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# Cosmid-derived markers anchoring the bovine genetic map to the physical map

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**Abstract.** The mapping strategy for the bovine genome described in this paper uses large insert clones as a tool for physical mapping and as a source of highly polymorphic microsatellites for genetic typing, and was one objective of the BovMap Project funded by the European Union (UE). Eight-three cosmid and phage clones were characterized and used to physically anchor the linkage groups defining all the bovine autosomes and the X Chromosome (Chr). By combining physical and genetic mapping, clones described in this paper have led to the identification of the linkage groups corresponding to Chr 9, 12, 16, and 25. In addition, anchored loci from this study were used to orient the linkage groups corresponding to Chr 3, 7, 8, 9, 13, 16, 18, 19, and 28 as identified in previously published maps. Comparison of the estimated size of the physical and linkage maps suggests that the genetic length of the bovine genome may be around 4000 cM.

### Introduction

Loci anchoring the physical and genetic maps of a species are a very important resource in the development of genome mapping strategies, both in building comprehensive maps and for the eventual use of those maps. The benefits that are derived from integrated physical and linkage maps are numerous, as exemplified by the work on humans. Several aspects of the latest human map (Hudson et al. 1995) should be considered in establishing maps in other species. For example, it was developed with Sequence Tagged Sites (STSs; Olson et al. 1989) and incorporates most of the STSs and Expressed Sequence Tags (ESTs) previously placed on human linkage maps, the latter being crucial for the postitional cloning of disease genes. The amalgamation of physical and genetic maps allows a direct estimation of the distance between the ordered STSs, thereby providing the best coverage of the human genome so far.

Genome maps have been developed over the last few years for

several domesticated animal species (Bumstead and Palyga 1992; Fries et al. 1993; Barendse et al. 1994; Bishop et al. 1994; Levin et al. 1994; Archibald et al. 1995; Crawford et al. 1995), the bovine genome map being the most extensive, ranking fourth after the maps available for human, mouse, and rat. An updated bovine linkage map currently comprises almost 800 markers, including 150 type I markers or genes (O'Brien et al. 1993), and is predominantly made up of type II STS markers (W. Barendse, personal communication). The development of a physical map of the bovine genome has lagged behind mainly because of the difficulty in identifying individual bovine chromosomes. Initially, physical mapping data were based on conserved type I markers mapped to syntenic groups with somatic hybrid cell panels (Womack and Moll 1986; Fries et al. 1993). However, direct physical mapping approaches, like fluorescence in situ hybridization (FISH), have recently become available, so that now all the synteny groups have been assigned to chromosomes (Mezzelani et al. 1994; Masabanda et al. 1996). This has been achieved in most cases by combining the localization of large-insert clones to chromosomes by FISH, with the PCR amplification of microsatellites derived from the clones on hybrid cell lines.

Obtaining highly polymorphic markers from cosmid and phage clones is a very efficient way of placing markers on both the genetic and physical maps (Solinas Toldo et al. 1993; Ellegren et al. 1994a; Eggen and Fries 1995; Mezzelani et al. 1995; Thieven et al. 1996). This approach was adopted by the BovMap group as the tool of choice to provide physical anchorage to the developing bovine linkage map, and the results are presented in this paper. To achieve the mapping goals, it was crucial importance to use a common set of animals on which to type the markers, and the work was carried out using the International Bovine Reference Family Panel (IBRP; Barendse et al. 1996).

We report here on the physical mapping of 83 cosmid and phage-derived bovine microsatellite marker loci that were simultaneously placed on the bovine linkage map. Thus, the correspondence between linkage groups, synteny groups, and chromosomes is directly confirmed. In addition, some linkage groups have been oriented compared with previously published maps (Barendse et al. 1994; Bishop et al. 1994), so that now all the linkage groups are oriented with respect to the cytogenetic map.

Finally, the set of markers employed allows the estimation of the correspondence between the physical map and the genetic map; this helps to validate the estimates of genome size inferred from chiasmata counts. Estimating genome size has gained a special importance after the finding that the mouse and the porcine genomes are about 2000 centiMorgans (cM) long, which is significantly shorter than that of human despite having the same physical size (Davisson and Roderick 1989; Ellegren et al. 1994b). Our data suggest the converse may be true in cattle, where the length of the genome may be close to that of human, and possibly larger.

### Materials and methods

Isolation and physical mapping of cosmids and phages. Cosmids and phage-containing microsatellites were identified with poly d(AC) probes. For the majority of clones (that is, IDVGA and ETH clones), QFQ-banded chromosome preparations were subjected to competitive in situ suppression hybridization (Lichter et al. 1990) and then hybridzed with biotinylated probes. FITC signal detection was performed with a computercontrolled CCD camera device (Photometrics, Tucson, Ariz.), as described by Solinas Toldo et al. (1993) and Mezzelani et al. (1995). Chromosomal band assignments were obtained by determining the fractional length (FLcen) with respect to the proximal border of the chromosome and superimposing the value to the idiogram of the standard chromosomes (Popescu et al. 1996). Details of the physical localization for the other clones can be found in the references in Table 1. Sequencing and development of the microsatellite markers was performed at a later stage, when the chromosomal location of clones was known. From nearly 200 large insert clones that were physically mapped, 83 were eventually selected and the embedded microsatellite regions sequenced either by subcloning or directly from the cosmid (Ferretti et al. 1994) or phage templates (Thieven et al. 1995), for use as genetic markers.

