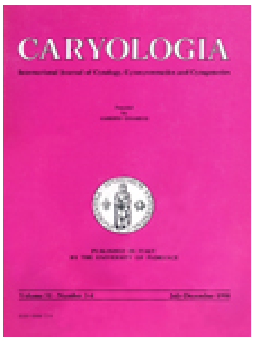

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A genetic physical map in river buffalo (*Bubalus bubalis*, 2*n*=50)

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SUMMARY — This study reports the first entire genetic physical map in river buffalo chromosomes by ISH-techniques. Thirty-six loci (mostly mapped by FISH), of which 19 were expressed genes and 17 were DNA-segments (cosmid derived-microsatellites), were localized in specific chromosomes. At least one molecular marker from all 31 bovine syntenic groups was assigned to each river buffalo chromosome. This allowed tentative assignment of all bovine syntetic groups to river buffalo chromosomes, also in consideration of high degree of chromosome banding homologies between the two species and that all mapped loci in river buffalo chromosomes were at the same cattle homoeologous chromosomes and chromosome bands.
By adding markers from bovine U1, U3, U7, U10, U13, U16, U17, U25, U28 and U29 assigned to the river buffalo genome by the somatic cell hybrid technique, a total of 54 markers is assigned to river buffalo genome.

Key words: river buffalo, gene mapping, chromosomes, *in situ* hybridization

INTRODUCTION

Of the 150 million buffaloes raised throughout the world, about 120 million are Asiatic water buffaloes *(Bubalus bubalis)* and the rest are African buffaloes *(Syncerus caffer)* (SHALASH 1991). Of the former, two types of buffaloes are known, the river buffalo (2*n*=50) and the swamp buffalo (2*n*=48), their karyotypes being differentiated by a tandem fusion translocation between river buffalo chromosomes (BBU) 4p and 9 originating the large chromosome 1 in swamp buffalo with a reduction in the diploid number from 50 to 48 (DI BERARDINO and IANNUZZI 1981; CHOWDHARY *et al.* 1989).

Crosses between these two types have been performed, especially to increase the milk production in the swamp type. Also in African buffaloes two types are known: the *Syncerus caffer nanus* (2*n*=54) and the *Syncerus caffer* (2*n*=52) (BUCKLAND and EVANS 1978). No common biarmed pair has been found between the Asiatic and African buffaloes, confirming a different evolution between the two types of buffaloes (IANNUZZI *et al.* 1983).

Standard G-, Q- and R-banded karyotypes are available in river buffalo (CSKBB 1994) and several genes have been assigned by both somatic cell hybrid (EL NAHAS *et al.* 1993, 1996a, 1996b) and ISH techniques (Hassaname *et al.* 1993, 1994; IANNUZZI *el al.* 1993a, b, 1997a, b, c) but a complete genetic physical map is still lacking in this important species.

In this study I summarized all the data available in the river buffalo genetic map, primarily referring to ISH (mostly by FISH) technique, by showing the first entire physical map on R-banded standard ideogram.

MATERIALS AND METHODS

Blood culture, slide preparations, R-banding techniques, bovine probe preparations and denaturation, *in situ* hybridization, signal detection, microscope observation, photographs, image acquisition and processing were reported in the original studies (Table 1). River buffalo chromosome banding nomenclature followed the standard karyotype (CSKBB 1994), while all the bovine markers assigned in river buffalo chromosomes were in agreement with the available cattle data (EGGEN and FRIES 1995; TEXAS NOMENCLATURE 1996).

RESULTS AND DISCUSSION

Table 1 reports all loci mapped in river buffalo with relative bovine syntenic groups and references, while figure 1 shows the river buffalo R-banded standard ideogram with the exact localization of mapped loci with a complete list of cattle syntenic groups (expressed genes only, EGGEN and FRIES 1995) indirectly assigned to specific river buffalo chromosomes. As shown, at least one molecular marker from all 31 bovine syntenic groups has been assigned to each river buffalo chromosome. Furthermore, with the exception of *IFNG* (BBU4q), *IGHG* (BBU20) and the cosmid clOBT945 (BBUX), all the remaining loci were assigned to single chromosome bands which were found to be almost all R-positive. With few exceptions due to banding pattern resolution achieved during *in situ* procedures, all mapped loci were localized at the same homoeologous chromosomes and chromosome bands between cattle and river buffalo, confirming that chromosome banding similarity is highly indicative for genetic homology. Also the homoeologous cattle chromosomes (BTA) are indicated ac[illegible]ng to the TEXAS NOMENCLATURE (1996). Only for BBU1p and BBU24 bo[illegible] A25 and BTA27 were indicated as homoeologues for the discrepancies betw[illegible]n the TEXAS NOMENCLATURE (1996) and both river buffalo stand[illegible]ryot[illegible]e (CSKBB 1994) and recent FISH-mapping data (IANNUZZI *et a*[illegible] /d) which followed the ISCNDA89 (1990) system.

