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Chromosome localization of the 31 type I Texas bovine markers in sheep and goat chromosomes by comparative FISH-mapping and R-banding

G. P. Di Meo*, A. Perucatti*, M. Gautier[†], H. Hayes[†], D. Incarnato*, A. Eggen[†] and L. Iannuzzi*

*National Research Council (CNR), ISPAAM, Laboratory of Animal Cytogenetics and Gene Mapping, Naples, Italy. [†]Laboratory of Biochemical Genetics and Cytogenetics, Department of Animal Genetics, INRA, Jouy-en-Josas, France

Summary

Bovine BAC clones containing the 31 genes, referred to as the Texas markers used earlier to definitively assign the 31 bovine syntenic groups (U) to cattle chromosomes, were mapped by fluorescent *in situ* hybridization to sheep and goat R-banded chromosomes according to ISCNDB2000. All 31 markers were localized on homoeologous chromosomes and chromosome bands of the two species in agreement with previous localizations obtained both in cattle and river buffalo, definitively confirming chromosome homoeologies between *Caprinae* and *Bovinae*. In addition, we have extended physical maps of sheep and goat as 11 genes (*HSD3B1*, *INHBA*, *CSN10*, *IGF2R*, *PIGR*, *MAP1B*, *DSC1*, *ELN*, *TNFRSF6*, *CGN1*, *IGF2*) and 14 genes (*SOD1*, *HSD3B1*, *CSN10*, *IGF2R*, *RB1*, *TG*, *PIGR*, *MAP1B*, *IGH@*, *LTF*, *DSC1*, *TNFRSF6*, *CGN1*, *IGF2*) were assigned for the first time to goat and sheep chromosomes, respectively.

Keywords cytogenetics, goat, nomenclature, sheep, type I marker.

Chromosome nomenclatures in bovids have often represented a problem for cytogeneticists due to the nature of the autosomes (all acrocentric in cattle and goat, almost all acrocentric in river buffalo and sheep), as well as to different banding techniques used by various laboratories. For this reason, official molecular markers were assigned to each cattle chromosome during the Texas Conference (1996). Another important step was achieved when the same BAC-clones containing the 31 markers referred as 'Texas markers' were mapped, by fluorescent *in situ* hybridization (FISH), to both G/Q and R-banded cattle chromosomes (Hayes *et al.* 2000). This study was the basis for the latest international conference (ISCNDB2000 2001) where new standard Q-, G-, and R-banded karyotypes were arranged for cattle, sheep and goat using only one common chromosome band nomenclature.

In this study, all the 31 Texas markers, earlier assigned to both cattle (Hayes *et al.* 2000) and river buffalo (Iannuzzi *et al.* 2001a) were FISH-mapped to both sheep and goat chromosomes, definitively confirming the chromosome

homoeologies among these four bovids and further extending their cytogenetic maps.

Concavalin A stimulated blood cell cultures were treated for late incorporation of both BrdU (20 µg/ml) and Hoechst 33258 (40 µg/ml) to obtain R-banded preparations.

Slides kept at -20 °C until use, were treated for FISH-technique as previously reported (Iannuzzi *et al.* 2000). The following biotinylated probes were used: 31 bovine BAC-clones each containing one of the 31 type I Texas markers characterized previously (Hayes *et al.* 2000; Gautier *et al.* 2001). Fifteen metaphases were studied for each probe and species and chromosome identification followed the standard karyotypes (Ansari *et al.* 1999; ISCNDB2000 2001). The frequency of FITC-signals (cells with spots on one chromosome or on both chromosomes) varied from 30% (*PGK1*) to 70% (*DEFB@*). These percentages were lower than those achieved in both cattle (Hayes *et al.* 2000) and river buffalo (Iannuzzi *et al.* 2001a), probably because of the bovine BAC-clones employed.