Markers and polymorphisms. For all but five loci, polymorphism in the length of the dinucleotide microsatellite repeat was used for genetic mapping by analysis of the PCR products generated with the primers described in Table I on denaturing acrylamide gel electrophoresis. For five loci that showed low heterozygosity, mapping was by single strand conformational polymorphisms (SSCP) described by Williams and associates (1996b). Table 1 summarizes the details of all the markers that are part of the BovMap database maintained at INRA, which is accessible on the Internet at the address: http://locus.jouy.inra.fr/cgi-bin/bovmap/. A database of D numbers, identifying the anonymous DNA loci, is maintained by R. Fries.

Typing and linkage analysis. A common set of 15 full sib families (330 animals), which are part of the International Bovine Reference Family Panel (IBRP; Barendse et al. 1996), was used for the genetic mapping of the markers. The data are collated in the Cattle Genotypic Database (CGD), managed by W. Barendse. Intervals between markers were estimated by two-point linkage analysis with the CRI-MAP program (Green et al. 1990). The lod scores and recombination fraction data can be obtained from barendse@magic.tap.csiro.au.

#### Results

Isolation and mapping of microsatellites. The goal of the project was to map large insert clones harboring microsatellite sequences by FISH and to use the microsatellite regions to locate the clones on the genetic linkage map. In the course of the project, 185 cosmid and 10 lambda phage clones were placed on chromosomes by FISH, and microsatellite markers were developed for a subset of 83 of these clones that were suitable distributed across the bovine karyotype. These markers are shown in Fig. 1 and Table 1. Fifteen chromosomes have at least three markers, six chromosomes have two markers, and nine have a single marker. Of the chromosomes with only one marker, only Chr 22 had additional FISH-mapped clones at positions distinct from the one that was genetically mapped, but for which it was not possible to develop a polymorphic marker. On the remaining chromosomes, the additional clones available mapped to the same chromosomal region as the previously genetically mapped marker.

The distribution of the clones described in Fig. 1 is not biased towards the telomeric regions of the chromosomes. However, the overall distribution of the complete set of FISH-mapped clones, that is, the 185 cosmids and 10 lambda phages, suggests a tendency for microsatellite sequences to be more frequent towards the telomeres (54 clones mapped to telomeric or subtelomeric bands out of 195); in addition, more microsatellite-containing clones were obtained from Chrs 15, 16, and 19 than expected compared with other chromosomes (data not shown).

Linkage. The STS markers developed from the large insert clones (Table 1) were placed on the linkage map by typing the animals of the IBRP panel. The results are shown in Fig. 1 (N.B., some markers for Chr 15 are not shown, for clarity). Physical and linkage mapping of the clones has been essential to define the orientation of various linkage groups in relation to chromosomes in the continuous process of updating the bovine genetic map. Thus, the linkage groups for the following chromosomes were anchored and in some cases oriented by use of the cosmids and the embedded microsatellite markers described in this paper: Chr 3, D3S24; Chr 4, D4S32; Chr 7, D7S20; Chr 8, D8S20; Chr 9, D9S14; Chr 10, D10S36; Chr 12, D12S17; Chr 13, D13S16; Chr 16, D16S5 and D16S23; Chr 18, D18S10; Chr 19, D19S4 and D19S18; Chr 28, D28S10. The physical assignment of marker D27S14 to 27q23 confirmed the recent identification of the last unassigned linkage group (Masabanda et al. 1996). Additionally, cosmid MAP1B, 20q14-q15, confirmed the orientation of the linkage group for Chr 20.

Finally, the following markers provided the first physical assignments that anchored the linkage groups corresponding to Chr 9, *D9S14*; Chr 12, *D12S17*; Chr 16, *D16S5*; Chr 29, *D29S2*, formerly Chr 25, and *D25S2* (ISCNDA 1989, 1990; Popescu et al. 1996).

Comparison of the physical and genetic maps. The markers defined intervals on 20 chromosomes as illustrated in Fig. 1. Each chromosome is represented with the idiogram of the corresponding G-banding pattern, in accordance to a recently agreed update of the ISCNDA standard (Popescu et al. 1996). All the idiograms are drawn to scale. The markers described in this study are indicated by the D-numbers flanking the idiograms, with the corresponding chromosomal assignments. The location of the markers on the genetic map is shown at the right of the idiograms, with the linkage group that defines each chromosome. Distances are in cM from the physically anchored microsatellite markers to the closest genes to bridge the gaps between the markers on the genetic map. Additional markers defining the ends of the linkage groups are included, where necessary. Thus, the genetic distances between the markers are obtained by adding the distances that separate all the markers for each chromsome. The complete linkage map will be published elsewhere (Barendse et al., 1996).

