# Genetic conservation of chromosome 1 of cattle, goat, sheep and river buffalo revealed by comparative FISH mapping

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ABSTRACT

By employing comparative physical mapping of five bovine chromosome 1 specific cosmid derived microsatellites, the hypothesis of conservation of genomic structure in four species of the family bovidae was examined. FISH mapping data revealed that the five cosmid clones mapped to identical band locations of homologous cattle and goat acrocentric chromosome 1 and the long arms of sheep and river buffalo meta/submetacentric chromosome 1 (1q). The study thus extends the physical gene maps of the four species and documents that within bovid chromosomes, homology of banding patterns corresponds to homology in genetic structure.

**Key Words:** physical mapping, Genetic conservation, Genetic homology, Chromosomal banding homology

## INTRODUCTION

In recent years the specifics about mammalian genome evolution has been vastly improved by means of DNA sequence comparison, comparative gene mapping studies and by comparative cytogenetics. Comparative chromosome banding studies suggested that goat chromosome 1; long arm of sheep chromosome 1 and long arm of river buffalo chromosome 1 are the equivalents of cattle chromosome 1 (Ford et al 1980; ISCNDA 1989; Hayes et al 1991; Kaftanovskaya and Serov 1994; CSKBB 1994, Iannuzzi and DiMeo 1995, DiBerardino and Burghete 1998). As each G, R or Q band spans over several megabases of the genome, it was appropriately realized that the pronounced correspondence of banded karyotypes of bovids might not essentially correspond to genetic homology (ISCNDA 1989, Chowdhary et al 1991). Comparative gene mapping information across species verifies the proposed cytogenetic homologies but are hitherto not amply developed to obtain fine comparisons especially when all the four species i.e. cattle, river buffalo, sheep and goat are considered simultaneously. If sufficient comparative chromosomal banding and gene mapping data is accessible in various species, the types and numbers of chromosomal reorganization incorporated in the lineage can be deduced. By extension, such evaluation will allow to depict karyotype evolution and consequently ascertain the primitive karyotypes and gene maps (Qumsiyeh and Baker 1988).

Fluorescence in situ hybridization (FISH) has proved to be a powerful tool for extending physical maps of various domestic species, especially for locating loci along the chromosomes. This facilitates the comparison of the physical organization of different genomes. Domestic cattle is the most extensively studied domestic species with more than 4600 loci assigned (Iannuzzi et al 2003b, Ihara et al 2004, Womack 2005). In other bovids, the physical maps are relatively poor especially when referring to specific chromosomal regions for comparison.

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The imperative objective is to physically localize gene/genetic markers on each of the homologous cattle, buffalo, sheep and goat chromosomes to validate that the similarities in banding patterns at the chromosomal level do reveal similarities in genetic organization at the DNA level. Consequently, in this study five autosomal type II loci, earlier FISH mapped in cattle (Prakash et al unpublished) were comparatively FISH mapped in river buffalo (BBU), sheep (OAR) and goat (CHI) chromosomes noticeably extending the comparative physical map in these four economically important bovids.

## MATERIAL AND METHODS

*Chromosome preparation*: metaphase chromosome preparations were obtained from pokeweed stimulated peripheral blood lymphocyte cultures using standard technique. To increase the yield of metaphase spreads and to obtain elongated chromosomes, cells were cultured in the presence of ethidium bromide (20μg/ml) during the last hour of incubation.

*Probes and their labeling*: A cosmid library was constructed from a female of Norwegian Red cattle (NRF) breed. Screening of cosmids was performed using a $(GT)_{10}$ oligonucleotide. Clones giving a positive signal to this probe and negative signals to bovine satellites 1709, 1715 and bovine cot 1 DNA were selected (Olsaker 1996). Cosmid DNA was purified using QIAGEN tips plasmid kit, labeled with biotin-14-dATP by nick translation using the BRL commercial nick translation system (Gibco-BRL-8247-SA) following the manufacturer's protocol with minor modifications as detailed earlier (Chowdhary et al 1995).

*In situ hybridization and signal detection*: Fluorescence in situ hybridization (FISH) was performed according to Chowdhary et al (1995), with modifications. Briefly, the post hybridization washings in 50% formamide were carried for 3 X 10 min instead of 3 X 15 min. The hybridization signals and chromosome images were screened, captured and processed using appropriate

across a wide range of mammalian species. The five type II loci (non coding DNA sequences) commonly FISH mapped in this study demonstrates that even non-coding sequences are precisely conserved in the four boivds. Further genetic map development is needed to improve the efficiency of genome screens in a larger number of bovidae species especially in terms of a more uniform coverage of map regions with highly polymorphic markers. It will also be vital to increase the number of links between the genetic maps and the sequence maps of other species, so as to be better able to use comparative information to assist with genome screens for traits of interest in economically important bovidae species.

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