KARYOLOGICAL SCREENING OF ANESTROUS HEIFERS

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Fertility is a multifactorial phenomenon which is influenced by several factors that often overlap. Unlike many other causes of reproductive failure, little attention appears to be given towards cytogenetic anomalies in farm animals (Yimer and Rosnina, 2014). Chromosomal abnormalities, such as translocations, tandem fusion and chimerism for sex cells, sex chromosome aneuploidy are among few important abnormalities in cattle, which are reported to be associated with variable degrees of infertility. Present study was undertaken to find out the incidence of chromosomal defects in reproductively abnormal animals and to recommend suitable strategies to eradicate such chromosomal abnormalities from the population.

MATERIALS AND METHODS

Five apparently normal heifers and 10 anestrous heifers (3 Frieswal, 3 Gir and 4 Holstein cross) were included in the present study. The history of the animals was obtained to judge the fertility status of heifers in relation to their age. Blood samples (5 ml) from each animal were collected to obtain chromosomal preparations. Lymphocytes were cultured in RPMI-1640 medium enriched with tryptose phosphate broth. Pokeweed mitogen (PWM) was added at the rate of 0.1 ml per culture vial. The media was supplemented with foetal calf serum (20%) just before addition of blood (0.5 ml) to the culture vials. Slides were stained with Giemsa and from each animal a minimum of 25 metaphase spreads were screened using trinocular research microscope (Leica-DM3000). For identification of individual chromosomes, G and C bandings were performed as per the standard procedures (Seabright, 1971; Sumner, 1972). Chromosome number, general morphology of chromosomes and the chromosomal abnormalities in number and structure, if any, were identified and catalogued. Abnormalities *vis* a *vis* reproductive status was studied to establish the association, if any.

RESULTS AND DISCUSSION

Cytogenetic screening of heifers belonging to different genetic groups revealed a modal chromosome number of 2n = 60. The karyotype of all the animals comprised of 29 pairs of acrocentric autosomes and one pair of sex chromosomes. The X chromosomes were larger submetacentric. Apparently, none of the animal revealed any deviation from the normal diploid chromosome number of cattle. Appannavar *et al.* (2004) and Mukherjee and Yadav (2004) also reported similar findings in different breeds of indigenous cattle. Screening of slides revealed presence of structural abnormalities like gaps, breaks, deletions, centromeric attenuations and chromosome fragmentation (Figure 1). The numerical abnormalities were recorded in the form of polyploidy. The frequency of gaps, breaks, deletions and polyploids were low in both normal and reproductively abnormal animals (Table 1). Whereas Sarkhel and Katpatal (1997) have demontrated higher frequencies of gaps, breaks, deletions and polyploidy cells in reproductively abnormal animals. Hassanane *et al.* (1995) illustrated that increased rates of chromatid breaks and gaps had been related to lowered fertility and anestrum in animals. Gaps and breaks points to the missing of genes carried by the deleted portion of chromosome. However, lower frequencies of gaps, breaks and deletions obtained in this study could not have affected fertility performance of animals. An aging heifer of Frieswal genetic group showed chromosome fragmentation and centromeric attenuations in 7 % and 8 % of metaphases, respectively. Saleh (2008) also reported a significant increase in frequency of centromeric attenuations, gaps, breaks and fragments in a group of anestrum cattle. The presence of centromeric attenuation and fragmentation in one anestrous heifer, might have some practical utility.

Table 1: Frequency (%) of various chromosomal abnormalities.

S. No.	Chromosomal abnormalities	Control animals	Anestrous heifers
1.	Gaps	6 (5)	4(10)
2.	Breaks	4 (5)	5(10)
3.	Deletions	2(1)	3(2)
4.	Fragmentations	0(0)	7(1)
5.	Centromeric attenuations	0(0)	8(1)
6.	Polyploidy	2(1)	1(1)

Figure in parenthesis indicate number of animals showed chromosomal abnormalities.

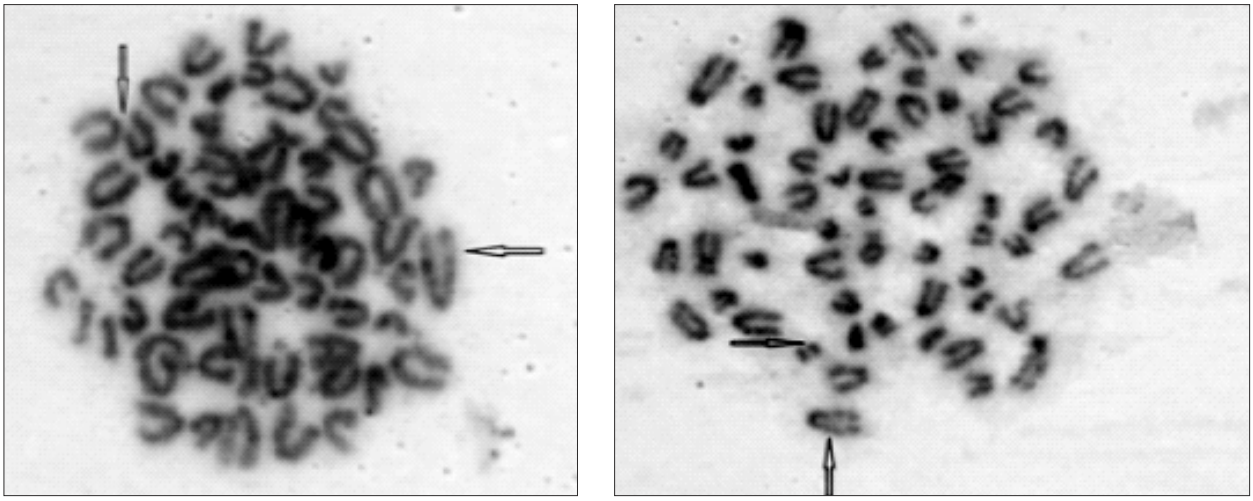

Figure 1: Metaphase plates of heifers showing (a) gaps and breaks, (b)gaps and centromeric attenuations.

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