The intervals defined by the FISH-mapped anchored loci were used to evaluate the alignment of the physical and the genetic map. With respect to physical mapping, the distances between markers were evaluated by measuring the length spanning hybridization peaks, defined by cytogenetic band assignments (Lichter et al. 1990). In the case of assignments to more than one band, the central position was taken. The length of the interval was estimated as a percentage of the total karyotype. Markers separated by less than 1% of the genome were not considered. No data are presented for Chrs 13 or X, which only have two markers mapped at adjoining positions. In total, intervals could be considered for 20 chro-

### Table 1. Markers information

NMA222         D1542         [19]2         Clinic Ax16G A1CGACT         CA         D164         Ma223         D1540         [19]41           AUB6         D1540         11941         Clinic Ax16G A1CGACT         CA         CA         D1640         D2522         D26         D2524         X0941 / (Williams et al. 1996)           AUB6         D1540         11941         Clinic Ax16G A1CGACT         CGACT	Marker	D No.	FISH	Primers (5' to 3')	Repeat	Size (BP)	Acc. No./Reference
AB6         D1540         1q41         GCARDITICACCULUTION         CACOUNT         D4200         X58441 / (Williams et al. 1996a)           RV16         D2522         2q14-q21         CTTATIGATAGATTGATCGATCGATCGATCGATCGATCGATCGATC	INRA212 HAUT33	D1S42 D1S63	1q12 1q31	* CTGTCAATGGTATCGGACTCA	* (CA)11G(AC)2	200 208	a X89258 / (Thieven et al. 1996)
B0918         D2S32         24p1-42         ATTAXISAGANATICAT (Explanded)         SSCP         140         X33164 / (Williams et al. 1996)           IDVCAM0         D2566         2q42         TUTESATOCT (Explanded)         (C)TTAL         98         X7956 / (Williams et al. 1996)           IDVCAM7         D2519         2q45         TOTALSCI CONCOLLANCE         (C)TTALSCI CONCOLLANCE         (C)TTALSCI CONCOLLANCE           IDVCAM2         D257         2q45         TOTALSCI CONCOLLANCE         (C)TTALSCI CONCOLL	JAB6	D1S40	1q41	GCAAGCTGTCAGCCTGTTCA GAGACATGCATCCCAACCACCC CATCACCTACAACCATCCCATC	(AC)6AT(AC)12	242/248	X98441 / (Williams et al. 1996a)
IDVCASP         D2546         2q42         TOTICLE INSTITUTE ANALOGE CARACTELE         (GT)21         96         X7555 / (Nijma et al. 1996)           IDVCASP         D257         2q45         TAAGGGE CARACTELE (INSTITUTE)         (AC)10         122         Z27071 / (Permit et al. 1996)           IDVCASP         D257         2q45         GAAGGE CARACTELE (INSTITUTE)         (AC)10         123         Z37071 / (Permit et al. 1996)           IDVCASP         D3525         3q35         CAGGE CARACTELE CARACTELE (INSTITUTE)         (AC)11         211         X38070 / (Mezzelani et al. 1995)           IDVCASP         D3525         3q35         CAGGE CARACTELE CARACTELE (INSTITUTE)         (AC)11         212         X38072 / (Mezzelani et al. 1995)           IDVCASP         D3525         3q35         TECELET CARACTELES         (AC)21         202         X38052 / (Mezzelani et al. 1995)           IDVCASP         D3525         3q37         TECELET CARACTELES         (AC)21         138         X38064 / (Mezzelani et al. 1995)           IDVCASP         D5530         5q25         TECELET CARACTELES         (AC)17         177         X38234 / (Thicese et al. 1996)           IDVCASP         D5530         5q33         ATECELET TARACTELES         (AC)17         157         X38234 / (Thicese et al. 1996)      <	RI918	D2S32	2q14-q21		SSCP	140	X93164 / (Williams et al. 1996b)
IDVGA37         D2S19         2q45         TAXAGE/AGE/CITAGE/IG         (AC)17(TC)10AT(AC)6         210         X83083 / Mezzelani et al. 1995)           IDVGA32         D2S7         2q45         GTAGCAAGE/AGE/CITAGE/GE         (AC)10         132         Z27071 / (Premeti et al. 1994)           IDVGA53         D3S29         3q21         GAGGAAGE/AGE/CITAGE/GE         (AC)11         221         X85073 / (Mezzelani et al. 1995)           IDVGA54         D3S24         3q35         ACATGE/CITAGE/CICC/CITAGE/GE         (AC)11         202         X89278 / (Mezzelani et al. 1996)           IMU731         D3S4         3q36         CATGE/CITAGE/GE         (AC)21         202         