Table 1 - Loci physically mapped in river buffalo chromosomes by *in situ* hybridization (mostly by FISH).

Locus name and symbol	Chromosome localization	Bovine syntenic group *	References
Uridine monoph. syntase *(UMPS)*	1q31	U10	Iannuzzi *et al.* 1994a
Beta-defensin *(DEFB@)*	1p12	U25	Iannuzzi *et al.* 1996c
Villin *(VIL)*	2q33	U17	Iannuzzi *et al.* 1997a
Major hystoc. complex *(MHC-Bubu)*	2p22	U20	Iannuzzi *et al.* 1993a
Omega interferon *(IFNW)*	3q15	U3	Iannuzzi *et al.* 1993b
Trophoblast interferon *(IFNT)*	3q15	U3	Iannuzzi *et al.* 1993b
IDVGA47 (DNA segment)	3p22	U21	Iannuzzi *et al.* 1997c
Lysozyme *(LZY)*	4q23	U3	Iannuzzi *et al.* 1993c
Gamma interferon *(IFNG)*	4q23>26	U3	Hassaname *et al.* 1994
Conglutinin *(CGN1)*	4p16	U29	Iannuzzi *et al.* 1994b
IDVGA49 (DNA segment)	5q21	U1	Iannuzzi *et al.* 1997c
IDVGA7 (DNA segment)	5p19	U7	Iannuzzi *et al.* 1997c
IDVGA53 (DNA segment)	6q15	U6	Iannuzzi *et al.* 1997d
Alpha-S2-casein *(CASN1S2)*	7q32	U15	Iannuzzi *et al.* 1996b
IDVGA61 (DNA segment)	8q34	U13	Iannuzzi *et al.* 1997d
Elogation factor 2 *(EEF2)*	9q15	U22	Iannuzzi *et al.* 1997b
Connexin *(GJA1)*	10q17	U2	Iannuzzi *et al.* 1998b
JAB10 (DNA segment)	11q13	U5	Iannuzzi *et al.* 1998b
1>3-galactosyltransferasi *(GGTA1)*	12q36	U16	Iannuzzi *et al.* 1997b
IDVGA41 (DNA segment)	13q15	U27	Iannuzzi *et al.* 1997d
Prion protein *(PRNP)*	14q15	U11	Iannuzzi *et al.* 1998a
IDVGA76 (DNA segment)	15q15	U24	Iannuzzi *et al.* 1998b
IDVGA32 (DNA segment)	16q25	U19	Iannuzzi *et al.* 1997d
Zinc finger protein *(ZNF164)*	17q24	U23	Iannuzzi *et al.* 1997e
Zinc finger protein *(X81804)*	18q24	U9	Iannuzzi *et al.* 1997e
Microtubule ass. protein *(MAP1B)*	19q13	U14	Iannuzzi *et al.* 1998b
Immun. gam. heavy chain *(IGHG)*	20q23>25	U4	Hassaname *et al.* 1993
Cathalecidins *(CATHL@)*	21q24	U12	Iannuzzi *et al.* 1998b
COSAE7 (DNA segment)	22q24	U28	Iannuzzi *et al.* 1998b
IDVGA59 (DNA segment)	23q22	U26	Iannuzzi *et al.* 1997d
IDVGA71 (DNA segment)	24q13	U8	Iannuzzi *et al.* 1997d
IDVGA82 (DNA segment)	Xq44	X	Iannuzzi *et al.* 1998b
clOBT314 (DNA segment)	Xq13	X	Prakash *et al.* 1997
clOBT945 (DNA segment)	Xq34-35	X	Prakash *et al.* 1997
clOBT1489 (DNA segment)	Xq47	X	Prakash *et al.* 1997
IDVGA50 (DNA segment)	Y	Y	Iannuzzi *et al.* 1998b

* For the complete list of expressed genes and DNA segments mapped in each bovine syntenic group, see Eggen and Fries (1995) and Ferretti *et al.* (1997).

