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Polymorphic Sperm Phenotype Suggesting Genetic Sperm Defects in a Jersey (*Bos taurus*) X Zebu (*Bos indicus*) Crossbred Bull

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ABSTRACT

Data on semen collections from a Jersey X Zebu crossbred bull, consistently producing semen with high proportion of sperm abnormalities was collected and the semen characteristics were analyzed. Observation of the records of 58 ejaculations from the bull has revealed that 96.6% of the ejaculates were rejected for poor semen quality especially in sperm morphology. A representative semen sample from the bull also showed sperm abnormalities up to 72.9%, of which 14.9% and 14.5% of the spermatozoa exhibited loose heads and microcephalic heads respectively. Coiled tail (16.7%) and short tail (14.9%) were the main tail defects observed. Since sperm defects were consistently appearing in high proportions in almost all the ejaculates the genetic cause of the sperm defect was suspected.

Keywords: Bull spermatozoa, Polymorphic sperm phenotype, Genetic sperm defects

Abnormal morphology of sperm is a noncompensable trait and the merit of the sperm morphology assessment as an *in-vitro* fertility test of bulls has been well established. Further, the relationship of proportion of morphologically normal and abnormal spermatozoa with conception has also been proven (Padrik and Jaakma, 2001). The most common causes of sperm abnormalities are environmental, andrological or endogenous especially for nonspecific head and flageller defects (Chemes and Rawe, 2003; Chenoweth, 2005). However, certain types of head shape and tail defects are genetic in origin. The causative factors lead to impaired spermiogenesis and sperm maturation (Chenoweth, 2005) which more often results in specific sperm defects. Since the sperm head, which carries the genetic material from the sire to the offspring, plays a vital role in the fertilization process, even a small proportion of sperm head defects significantly affects fertility (Al-Makhzoomi *et al.*, 2008). Decapitated sperm, microcephalic sperm heads and short tail are some of the genetic sperm defects which influence the fertility

adversely (Chenoweth, 2005; Chemes, 2012). The present study is on a Jersey X Zebu crossbred bull, repeatedly producing ejaculates containing high proportion of different types of sperm defects.

A Jersey X Zebu crossbred bull, aged 30 months, maintained at a frozen semen production centre in Tamil Nadu State, India was reported to have consistently producing ejaculates with high proportion of sperm abnormalities. The ejaculates from the bull were evaluated for semen volume, sperm concentration, sperm motility and sperm morphology by conventional methods using phase contrast microscope and photometer. Morphological evaluation was done using 3% Rose Bengal stained sperm slides. A total of 200 spermatozoa were counted for assessment of sperm abnormalities. Semen characteristics recorded for the 58 ejaculates collected from the bull at the frozen semen production centre were studied. A representative ejaculate from the bull was taken using artificial vagina and the semen sample was evaluated for

semen characteristics including sperm morphology as described earlier.

Since the first collection in January 2013, a total of 58 ejaculates were taken from the bull, till February 2014. Almost all the semen samples (96.6%) were rejected because of poor sperm motility and high sperm abnormalities, exceeding the 20% permissible level. The average values recorded for the sperm characteristics were 3.56 ml, 1085.28 million/ml and 18.4% for ejaculate volume, sperm concentration and sperm motility respectively.

The ejaculate from the bull collected for the present study consisted of 4.4 ml of semen, containing 1367 million spermatozoa per ml of semen. The sperm motility was 70%. The morphology evaluation of the stained sperm slides revealed that majority of the spermatozoa were abnormal of which the head and tail defects were in high proportions (Table 1).

Table 1. Sperm abnormalities in the representative ejaculate of the Jersey x Zebu crossbred bull

Type of abnormalities	Percentages
Loose heads	14.9
Microcephalic heads	14.5
Macrocephalic heads	1.80
Pyriform heads	4.52
All head defects	35.7
Coiled tail	16.7
Short tail	14.9
Bent tail	4.52
Broken tail	0.90
All tail defects	37.0
Overall abnormalities	72.7

The polymorphic phenotypes of spermatozoa are depicted in the Fig. 1. Loose heads and microcephalic heads were the main head abnormalities detected, while a few macrocephalic and pyriform heads were also seen. Most of the tail defects were coiled tail and short tail and a few bent and broken tails were also noticeable. Only 27.1% of the spermatozoa appeared normal in morphology. Nearly 50% of the loose heads were normal in shape and size and major proportion of the remainder were microcephalic.