X89278 / (Mezzelani et al. 1996)           IMU731         D3S4         3q36         CATGE/CITAGE/GE         (AC)21         202         X89278 / (Mezzelani et al. 1996)           IMU731         D3S4         3q37         TGE/CITAGE/GE/GE/CITAGE/GE         (AC)21         124         Z2779 / (Gigen et al. 1996)           IMU731         D3S5         5q32         4q11         ATTGE/CITAGE/GE/CITAGE/GE/CITAGE/AGE/CITAGE/GE/CITAGE/AGE/CITAGE/GE/CITAGE/AGE/CITAGE/GE/CITAGE/AGE/CITAGE/GE/CITAGE/AGE/CITAGE/GE/CITAGE/AGE/CITAGE/GE/CITAGE/AGE/CITAGE/GE/CITAGE/AGE/CITAGE/GE/CITAGE/AGE/CITAGE/GE/CITAGE/GE/CITAGE/AGE/CITAGE/GE/CITAGE/GE/CITAGE/GE/CITAGE/GE/CITAGE/GE/CITAGE/GE/CITAGE/GE/CITAGE/GE/CITAGE/GE/CITAGE/GE/CITAGE/GE/CITAGE/GE/CITAGE/GE/CITAGE	IDVGA80	D2S46	2q42	TCTTGGATGTCTGACAAGATGC	(GT)21	98	X97565 / (Nijman et al. 1996)
IDVGA2         DS7         2q.45         CTASCA ADDRAGES         (AC)10         132         Z27071 / Ferrenti et al. 1996)           IDVGA35         DS72         3q.21         ACCEGACCCTCCGTCAGC         (AC)11         210         X85097 / (Mazzelini et al. 1996)           IDVGA35         DS725         3q.35         AGT CCCCTCAGCACAGC         (AC)11         210         X85092 / (Mazzelini et al. 1996)           IDVGA35         DS725         3q.35         ACCEGACCTCCCAGACC         (AC)21         202         X85092 / (Mazzelini et al. 1996)           IAM739         DS525         3q.36         TTCCCTCAGACCCAGATC         (AC)21         180         X73947 / (Fegen et al. 1996)           IDVCA31         DS52         4q.31         TTCCCTCAGCCAGATT         (AC)9         138         X85046 / (Mazzelini et al. 1995)           IDVCA31         DS53         5q.35         GTTAGACTCOTTTECCA         (AC)17         17         X85086 / (Mazzelini et al. 1995)           IDVCA31         DS53         5q.35         GTTAGACTCOTTTECCA         (AC)17         17         X85047 / (Mazzelini et al. 1996)           IDVCA31         DS53         5q.35         GTTAGACTCOTTTECCAC         (AC)17         17         X89254 / (Mazzelini et al. 1996)           IDVCA34         DS53         5q.35	IDVGA37	D2S19	2q45	TAACAGGACAAGTCTTCAGGTG	(AC)17(TC)10AT(AC)6	210	X85053 / Mezzelani et al. 1995)
IDVGA53         DSS29         Sq21         ACCEGACCECTCGETCACTE. CACEGACAGGGAAAATTACAC         CAC)11         211         X85070 / (Mezzelini et al. 1995)           IDVGA53         DSS25         Sq35         AASTGCCATCACCTCCCCTTCACC         (CC)21         202         X85052 / (Mezzelini et al. 1995)           IAUT31         DSS41         Sq35         CACEGACTCCCCCTTCACCC         (CC)21         202         X85052 / (Mezzelini et al. 1996)           IAUT31         DSS41         Sq37         TGCTCACTCACAGACACACATEC         (AC)21         202         X85062 / (Mezzelini et al. 1995)           IDVCA27         DSS3         Sq37         TGCTCACTCACAGACACACATEC         (AC)21         138         X85064 / (Mezzelini et al. 1995)           IDVCA27         DSS3         Sq35         GTCCACACACACACACACACACACACACACACACACACAC	IDVGA2	D2S7	2q45	GTAGACAAGGAAGCCGCTGAGG	(AC)10	132	Z27071 / (Ferretti et al. 1994)
IDVGA15         D3525         D3535         D3536         <	IDVGA53	D3S29	3q21	ACGGGACGCCTCGGTCAGTG	(AC)11	221	X85070 / (Mezzelani et al. 1995)
IAUT31         D3541         3q36         CALL 12 THE INCLUMARAN         CACUST CONSTRUCT         CACUST CON	IDVGA35	D3S25	3q35	AAGTGCCATCACCTCCCCATTC	(TG)20CG(TG)6(AG)6	233	X85052/(Mezzelani et al. 1995)
NRAL197         D3526         3q.36         TEGRAFAGE TEGRAFICE AGREGITING TO GALACETCARGED TITES         (AC)23         180         X73947 / (Eggen et al. 1996)           IDVCA27         D3524         3q.37         TEGRAFICE AGREGITING TO GALACETCARGED TITES         (AC)9         138         X55066 / (Mczzchani et al. 1995)           IDVCA51         D4532         4q.11         ATECCTFCARGED TITES         (AC)11         174         X55066 / (Mczzchani et al. 1993)           IATTO         D553         5q.32         GETTRAGECT TATTAGETCTARTAGETCARGED TITES         (AC)17         157         X5924 / (Thieven et al. 1996)           IATTO         D553         5q.33 - q4         GETCTECATACTTGEGCACE         (AC)17         157         X29243 / (Williams et al. 1996)           IATTO         D552         5q.35         CECLACARGECT CICCATACT         (AC)11         203         Z27075 / (Ferretti et al. 1996)           IDVCA60         D552         5q.35         CECLACARGECT CICCA         (TO)10         204         Z67734 / (Kihn et al. 1996)           IDVCA60         D552         5q.35         GETCECTEGCTTATACGEC         (AC)11         203         Z27075 / (Ferretti et al. 1996)           IDVCA60         D552         5q.35         GETCECTEGCTTATACGEC         (CA)16         177         Z2743 / (Kihn et al. 1996)	HAUT31	D3S41	3q36	CAGGTATCGGTGGTGAGAAT	(AC)21	202	X89258 / (Thieven et al. 1996)
