TELOMERIC BANDS ON *BOS TAURUS* MITOTIC CHROMOSOMES

BANDAS TELOMÉRICAS EN CROMOSOMAS MITÓTICOS DE *BOS TAURUS*

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Additional keywords

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Palabras clave adicionales

Bandas T. Secuencias teloméricas. Brazos eucromatínicos cortos.

SUMMARY

The quantitative pattern of T-banding was described for bovine chromosomes. The telomeric and pericentromeric T bands covered about 21 p. cent of chromosomal length. There were only five interstitial bands (7q13, 7ql5, 7q22, 7q24, 16q16-21). We have not found any specific distribution definited intracromosomal localization (Eggen and Fries, 1995) between T (5 genes), R (10 genes) and G (7 genes) bands. The intrachromosomal position of (TTAGGG)n telomeric sequences was determined by direct R-banding FISH. The hybridization signals were observed at the pericentromeric regions and termini of all autosomes. The sex chromosomes have got only telomeric pattern. Any interstitial sites were absent. According to the results and our data concerning satDNA II and IV intrachromosomal distribution it's possible to speak about chromatic arms of cattle autosomes.

RESUMEN

Se describió el patrón cuantitativo de las bandas T en cromosomas bovinos. Las bandas T teloméricas y pericentroméricas cubrían alrededor del 21 p. cien de la longitud cromosómica. Sólo había cinco bandas intersticiales (7q13, 7ql5, 7q22, 7q24, 16q16-21). No hemos encontrado ninguna distribución específica de genes con localización intracromosómica definida (Eggen y Fries, 1995) entre las bandas T (5 genes), R (10 genes) y G (7 genes). La posición intracromosómica de las secuencias teloméricas (TTAGGG)n fue determinada mediante bandeo R directo y simultáneo con la hibridación *in situ* fluorescente (FISH). Las señales de hibridación se observaron en las regiones pericentroméricas y terminales de todos los autosomas. Los ceromosomas sexuales sólo poseen patrón telomérico. No existían localizaciones intersticiales. De acuerdo con estois resultados y con nuestros datos acerca de la distribución intracromosómica del DNA satélite II y IV, es posible hablar de brazos eucromatínicos cortos en los autosomas bovinos.

INTRODUCTION

It is very typical for modern cytogenetic using a lot of methods characterized structural and functional

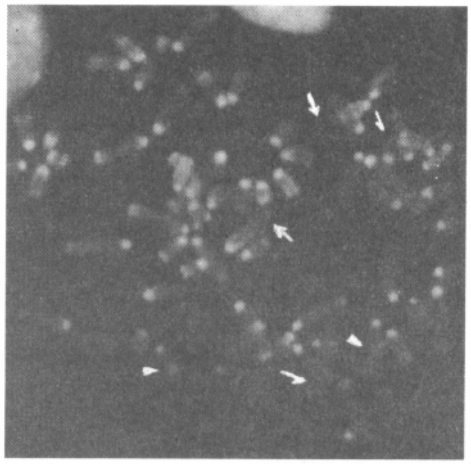

Figure 1. *The distribution of THA stained blocks along* Bos taurus *mitotic chromosomes. Interstitial T bands on chromosome 7 (→) and chromosome 16 (▻); sex chromosome (⇀).* (Distribución de los bloques teñidos con THA entre los cromosomas mitóticos de *Bos taurus*. Bandas intersticiales en el cromosoma 7 (→) y cromosoma 16 (▻); cromosoma sexual (⇀)):

aspects of chromosomal heterogeneity, communication between cytochemical and molecular levels of resolution. So called T bands bring notice from this viewpoint as GC richest and maximally termoresistant fraction of R blocks. For the first time specific T method of differential staining have been used for human chromosomes (Dutrillaux, 1973 and 1977). Then the data appeared about telomeric localization of T bands its enrichment of short intermediate repetitive DNA sequences (SINEs), earliest replicating in S phase (Holmquist, 1990; Bernardi, 1995). The most impressivable phenomenon is the highest gene concentration in T bands according to different methods of estimation such distribution (Saccone *et al.*, 1992; Craig, Bickmore, 1993 and 1994). The cytogenetical study of T banding is not satisfactory in spite of undoubted biological importance of the phenomenon. There are no practically data concerning principles of distribution of such blocks along chromosomes of other apecies except Homo sapiens. That is why we were going to describe the location of T bands for *Bos taurus* mitotic chromosomes, estimate its telomeric origin and comparative density of localized genes.

MATERIAL AND METHODS

The chromosomal slides have been prepared from stimulated peripherial blood lymphocytes of 16 black and white anlmals. Bovin chromosomes were identified by RBA methods of differential staining. T banding was done according to THA staining procedure (Dutrillaux, 1979). For *in situ* hybridization the probe contained 181 b.p. insection of TTAGGGn telomeric repeat in Eco R1 site of Bluescript KS vector was used (Weber *et al.*, 1990). The biotinylation of the probe was achieved by nick-translation in presence of biotin-11-dUTP. After hybridization the slides were rinsed and procesed for detection of bybridization signal with FITC (Lichter *et al.*, 1991).

