study aimed at clarifying the effect of age of the bull and length of the storage period on viability and fertilizing capacity of buffalo frozen semen.

MATERIAL & METHODS

In a laten square design, a total of 198 frozen semen straws, 0.5 ml capacity were taken from 22 buffalo bulls (< 5, 5-10 & >10 years old) at three storage periods (1, 2 & 3 years) with three repetitions. Each time, the frozen semen was thawed at 37°C for 30 seconds to estimate the following:

(1) Percentage of the post-thawing motility (PTM) according to Salisbury, et al. (1978).
(2) Percentage of live spermatozoa (PIS) using the eosin - nigrosin Staining technique (Campbell, et al., 1956).
(3) Percentage of intact acrosome (PIA) using the spermac stain technique (Oettle, 1986).
(4) The hypo-osmotic swelling test (HOST) indicating percentage of coiling and bending tails of spermatozoa (Jeyndran, et al., 1992).
(5) The semen quality test (SQT) according to Reddy & Bordekar (1999).

For most of buffalo bulls, under investigation, the conception rate (CR) was estimated on a total of 66 trials of insemination for 3682 buffalo cows inseminated during the period from January to December, 2002 according to the data base program allocated in the General Organization for Veterinary Services.

All data obtained were tabulated and statistically analyzed according to the statistical computer program, Statease (1986).

RESULTS

Regarding the effect of age of the buffalo bull, the present results revealed more improvement in the sperm viability and fertilizing capacity for bulls aged 5 – 10 years when compared to other age groups. In the mean time, although there was a non-significant improvement in the quality tests of spermatozoa a non-significant decrease appeared in the conception rate of the buffalo frozen semen stored for three years or more (Table, 1).

Table (1): Effects of age of the bull and period of the storage on viability and fertilizing capacity of buffalo frozen semen (Mean ± S.E.)

Factors	PTM (%)	PLS (%)	PIA (%)	HOST (%)	SQT	CR (%)
Age of the bull:						
- < 5 years	37.27 ± 0.46^{ab}	48.91 ±4.14^{ab}	80.46 ±0.59^{a}	26.36 ±2.82^{a}	1.71 ±0.22^{a}	62.50 ±1.03^{a}
- 5 – 10 years	40.42 ± 2.64^{a}	49.75 ±3.11^{a}	78.92 ±0.89^{b}	23.25 ±2.62^{a}	2.16 ±.018^{a}	60.08 ±1.50^{a}
- > 10 years	34.14 ± 0.71^{b}	43.73 ±2.10^{a}	78.56 ±0.48^{a}	24.78 ±1.01^{a}	2.00 ±0.10^{a}	62.86 ±0.36^{a}
Period of storage:						
- 1 year	35.86 ±1.14^{ab}	46.74 ±1.58^{a}	78.57 ±1.22^{ab}	23.26 ±1.42^{b}	1.43 ±0.08^{b}	62.47 ±0.81^{a}
- 2 years	32.69 ±1.93^{b}	42.23 ±2.72^{b}	77.00 ±0.98^{b}	30.00 ±1.73^{a}	3.11 ±0.360^{a}	63.30 ±0.87^{a}
- 3> years	38.21 ±2.60^{a}	48.29 ±3.20^{a}	80.14 ±0.47^{a}	20.21 ±1.95^{b}	2.73 ±0.30^{a}	60.15 ±1.76^{a}

Values with different superscripts within the same column for the same Factor are significant at least at P < 0.05

DISCUSSION

It has been emphasized that the semen quality and freezability of spermatozoa for some buffalo bulls are generally inferior comparing to others under the same conditions of management, a finding which might be attributed to species variation (Chinnaiya and Ganguli, 1990; Anchieta et al, 2001), age of the bull (Stalhammar, et al., 1989; Rao and Streemannarayana, 1996) and even individuality (Siratskil, 1993; El-Azab et al., 1998a). However, semen manipulation during freezing, thawing and handling can not be ignored (Salisbury, et.al., 1978). Without justification of all these events, profitability of producing semen of good fertilizing capacity will be absent.