IDVGA27         D3S24         3q37         TGSTLACK LIGKARGE (ALC) TGSTLACK LIGKARGE (ALC) ATGCTLACK LIGKARGE (ALC) ATGCT	INRA197	D3S26	3q36	AGCATCAGCCTCAGAAGTGG	(AC)23	180	X73947 / (Eggen et al. 1996)
IDVGAS1         D4S32         4q31         ATGC/TGT/ATCC/ATGC/ATT         (AC)11         174         X85068 / (Mczzelni et al. 1995)           ETH10         DSS3         Sq25         CTECTGATGT/ATGC/ATA         (CA)19         214         Z22739 / (Solinas Toklo et al. 1993)           HAU729         DSS49         Sq33-q34         ATGC/ATGC/ATTTGGC/ACT         (CA)17         157         X89254 / (Thieven et al. 1996)           JAB2         DSS30         Sq33-q34         GGT/ATTTGGC/ACT         (CA)17         157         X89254 / (Thieven et al. 1996)           JAB2         DSS20         Sq33-q34         GGT/ATTTGGC/ACT         (CA)16         177         Z22743 / (Solinas Toklo et al. 1993)           JDVGA9         DSS27         Sq35         CCTC/ACAGT/CAAGCC/ATGC/AC         (CA)11         203         Z27075 / (Ferretti et al. 1994)           JDVGA9         DSS24         6(12-4)         CACAGACT/CAAGCC/ATGC/AC         (AC)20         187         Z3084 / (Harlzins et al. 1996)           JDVCA65         DSS29         6q22         ACTACACACT/AAGCC/ATGC/AC         (AC)20         187         Z3084 / (Marlzins et al. 1996)           JDVCA642         DTS20         7q14-q15         GGGACACCT/ATGC/AC         (CA)20         187         Z3084 / (Marlzins et al. 1996)           JDVCA642	IDVGA27	D3S24	3q37	AGAAACTGCAGGGIGIIGTGG TGGTCAGTCACAGAGAAGCACAG	(AC)9	138	X85046 / (Mezzelani et al. 1995)
ATTCCTTCATIGGTTAATGGTTA         (CA)19         214         222739 / (Solinas Toklo et al. 1993)           IAUT29         D5549         5q3-q4         ATTCCTAGCCACCTTTCTTCTC         (AC)17         157         X89254 / (Thieven et al. 1996)           JAB2         D5530         5q3-q4         ATTCCTAGCCACCTTGCTC         (AC)17         157         X89254 / (Thieven et al. 1996)           JAB2         D552         Sq35         GCACACGTCTGGCATACTTGGGCAC         (CA)16         177         Z22743 / (Solinas Toklo et al. 1993)           CATTCGACCTACGTGGCATGCCC         (CA)16         177         Z22743 / (Solinas Toklo et al. 1993)           IDVGA9         D552         Sq35         GCACACGGTTACTGGTGCACCTCCA         (CA)16         177         Z27034 / (Khn et al. 1996)           IDVGA9         D552         Gq12-q14         CCAGGGTTACTGGTGCACTCCCA         (CG)10         204         Z50734 / (Khn et al. 1996)           IDVGA9         D7520         7q14-q15         TGCAGTCACACTAGCA         ACQ20         187         X86815 / (Zhang et al. 1995)           IDVGA9         D7520         7q23-q24         AGACACATTGGCACGAGACTAA         (AC)19         230         Z27077 / (Ferretti et al. 1994)           IDVGA62         D8514         Sq15         CCAGTCTGGCACCACTTGGCA         (CA)19         230         Z	IDVGA51	D4S32	4q31	GAAAGCCTGGTACTCATGGAATA ATGGCAATATTTTGTTCTTTTC	(AC)11	174	X85068 / (Mezzelani et al. 1995)
LHUT29         DSS49         Sq3-q44         ASTECISACIENT CECCECT THE CECTURE CECCENT THE CECTURE CECTURE CECCECT THE CECTURE CECCENT THE CECTURE CECCENT CECCECT C	ETH10	D5S3	5q25	ATTCCTTGATGGTCTAATGGTTA GTTCAGGACTGGCCCTGCTAACA	(CA)19	214	Z22739 / (Solinas Toldo et al. 1993)