RESULTS AND DISCUSSION

The pericentromeric and telomeric regions of the most bovine chromosomes revealed bright fluorescence after THA staining **(figure 1)**. The chromosome 7 was one of the exceptions without

telomeric T blocks and some interstitial bright R bands (7q13,7q15, 7q22, 7q24). Besides of telomeric block on chromosome 16 there was also bright q23 band. The X chromosome have not found any sharp THA staining. Chromosome Y revealed only T block coincided with R+ region of short arm.

The genomic share of centromeric, telomeric and interstitial THA stained bands was correspondingly 11.5 p. cent, 3.3 p. cent and 1.5 p. cent. We tried to analyze relationship between T blocks and R or G bands (Di Berardino *et al.*, 1990).

According to our results all telomeric and interstitial T bands conformed to R blocks. However, about 42 p. cent of such chromosomal regions revealed more complex origin because of the bands included simultaneusly R and G blocks (chromosome 4, 6, 8, 9, 10, 12, 14, 16, 18, 19, 20, 23, 24, 28).

The comparison of distribution T bands along human (Saccone *et al.*, 1992) and bovine chromosomes displayed some differences and common characters **(table I)**.

The bovine chromosomes have not practically revealed interstitial THA pattern of staining. Majority of T blocks

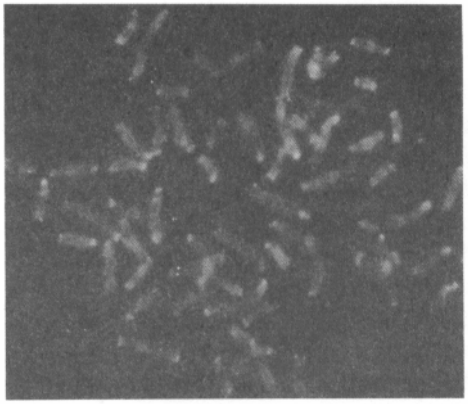

Figure 2. *FISH on* Bos taurus *chromosomes using the probe containing telomeric repeat (TTAGGG)n.* (FISH en los cromosomas de *Bos taurus* usando una sonda que contiene la repetición telomérica (TTAGGG)n)

from both apecies coincided with R bands. The proportion of telomeric plus interstitial ones was very like for both species. It was 9 p. cent for cattle and 15 p. cent for human.

Probably, the pericentromeric T blocks of *Bos taurus* were result of CG enrichment of bovine satellites DNA familles. It was the reason of preferential THA and CMA staining of such

TableI. *Cytogenetical properties of T bands from* Bos taurus *and* Homo sapiens *(Holmquist, 1990; Craig, and Bickmore, 1993) mitotic chromosomes.* (Propiedades citogenéticas de las bandas T de *Bos taurus* y *Homo sapiens* (Holmquist, 1990; Craig and Bicmore, 1993).

Species	Share of T bands in chromosomal set (p. cent)	Share of T bands, (p. cent) Centromeric	Interstitial	Telomeric
Human	15	-	35	65
Cattle	17	48	3	49

chromosomal regions. The molecular composition of T blocks coincided according to cytochemical criteria for both species. The results of THA banding, specific chromomycin A3 (CMA) staining, distribution of heavy H3 isochore were the same (Saccone *et al.*,1992; Bernardi, 1995). We have also found coincidence of bovine T blocks after THA and CMA differential staining. One of the possible reasons of appearance of bovine pericentromeric T bands may be also availability of small short arm of acrocenric autosomes we have used TTAGGG repeats *in situ* hybridization to verify such suggestion. The probe hybridized intensely to the pericentromeric and telomeric regions of all autosomes and telomeric parts of sex chromosomes. Interstitial hybridization have not been noted **(figure 2)**.

Probably, it means only small parts of bovine pericentromeric bright THA staining pattern correspond to human T bands. There is opinion the most of mammals have got such blocks only in telomeric positions (Dutrillaux, 1979; Ambros and Sumner, 1987).

We tried to compare the distribution of bovine T, R and G bands and genes which have been asigned (Eggen and Fries, 1995). Unfortunately, there are only 22 intrachromosomally localized genes for *Bos taurus*. Of these, 10 map to R bands, 7 map to G bands and 5 maps to T bands. So, it is not posible to indicate preferential localization of bovine genes in the T bands at least nowadays. Over 1000 human genes have been mapped to single band. 464, 336 and 200 of genes were correspondingly mapped to T, R and G bands (Holmquist, 1990; Craig and Bickmore, 1993; Bernardi, 1995).

The data also raise the question of why T bands are often found at telomeres. It is interesting to note that although it has been postulated that all T bands may have telomeric origins. T bands-syntenic regions in the mouse are mostly nontelomeric (Craig and Bickmore, 1993).

So, it is necesary to verify conspicuous properties of G, T, R bands developed only for human genome. The experimental material for other species will be able to accumulate and take into consideration.

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