All 31 Texas markers mapped to homoeologous chromosomes and chromosome bands of the two species, as well as to homoeologous chromosomes and chromosome regions of sheep/goat and cattle, as reported in Table 1. This further supports the chromosome homoeology between these two species as well as between *Caprinae* and *Bovinae*. Indeed, goat and sheep chromosome localizations agree with those obtained in both cattle (Hayes *et al.* 2000) and river buffalo

Address for correspondence

L. Iannuzzi, CNR-ISPAAM, Laboratory of Animal Cytogenetics and Gene Mapping, Via Argine 1085, 80147 Naples, Italy.
E-mail: l.iannuzzi@iabbam.na.cnr.it

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Table 1 List of mapped loci and their localization on sheep (OAR) and goat (CHI) chromosomes. Comparisons with cattle chromosome (BTA) localizations (Texas Conference 1996; Hayes *et al.* 2000; ISCNDB2000 2001), relative syntenic group (U) and presumed localizations on the basis of Ansari *et al.* (1999) sheep standard are also reported.

Locus Symbol	Locus name	OAR	CHI	BTA	U	OAR, Ansari <i>et al.</i> 1999
<i>SOD1</i>	superoxide dismutase 1, soluble (amyotrophic lateral sclerosis 1 (adult))	1q12.2	1q12.2	1q12.2	U10	1q13
<i>VIL1</i>	villin 1	2q43	2q43	2q43	U17	2q43
<i>HSD3B1</i>	hydroxy-delta-5-steroid dehydrogenase, 3 beta- and steroid delta-isomerase 1	1p21	3q21	3q21	U6	1p21
<i>INHBA</i>	inhibin, beta A (activin A, activin AB alpha polypeptide)	4q26	4q26	4q26	U13	4q27
<i>IFNG</i>	interferon gamma type 2	3q23	5q23	5q23	U3	3q23
<i>CSN10</i>	casein, kappa	6q32	6q32	6q32	U15	6q32
<i>RASA1</i>	GTPase activating protein RAS p21	5q25.2	7q25.2	7q25.2	U22	5q32
<i>IFN1@</i>	interferon omega	2p15	8q15	8q15	U18	2p15
<i>IGF2R</i>	insulin-like growth factor 2 receptor	8q26	9q26	9q26	U2	8q26
<i>CYP19</i>	aromatase (cytochrome P450, subfamily XIX)	7q26	10q26	10q26	U5	7q34
<i>LGB</i>	lactoglobulin, beta	3p28	11q28	11q28	U16	3p27
<i>RB1</i>	retinoblastoma 1 (including osteosarcoma)	10q13	12q13	12q13	U27	10q13
<i>IL2RA</i>	Interleukine 2receptor alfa	13q13	13q13	13q13	U11	13q13
<i>TG</i>	thyroglobulin	9q13	14q13	14q13	U24	9q21
<i>FSHB</i>	follicle stimulating hormone, beta polypeptide	15q26	15q26	15q25-q26	U19	15q32
<i>PIGR</i>	polymeric immunoglobulin receptor	12q12	16q12	16q12	U1	12q12
<i>FGG</i>	fibrinogen, gamma polypeptide	17q13	17q13	17q13	U23	17q13
<i>GPI</i>	glucose phosphate isomerase	14q24	18q24	18q24	U9	14q24
<i>GH1</i>	growth hormone 1	11q22	19q22	19q22	U21	11q32
<i>MAP1B</i>	microtubule-associated protein 1B	16q13.1	20q13.1	20q13.1	U14	16q13
<i>IGH@</i>	immunoglobulin heavy locus	18q24	21q24	21q24	U4	18q24
<i>LTF</i>	lactotransferrin	19q24	22q24	22q24	U12	19q24-26
<i>BOLA-DYA</i>	major histocompatibility complex, class II,DY alpha	20q13	23q13	23q13	U20	20q13
<i>DSC1</i>	desmocollin 1	23q21	24q21	24q21	U28	23q13
<i>ELN</i>	elastin (supravalvular aortic stenosis, Williams-Beuren syndrome)	24q22	25q22	25q22	U8	24q17
<i>TNFRSF6</i>	tumor necrosis factor receptor superfamily, member 6	22q13	26q13	26q13	U26	22q13
<i>DEFB@</i>	beta defensin gene cluster	26q13	27q13	27q13	U25	26q13
<i>CGN1</i>	conglutinin 1	25q17	28q17	28q17	U29	25q21
<i>IGF2</i>	insulin-like growth factor 2 (somatomedin A)	21q24	29q24	29q24	U7	21q23
<i>PGK1</i>	phosphoglycerate kinase 1	Xq26	Xq28	Xq25	X	Xq23
<i>ZFY</i>	zinc finger protein Y-linked	Yp12	Yp12	Yp12	Y	Yp12