LAB2         DSS30         593-94         CETCICACATCETICAGACCLACTTI         (AC)9         224/228         X98435 /(Williams et al. 1996a)           ETH2         DSS2         593-94         CCALAGGGETTGGGACAGCC GCALAGGGETTGGGACAGCC CCALGGGATTGGCTACTTGGACGGCC CCALGGGATTGGCTACTTGGTAACA CTTCICTGCACAGGTCTGGACAGCC CCALGGGATTGGCACAGCC CCALGGGATTGGCACAGCC CTCICTGCACAGGTCTGGACAGCC CCALGGGATTGGCACACTCICTGG CCALGGGATGTGGACAGCC CTCICTGCACAGGTCTGGACGAGC CCALGGGATGTGGACGACACTCICTGG CCALGGGACC CCALGGGATGTGGACGACGCCCATCT CTG)10         204         Z50734 /(Kähn et al. 1996)           JDVGA6         DSS2         6q12-q1         CCALGGGATTGGCGACGAC CCALGGGACACTCICTGGCCACACTCI CTG)10         204         Z50734 /(Kähn et al. 1996)           JDVGA63         D6S29         6q22         TATATGAAAGTTAATAAAGC CTTCICGGCACACACCCCCCCCAGCC CCALGGACACTCICTCICAGGGACA         (AC)20         187         X86811 / (Zhang et al. 1995)           JDVGA62         DTS21         7q15         GGGATTGCCCAGGCCCATAC CCACCCACTCICTCICA CGCGTACACCACCCCCCCTCI CTG)15         264         X03162 /(Amatssindis et al. 1995)           JDVGA11         D8S20         8q14         CCTTCICGGCACCACCCCTCICT CTGCGGACCCACCTGCTGCA CCCCCTCGCGCCC CCCCCCCCCC	HAUT29	D5S49	5q33-q34	CCTCCAGCCCACTTTCTCTTCTC AGTGGTTAATTTAAGACTGTGC	(AC)17	157	X89254 / (Thieven et al. 1996)
ETH2         DSS2         Sq35         ECACAGGCT GRGCATEGCC CCATGGATTTCCCTCGCTAGCT CCATGGATTTCCCTGCTAGCT CCATGGATTTCCCTGCTAGCT CCATGGATTTCCCTGCTAGCT CCATGGATTTCCCTGCTAGCT CCATGGATTTCCCTGCTAGCT CCATGGATTTCCCTGCTAGCT CCATGGATTTCCCTGCTAGCT CCATGGATTTCCCTGCTAGCT CCATGGATTTCCCTGCTAGCT CCATGGATTTCCCTGCTAGCT CCATGGATTTCCCTGCTAGCT CCATGGATTTCCGCTGGCTAGCT CCATGGATTTCCGCTGGCT CCATGGATTTCCGCTGGCT CCATGGATTTCCGCTGGCT CCATGGATTTCCGCCGCCT CCGTGACGCCCTGCCT CCGTGACGCCCGTGCCG CCGTGACGCCCGTAGCGCGCT CCGTGACGCCCGTGCCGCGCC CCGTGACGCCGTGCCGCCGCCGCC CCGTGACGCCGTGCCGCCGCCGCCGCCGCC CCGTGACGCCGTGCCGT	JAB2	D5S30	5a33-a34	CTCTGCCCTCAGCCCACTTT GCTCTCACATACTTGGGCAC	(AC)9	224/228	X98435 / (Williams et al. 1996a)
LDW         CLATEGGATTIGECCTECTAGET         CONS         Cons         Const         Const <thcons< th=""> <thcons< th="">         Const</thcons<></thcons<>	ETH2	D5S2	5035	GGAATAGCTATCTGGAGAGGC	(CA)16	177	722743 / (Solinas Toldo et al. 1993)
DirectoryDeschLappoCTICTICACATICAACTICIAC(CC)TLos <td>IDVGAQ</td> <td>D5827</td> <td>5035</td> <td>CCATGGGATTTGCCCTGCTAGCT</td> <td>(40)11</td> <td>203</td> <td>727075 / (Ferretti et al. 1004)</td>	IDVGAQ	D5827	5035	CCATGGGATTTGCCCTGCTAGCT	(40)11	203	727075 / (Ferretti et al. 1004)
Parto         Data         Delta         Delta         CACGGACTEAAGGACTTAGEGAC         (10)         Delta         Extraction         Caccing           IDVGA65         D6S29         6q22         ACATATGAAAGGACTTAGEGAC         SSCP         23         Z73084 / (Hartizus et al. 1996)           IDVGA65         DFS30         7q14-q15         TGATTGCAAGTTAGEGAC         SSCP         23         Z73084 / (Hartizus et al. 1995)           IDVGA62         D7S21         7q15         GGGACACCCAGATAGEGA         (AC)20         187         X86815 / (Zhang et al. 1995)           IOBT30         D7S20         7q23-q24         AGCACCACTATCCACCAGEGATGTA         (AC)20         163         X86811 / (Zhang et al. 1995)           IOBT30         D7S20         7q23-q24         AGCACCACTATTCCATCTGCATGTG         (TG)15         264         X03162 / (Anastassiadis et al. 1996)           IDVGA12         D8S21         8q23         GAATCGAACCTTCATGTGCAGCACACTATT         (AC)20         200         X74174 / (Eggen et al. 1995)           IARIA         D9S1         9q25         TGGGTGGGGAGTCACTCAC         (AC)20         200         X74174 / (Eggen et al. 1996)           IARIA         D9S1         Sq275         GGTGGGGGTGACTCACCA         (AC)20         200         X741174 / (Eggen et al. 1996)           I	ERNS	D6524	6012 014		(TC)10	205	750724 / (Köhn et al. 1994)
DVGA90         D62.2         ACTAR IDAAAGT TA TAACT         SSCP         2.3         Z-/3084 (Tranticus et al. 1996)           IDVGA90         D7S30         7q14-q15         TGACAGTTCGAGGAGA         (AC)20         187         X86815 / (Zbang et al. 1995)           IDVGA62         D7S21         7q15         GGGTACACTCTGACCAGAGGTA         (TA)8(TG)10         163         X86811 / (Zbang et al. 1995)           IDVGA62         D7S20         7q23-q24         AGCACCACTATCCACCTGTCT         (TG)15         264         X03162 / (Anastassiadis et al. 1996)           IDVGA11         D8S20         8q14         CCTTCTGGCAACCACTATTGGAG         (AC)21         215         X85069 / Mezzelani et al. 1994)           IDVGA52         D8S21         8q23         GAATCGAACCTTTGGCA         (AC)21         215         X85069 / Mezzelani et al. 1994)           IDVGA52         D8S21         8q23         GAATCGAACCTTTCGCAC         (AC)20         200         X74174 / (Eggen et al. 1994)           IDVGA52         D8S51         10q15         GCTGAGCACAGGGTTGCTGGA         (AC)20         X8945 / (Williams et al. 1996)           IJAB10         D10S56         10q15         GCTGAGCACCGCTGTTACTTCG         (AC)21         15         X74201 / (Vaima et al. 1994)           JAGCTCACTTAGAGGCACAGGGTGCTGG         (AC)21         15	TDNG 4 (5	D0324	6412-414	ACACGACTGAAGCGACTTAGCAG	(10)10	204	Z30734 / (Kulli et al. 1990)