Although no single test or a combination of tests has been proven to be efficiently good to judge on the enfutured fertility rate of a bull, a high significant positive correlation has been indicated between fertility and percentage of the post-thawing motility of spermatozoa (Gibson and Graham, 1969; Chandler, et al., 1978). From the present study, it has been found that the percentage of PTM of buffalo bull spermatozoa differed significantly between the different age groups. It appeared much better at 5 - 10 years old after which it declined. From field experiment, it was noticed that the semen quality of the buffalo bull is improved by advancing age of the bull as the libido is improved and the testes become more active. On the other hand, it has been found that the percentage of PTM of spermatozoa seems to be more fluctuated between periods of storage of the frozen semen of buffalo bulls. Such fluctuation might be attributed to the different trials adopted to establish the extender being used (Belorkar, et al., 1993; El-Azab, et al., 1998b), the system of freezing (Shannon, 1978; Salisbury et al., 1978) and the technique of thawing (Hube et al., 1983; Youssef, 1997) and handling the frozen semen. Parallel to any fluctuation in percentage of the PTM of spermatozoa, there is a positive correlation to percentage of the live spermatozoa, a finding which was confirmed in a previous study (El-Chahidi et al, 1977).

The percentage of cells that have an intact acrosome and are able to

perform the acrosome reaction upon triggering is regarded as important semen characteristic (De leeuw,et al., 1991; Wielders, 1991). Thereby, a high correlation with fertility has been indicated for the percent intact acrosome of spermatozoa (Saake and White, 1972; Berndtson, et

al.,1981). The PIA of buffalo bull spermatozoa was found to decrease non-significantly by advancing age of the bull, a finding which came in accordance with some previous reports (Stalhammer, et.al., 1989; Chinnaiya and Ganguli, 1990). However, a significant difference in PIA of buffalo bull spermatozoa was observed in relation to period of storage of the frozen semen. Such difference might be attributed to the change in techniques of processing, storage and thawing the frozen semen (Youssef, 1997; El-Azab et al., 1998b).

Cryopreserved spermatozoa are very susceptible to changes in the osmotic conditions encountered during freezing and thawing (Fisher and Fairfull, 1984; Zavos, 1991). Abrupt changes in osmotic pressure result in occurrence of osmotic shock, reduced sperm viability and sperm membrane damage (Correa and Zavos., 1995; Correa, et al., 1997). From the present study, an improved in the HOST of buffalo frozen semen incorporated with buffalo bulls aged 5 – 10 years. In our concern, this finding might be attributed to inability of either the young or advanced aged bulls to resist any change in the semen buffering capacity. Moreover, a significant change in the HOST of buffalo frozen semen associated with the change in period of the storage, a finding which came in consistent with the close relationship to the change in percentage of PTM and PIA of spermatozoa (Correa and Zavos, 1995; Correa, et al., 1997).

The semen quality test is implemented with the HOST to evaluate the fertilizing capacity of spermatozoa by measuring the changes in sperm membrane functional status and permeability (Correa ,et al., 1997). fluctuant results were noticed in the SQT between different ages of bull and buffalo

bulls, a finding which support that the semen quality is greatly influenced by the change in age of the bull (El-Azab et al, 1998a). As being expected the SQT might be altered in association with the change in techniques of freezing, storage and handling of the frozen semen. This finding was emphasized in the present study from the significant difference in the SQT between the different years of processing the frozen semen like that in the HOST.

The conception rate seemed to be nearly comparable for the different age groups and periods of storage of buffalo frozen semen. This finding suggested that the conception rate depends mainly on the semen quality and efficiency of processing, storage and handling the frozen semen rather than either age of the bull itself or period of the semen storage. For this reason, a buffalo bull might be utilized several years in production of the frozen semen as long as its fertility is high and being acceptable to the producer and inseminator.

From the present study, it can be concluded that, buffalo bulls aged up to 10 years have more efficiency in artificial insemination purposes based on their high semen quality and fertilizing capacity; storage of the frozen semen seems to depend more on the change in systems of freezing, thawing and handling than the length of the storage period.

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