The markers assigned to the biarmed pairs allowed the following genetic association between bovine syntenic groups to be established: U10/U25 in BBU1, U17/U20 in BBU2, U18/U21 in BBU3, U3/U29 in BBU4 and U1/U7 in BBU5.

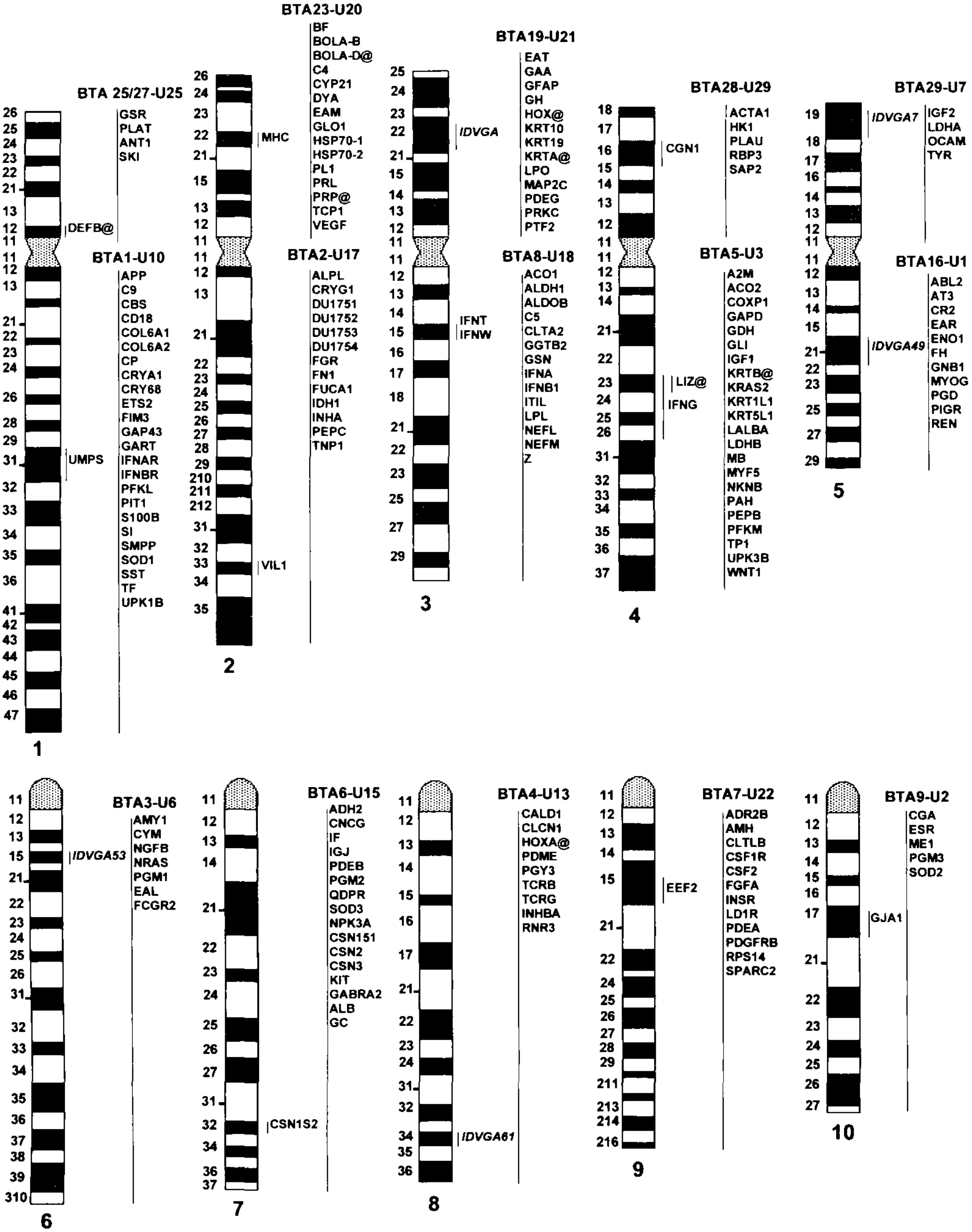
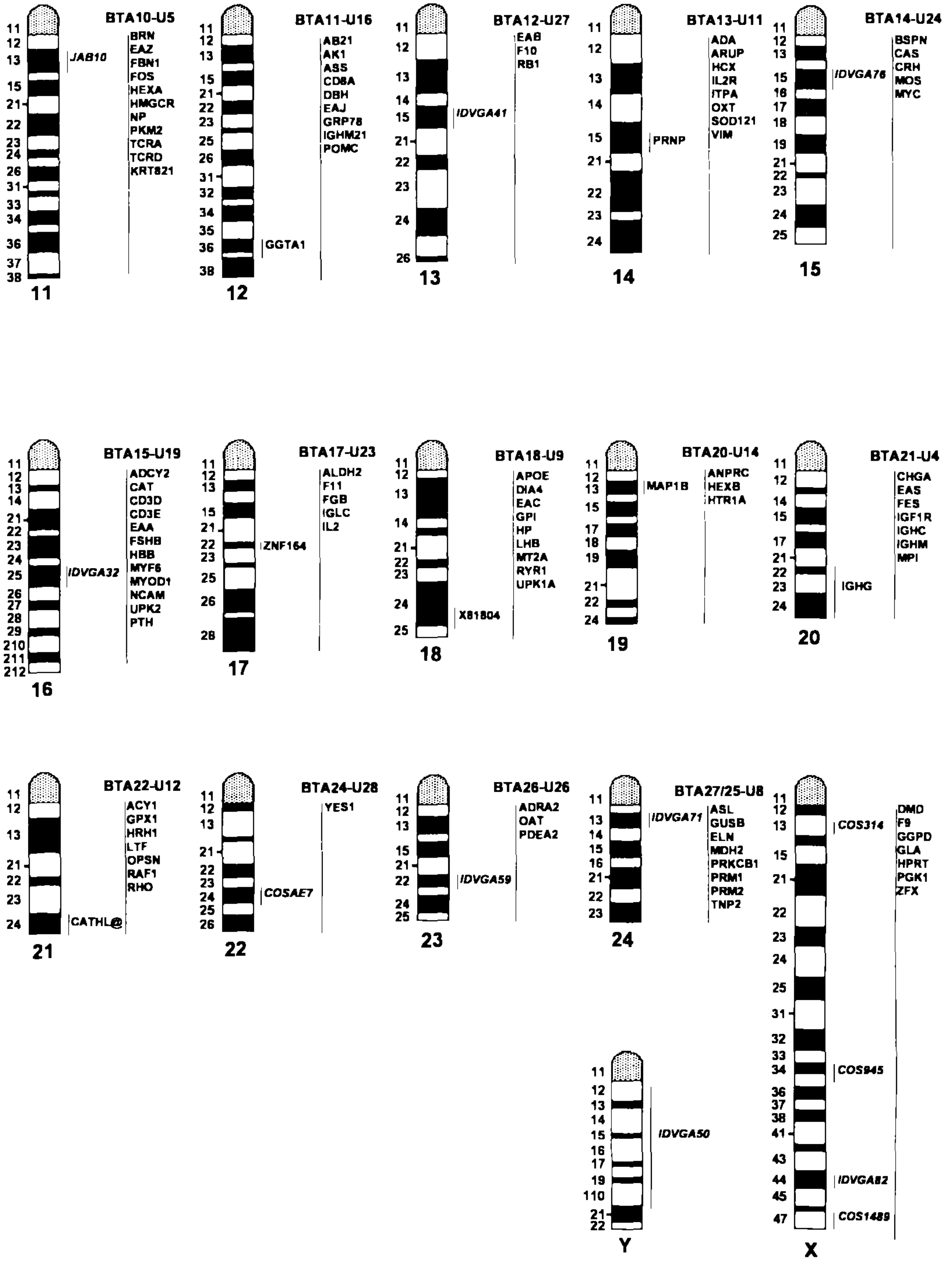


Fig. 1. — River buffalo R-banded standard ideogram with the ISH-mapped loci (mostly by FISH). The homoeologous cattle chromosomes (BTA) and relative bovine syntenic groups indirectly assigned (only expressed gene loci, Eggen and Fries 1995) are also shown. The DNA segments (mostly cosmids derived microsatellites) are reported in italics.