IDVCA30         D7830         7q14-q1S         IBA/RATCLARAGE LATAA         (AC)20         187         XB6815 (Zbang et al. 1995)           IDVCA62         D7521         7q15         GGCATCTTGGACCASATAGCA         (TA)8(TG)10         163         XB6811 / (Zbang et al. 1995)           IDBV70662         D7520         7q23-q24         AGCACCACTATCCATCTGTCTC         (TG)15         264         X03162 (Anastassiadis et al. 1996)           IDBV70611         D8520         8q14         CCTTCTGGCAACCACTATTGTG         (AC)19         230         Z27077 (Ferretii et al. 1994)           IDVCA52         D8521         8q23         GAATCGAACCACTATCA         (AC)21         215         X85069 / Mezzelani et al. 1995)           INRA144         D9514         9q25         TCGGTGTGGGAGGTGCTCGTACC         (AC)20         200         X74174 / (Eggen et al. 1994)           IJAB10         D10536         10q15         GCTGAACCACGAGGGTGTCGTG         (AC)22         213         X89255 / (Thieven et al. 1994)           IARA177         D11S17         11q16         CAGGTAGGAATATGAGGGTGT         (AC)21         158         X74201 / (Vaiman et al. 1994)           IAGGACTATCAAGGGTTCCTGC         (AC)21         195         X74201 / (Vaiman et al. 1994)         AGGACTCAAAACCCCACTTCTG           IARA177         D11S17	IDVGA05	D6529	6q22	TGATTCCAAGTTGAGGAGA	SSCP	223	2/3084/(Hariizius et al. 1996)
IDVGA62         DYS21         7q15         GelGALCTIGGALCAGAAGGIA         (TA)8(TC)10         163         X86811         / (Zhang et al. 1995)           IOBT930         D7S20         7q23-q24         ACCACCACTATCCATCTCTC         (TG)15         264         X03162 / (Anastassiadis et al. 1996)           IDVGA11         D8S20         8q14         CCTTCTGGCAACCACCTATTCATTGT         (AC)19         230         Z27077 / (Ferretti et al. 1994)           IDVGA52         D8S21         8q23         GAATCGAACCACCTTATCA         (AC)20         200         X74174 / (Eggen et al. 1994)           IDVGA55         D8S21         8q23         GAATCGAACCACCTTACCA         (AC)20         200         X74174 / (Eggen et al. 1994)           IACGTCACCTGACCACCACCGTGACCACCA         (AC)20         200         X74174 / (Eggen et al. 1994)         TGCTGGGGGTGCTGAC           JAB10         D10S36         10q15         GCTGACCTGCGCACCACCACCACCACCACCACCACCACCACCACCACC	IDVGA90	D7830	7q14-q15	GCCCGTACACCACATAGCA	(AC)20	187	X86815 / (Zhang et al. 1995)
IOBT930         DTS20         7q23-q24         ACCACCATCATCATCGATC         (TG)15         264         X03162 / (Anastassiadis et al. 1996)           IDVGA11         DSS20         8q14         CCTTCTGGCAACCATATTGT         (AC)19         230         Z27077 / (Ferretti et al. 1994)           IDVGA52         DSS21         8q23         GAATCGAACCTTGTCAC         (AC)21         215         X85069 / Mezzelani et al. 1994)           IDVGA52         DSS21         8q23         GAATCGAACCTTGCTGATGA         (AC)21         215         X85069 / Mezzelani et al. 1994)           IABCTCACACTTGCACCTGAC         AACCTCAACCTGGCACC         (AC)20         200         X74174 / (Eggen et al. 1994)           JAB10         D10S36         10q15         GCTGACCGTGTAACT         (AC)20         200         X74174 / (Eggen et al. 1994)           JAB10         D11S56         11q12-q13         ACGGTAGGGTGCT         (AC)22         270 / 290         X98445 / (Williams et al. 1996)           JNRA177         D11S17         11q13         LGGTAGGGTATGTAACCACGGGTGCT         (AC)24         195         X74201 / (Vaiman et al. 1994)           JDVGA3         D11S27         11q23         GGATAGGTTCCAACCACGGGTC         (AC)17         220         X73945 / (Eggen et al. 1996)           JAB67         D11S34         11q26	IDVGA62	D7S21	7q15	GGGATCTTGGACCAGAAGGTA GGTGACACTCCTCCCATCT	(TA)8(TG)10	163	X86811 / (Zhang et al. 1995)
