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## RESEARCH ARTICLE

## KARYOTYPING TO IDENTIFY INDIGENOUS MALE PARENTAGE IN CROSSBRED CALVES

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### Abstract

Cytogenetics is the study of structure and properties of chromosomes and chromosomal behaviour during cell division. Chromosome analysis is done routinely to examine the animals for breeding soundness and selection. A karyotype is the arrangement of chromosomes in homologous pairs in a systematic manner. The diploid chromosome number in cattle is 60 (29 pairs of autosomes and one pair of allosomes). The unique chromosomal feature that differentiates cattle of exotic origin (*Bos taurus*) from that of indigenous cattle (*Bos indicus*) is the structure of the Y-chromosome. The *Bos taurus* bulls have meta- or submetacentric Y-chromosome whereas the *Bos indicus* bulls bear an acrocentric Y-chromosome. Blood samples from the selected males which resembled Jersey crossbreds phenotypically were received at this department for karyotyping. Short-term leucocyte culturing was carried out with each sample. The karyotypes were made from good spreads by arranging the chromosomes in homologous pairs using Applied Spectral Imaging software. Analysis of karyotypes of 30 samples revealed that three karyotypes were found with acrocentric Y-chromosome. The acrocentric Y-chromosome should have been inherited from the indigenous sires only. Though it is not possible to identify the exact parentage by using this method, this is one of the best and cheapest methods to eliminate crossbred calves born to indigenous sires.

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## INTRODUCTION

A karyotype is a full set of metaphase chromosomes from one diploid cell arranged in descending lengths. Cattle have 60 chromosomes, 29 pair of autosomes and 1 pair of sex chromosomes. In spite of their common ancestral base Asiatic Zebu cattle (*Bos indicus*) and European taurine cattle (*Bos taurus*) exhibit several morphological and physiological differences (Halnan and Watson, 1982). The karyotypes of these two bovine sub-species are similar, consisting of 58 acrocentric autosomes and two sexual chromosomes, a submetacentric X chromosome and a Y chromosome whose morphology varies depending on the sub-species (Kieffer and Cartwright, 1968); the Y-chromosome found in *B. taurus* being submetacentric while that in *B. indicus* is acrocentric. The Y is established as a marker for paternal descent in defined cases.

## Materials and Methods

In Tamil Nadu state, all young bulls selected for AI have been subjected to chromosome analysis. As per the state government breeding policy for cattle, the exotic level of inheritance should be maintained at a level of 50 per cent. Hence production of high genetic merit crossbred bulls is of great importance. One of the methods is by field Progeny Testing by which crossbred / non-descript females are inseminated with crossbred / exotic purebred semen respectively and the calves thus produced were selected for production of semen for AI. As these bull mothers are maintained in farmer's herds, verification of the parentage becomes essential to establish their exotic inheritance.

Blood samples were received in heparinized vacutainers from 30 select crossbred bulls. Short-term lymphocyte culture technique was adopted as per Halnan (1977) with slight modifications. Each culture contained 7.5 ml of RPMI culture medium, 2 ml of autologous plasma, phytohemagglutinin (10µg/ml) and little buffy coat. The culture was incubated at 37° C for 72 h and colchicine was added one-and-half hour prior to harvesting. Potassium chloride hypotonic (0.075 M) treatment was given for 20 minutes at 37° C and fixations were done in methanol: acetic acid (3:1) Carnoy's fixative. Air-dried smears prepared in slides were stained with 2 per cent Giemsa for 10 minutes. Slides were screened for non-overlapping metaphase spreads with complete chromosome complement and were photographed and karyotyped as per standard nomenclatures (ISCNDA, 1989; ISCNDB, 2000).

## Results and Discussion

Out of the 30 samples karyotyped, all the samples had 60, XY karyotype indicating the male sex. The X-chromosome was large sub-metacentric and the Y-chromosome was small metacentric in 27 of the samples, while three samples had acrocentric Y-chromosome, size comparable to that of the 29<sup>th</sup> pair of autosomes (Fig 1 and 2).

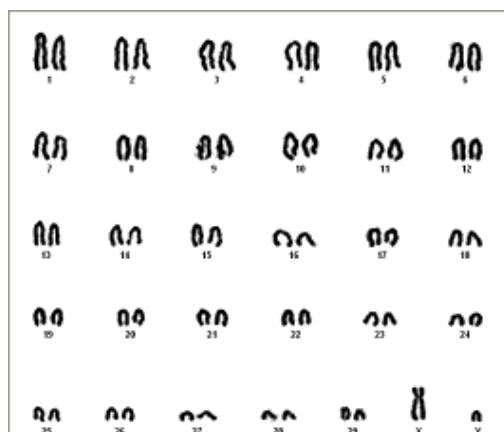


Fig 1. *Bos indicus* bull karyotype

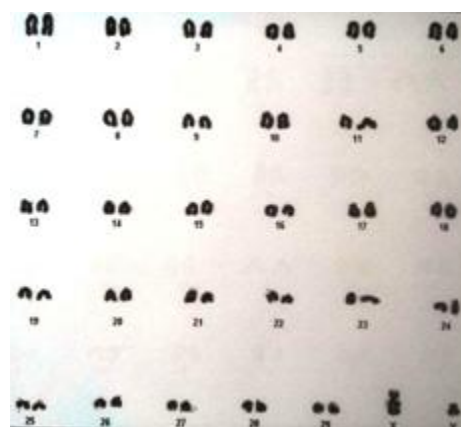


Fig 2. *Bos taurus* bull karyotype

Between *Bos indicus* of Indian origin and *Bos taurus* of European origin, Y-chromosome polymorphism is a well known phenomenon (Halnan and Francis, 1976; Stranzinger *et al.*, 1987). The bull samples were received from bull calves supposed to be of high genetic merit and born to exotic bulls through AI. It is understood from the fact of Y-chromosome inheritance that the calves with acrocentric-Y must have been inherited from the indigenous bulls only. This could be due to the fact that mismating had happened and the conception was effected with indigenous sperm. Though other techniques are available to identify the parentage this is a simple and effective method which could be performed easily even in field conditions and the bulls that show acrocentric-Y can be culled at an earlier stage of selection which could prevent the economic loss involved in Progeny Testing of these bulls.

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