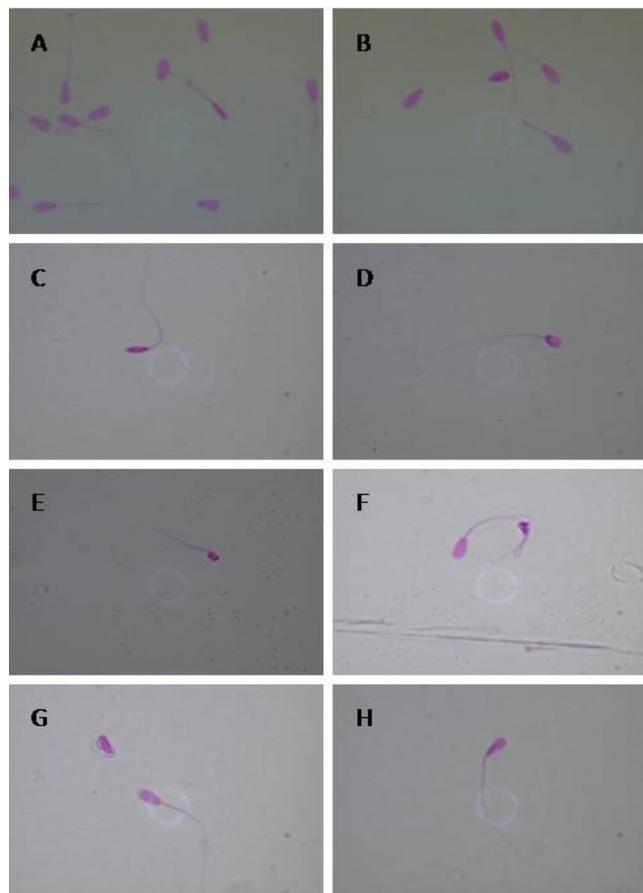


Figure 1. Polymorphic sperm phenotype in a Jersey X Zebu crossbred bull (A) Sperm with loose heads, microcephalic head and coiled tail (B) Sperm with loose heads and short tail (C) Microcephalic sperm head elongated (D) Microcephalic sperm head rounded (E) Microcephalic sperm head rounded with short tail (F) Microcephalic sperm head conical with coiled tail and a sperm with normal head and coiled tail (G) Microcephalic sperm head with coiled tail and a sperm with normal head with bent tail (H) Sperm with pyriform head

Very high proportion of microcephalic sperm has already been recorded in bulls (Hancock and Rollinson, 1949; Alun-Jones, 1962). Settergren and Nicander (1968) and Pant *et al.* (2002) found lower percentages of decapitated sperm heads in bull semen. In our study also relatively lower proportion of loose heads (22.6%) was recorded. Decapitated sperm defect was found to be a sterilizing defect in bulls (Hancock and Rollinson, 1949; Chenoweth, 2005) especially if present in very high proportion. Loose heads, headless tail and abnormal alignment are due to abnormalities in sperm centrioles and the head-neck junction due to genetic causes (Chemes, 2012). And the

hereditary nature of the defect has been well documented (Hancock and Rollinson, 1949; Alun-Jones, 1962; Chemes and Rawe, 2003).

Occurrence of microcephalic and macrocephalic sperm along with normal spermatozoa in spermogram of bulls has been reported (Revay *et al.*, 2010; Morrell *et al.*, 2014). In the present study, the microcephalic heads were seen in various sizes and shapes. Elongated and rounded microcephalic heads were more frequent in appearance while a few of the sperm heads were conical. Most of the microcephalic spermatozoa carried the normal tail while a few sperm had short tail or coiled tail having normal length. Isolated macrocephalic sperm with normal or short tail were also seen. A small proportion of spermatozoa showed pyriform heads. The fertility of males having sperm head defects may be depended on the degree of the abnormality (Nothling and Arndt, 1995) possibly due to reduction of its total surface area (Siqueira *et al.*, 2010). Even a small proportion, up to 10 to 20% of abnormal sperm heads can lead to sub-fertility (Al-Makhzoomi *et al.*, 2008). Therefore, sperm head shape and size deviations even in small proportions as it appears in the present case, should be viewed with caution.

The sperm picture of the bull in question has shown significant proportion of tail abnormalities namely, short tail, coiled tail, bent tail and broken tails. Of these, coiled tail and short tail defects were predominant. The coiled tails did not show any characteristic pattern to suggest any specific abnormality. Bent tail and broken tails were relatively in very small proportions. Short tail defect, a specific sperm defect has occurred in bulls (Siqueira *et al.*, 2010). It is also known to affect the fertility of the affected individual at varying degrees from sub-fertility, infertility to sterility (Kopp *et al.*, 2008). Faulty spermiogenesis causing short tail defect may be because of certain environmental factors (Siqueira *et al.*, 2010) or may be of genetic origin involving a single autosomal recessive gene with a complete penetrance in males (Andersson *et al.*, 2000; Kopp *et al.*, 2008).

CONCLUSION

Consistent occurrence of sperm abnormalities in the bull suggested impaired spermiogenesis. Environmental factors causing faulty spermiogenesis could not be suspected in the present case as many other bulls maintained in the

same environment and management conditions produced ejaculates of good quality and were successfully used for frozen semen production. Therefore, the abnormal sperm phenotype and pattern of its occurrence in the Jersey X Zebu crossbred bull was suggestive of genetic sperm defects and advice was given to eliminate the bull from the semen collection programme.

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