(Iannuzzi *et al.* 2001a). Special attention should be given to CHI25, CHI27, CHI28 and CHI29, homoeologous to OAR24, OAR26, OAR25 and OAR21, respectively. Indeed, these chromosomes have been often confused during standard karyotype constructions (reviewed in Iannuzzi *et al.* 1997). Details of their Q-, G- and R-banding patterns and chromosome differences have been reported in the ISCNDB2000 (2001). To confirm identification of these chromosomes in chromosome rearrangements or abnormalities, we suggest the use of FISH with official chromosome specific markers such as the Texas set of markers (Di Meo *et al.* 2000; Iannuzzi *et al.* 2001b, c, d). The presumed localizations of mapped loci made according to the sheep standard karyotype published by Ansari *et al.* (1999) are also reported in Table 1 (last column). The differences in band numbers are due essentially to a different numbering of landmarks and, in a few cases, to subdivision of bands.

The 31 type I markers assigned to sheep and goat in the present study extend the physical map of these two important

species. Indeed, 11 genes (*HSD3B1*, *INHBA*, *CSN10*, *IGF2R*, *PIGR*, *MAP1B*, *DSC1*, *ELN*, *TNFRSF6*, *CGN1*, *IGF2*) and 14 genes (*SOD1*, *HSD3B1*, *CSN10*, *IGF2R*, *RB1*, *TG*, *PIGR*, *MAP1B*, *IGH@*, *LTF*, *DSC1*, *TNFRSF6*, *CGN1*, *IGF2*) were localized for the first time on goat and sheep chromosomes, respectively (GoatBase: <http://locus.jouy.inra.fr/cgi-bin/lgbc/mapping/common/main.pl?BASE=goat>; Sheep Base: <http://www.thearkdb.org/>, November 2002). Moreover, the position of some loci previously mapped in these species has been revised according to both present data and the latest chromosome nomenclature (ISCNDB2000 2001). Indeed, *VIL1* maps to OAR2q43 and CHI2q43 (instead of OAR2q33 and CHI2q33), *RASA1* maps to OAR5q25.2 (instead of OAR5q21.4), *IFN1@* maps to CHI8q15 (instead of CHI8q22), *CYP19* maps to OAR7q26 (instead of OAR7q24-31), *LGB* maps to OAR3p28 (instead of OAR3p27), *GPI* maps to OAR14q24 and CHI18q24 (instead of OAR14q23-26 and CHI18q22), *ELN* maps to OAR24q22 (instead of OAR24q16-19).

In conclusion, the same BAC-clones containing the 31 type I Texas markers, previously localized in both cattle and river buffalo, were FISH-mapped to sheep and goat chromosomes definitively confirming chromosome homologies among sheep, goat, cattle and river buffalo and further supporting the common origin among the four species. To date, the only small difference between *Bovinae* (cattle and river buffalo) and *Caprinae* (goat and sheep) autosomes, revealed by comparative FISH-mapping data, concerns the *Bovinae* and *Caprinae* chromosomes 9 and 14 which differ in their pericentromeric region. This rearrangement has been explained by a simple chromosome translocation between ancestral forms of the two chromosomes (Iannuzzi *et al.* 2001e). More complex are the rearrangements (transpositions) that modify the size, shape, chromosome banding and gene order of the X-chromosomes in these bovids (Piumi *et al.* 1998; Iannuzzi *et al.* 2000).

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