BTA10-U5
JAB10
BRN
EAZ
FBN1
FOS
HEXA
HMGCR
NP
PKM2
TCRA
TCRD
KRT821
11
BTA11-U16
AB21
AK1
ASS
CD8A
DBH
EAJ
GRP78
IGHM21
POMC
GGTA1
12
BTA12-U27
IDVGA41
EAB
F10
RB1
13
BTA13-U11
PRNP
ADA
ARUP
HCX
IL2R
ITPA
OXT
SOD121
VIM
14
BTA14-U24
IDVGA76
BSPN
CAS
CRH
MOS
MYC
15
BTA15-U19
IDVGA32
ADCY2
CAT
CD3D
CD3E
EAA
FSHB
HBB
MYF6
MYOD1
NCAM
UPK2
PTH
16
BTA17-U23
ZNF164
ALDH2
F11
FGB
IGLC
IL2
17
BTA18-U9
X81804
APOE
DIA4
EAC
GPI
HP
LHB
MT2A
RYR1
UPK1A
18
BTA20-U14
MAP1B
ANPRC
HEXB
HTR1A
19
BTA21-U4
IGHG
CHGA
EAS
FES
IGF1R
IGHC
IGHM
MPI
20
BTA22-U12
CATHL@
ACY1
GPX1
HRH1
LTF
OPSN
RAF1
RHO
21
BTA24-U28
COSAE7
YES1
22
BTA26-U26
IDVGA59
ADRA2
OAT
PDEA2
23
BTA27/25-U8
IDVGA71
ASL
GUSB
ELN
MDH2
PRKCB1
PRM1
PRM2
TNP2
24
IDVGA50
Y
COS314
COS945
IDVGA82
COS1489
DMD
F9
GGPD
GLA
HPRT
PGK1
ZFX
X

The assignments of bovine molecular markers to BBU1p (U25), BBU4p (U29), BBU5p (U7) and BBU24 (U8) should allow us to easily resolve cattle nomenclature inconsistencies (GALLAGHER *et al.* 1993; IANNUZZI *et al.* 1994b; IANNUZZI and DI MEO 1995; IANNUZZI 1996; IANNUZZI *et al.* 1996c, 1997c, d). In fact, since high resolution G- and R-banded karyotypes are available in river buffalo (IANNUZZI *et al.* 1990a, b; CSKBB 1994), it will be very easy to check the G- and R-banding patterns in cattle (and related species) by using the river buffalo as marker chromosomes and the TEXAS NOMENCLATURE (1996) as reference point for molecular marker assignments. I should point out that BBU3p (BTA19/U21), BBU4p (BTA28/U29), BBU6 (BTA3/U6), BBU21 (BTA22/U12), BBU23 (BTA24/U26) and BBU24 (BTA27/U8) are nucleolus organizer chromosomes (IANNUZZI *et al.* 1996a). In particular, BBU24 (U8) (IANNUZZI *et al.* 1997d) is homoelogous to BTA27 or to BTA25 according to ISCNDA89 (1990) R-banded standard karyotype and TEXAS NOMENCLATURE (1996), respectively. Furthermore, the same U8 molecular marker assigned to BBU24 (IANNUZZI *et al.* 1997d) has been assigned to BTA27 (ISCNDA89 1990) or to BTA25 (TEXAS NOMENCLATURE 1996) by sequential FISH/RBA/Ag-NOR techniques (IANNUZZI 1998) confirming the homoeology between BBU24 and BTA27 (or BTA25) and that the bovine U8 chromosome is NOR-bearing.

The total number of loci assigned by ISH (mostly by FISH) is 36 and other ones from bovine syntenic groups U1, U3, U7, U10, U13, U16, U17, U25, U28 and U29 have been assigned to the river buffalo genome by somatic hybrid cell procedures (EL NAHAS *et al.* 1993, 1996a, 1996b), bringing to 54 the total number of loci assigned to this species. Future steps for the genetic improvement of this important species should include and increase in the number of loci assigned by FISH on each chromosome arm and linkage maps.

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