IDVGA11         DSS20         8q.14         CCTCTCT6GCAACCACCTATTGT CCACCTGAAGGAT         (AC)19         230         Z27077 / (Ferretti et al. 1994)           IDVGA52         DSS21         8q23         GAATCGAACCTTTCATGAGA CCACCTGACCACTGACCACTGACCACCT         (AC)21         215         X85069 / Mezzelani et al. 1995)           INRA144         D9S14         9q25         TCGGTGTGGGAGGTACTACAT TGGTGTGGGGGCTCCGTCACC         (AC)20         200         X74174 / (Eggen et al. 1994)           JAB10         D10S36         10q15         GCTGACCACAGGGTTGCTG CCCACCGTGTTACTTCAC         (AC22)         270 / 290         X89445 / (Williams et al. 1996)           JAB10         D11S56         11q12-q13         ACGGTAGGACAGGGTTGCTG CCCACCATCCCCCCCTTCTG CCCACCATCCCCCCCCTTCTG CCCACCATCCCCACCTTCG CCCACCATCCCCCCCCTTCG ACGGAATTAGCACATCCCACCA         (AC)24         195         X74201 / (Vaiman et al. 1996)           IDVGA3         D11S27         11q12         GGATAGGTATATCCACAC AGGAATTACCACACGG         (AC)17         220         X73945 / (Eggen et al. 1996)           IDVGA3         D11S27         11q23         GGGAATATCCACAGGATCCACAGG AGGATTACCACAGGGTTAGCCACAGG         (AC)17         220         X73945 / (Eggen et al. 1996)           IDVGA3         D11S34         11q26         CCGGGAGTATACCACAGGATCACCAGG GGAATACCCCCGGGCTAGCTC         (AC)17         260         X73	IOBT930	D7S20	7q23-q24	AGCACCACTATCCATCTGTCTC TGGAGTCTAGTCAGAGCGTCAGAA	(TG)15	264	X03162 / (Anastassiadis et al. 1996)
IDVGA52         D8S21         8q23         GAATCGAACCTTTCCTTCAC         (AC)21         215         X85069 / Mezzelani et al. 1995)           INRA144         D9S14         9q25         TCGGTGTGGGAGGTCACTGACT         (AC)20         200         X74174 / (Eggen et al. 1994)           JABI0         D10S36         10q15         GCTGACGGTCACTG         (AC)20         270 / 290         X98445 / (Williams et al. 1994)           HAUT30         D11S56         11q12-q13         ACGGTAGGGGTTGCTG         (AC)21         213         X89255 / (Thieven et al. 1996)           INRA177         D11S17         11q16         CAGCACCAAGGGTTGCTG         (AC)24(CA)22         213         X89255 / (Thieven et al. 1996)           IDVGA3         D11S27         11q23         GGATAGTAGTACCAGGG         (AC)11         158         Z27072 / (Ferretti et al. 1994)           AGGGACTTAGCACATACTCAAAACCCCAAGG         (AC)17         220         X73945 / (Eggen et al. 1996)         AGGGACTTAGCACATACTCAACACAGGAGGTTGCTG         (AC)17         365/377         X98442 / (Williams et al. 1996)           JAB7         D11S38         11q25         CTCCACCTCGGGCCTACT         (AC)17         365/377         X98442 / (Williams et al. 1996)           JAB7         D11S38         11q26         CGGGAAGTACGGGCTCA         complex <sup>b</sup> 188         X81349 /	IDVGA11	D8S20	8q14	CCTTCTGGCAACCACCTATTTGT CCACCTAAGTGTCTCCTGATGGA	(AC)19	230	Z27077 / (Ferretti et al. 1994)
INRA144         D9S14         9q25         TCGCTGTGGGGGGTGACTACAT TGCTGGGGGCTCGCTACT         (AC)20         200         X74174 / (Eggen et al. 1994)           JAB10         D10836         10q15         GCTGACCGTGTAACTTCAC         (AC22)         270 / 290         X98445 / (Williams et al. 1996)           HAUT30         D11856         11q12-q13         ACGGTAGGATATAGGGGTGT         (AC2)         213         X89255 / (Thieven et al. 1996)           INRA177         D11817         11q16         CAGCACCACCACTTCTG         (AC)2(CA)2T(CA)22         213         X89255 / (Thieven et al. 1996)           INVGA3         D11827         11q2         GGATAGGTATAATTAAGTTTCTGGC         (AC)24         195         X74201 / (Vaiman et al. 1994)           INVGA3         D11834         11q25         CTCCACCTTGCCCAGTCC         (AC)17         220         X73945 / (Eggen et al. 1996)           INVGA57         D11S38         11q26         CTGCACCTGGACTGATCAC         (AC)17         365/377         X9842 / (Williams et al. 1996)           INVGA57         D12S29         12q13         ATGGGTCTCGGGTCACCTGATC         (CT)17         365/377         X9842 / (Williams et al. 1995)           INRA209         D12S17         12q26         CTGAGGGATCACTGAC         complex <sup>b</sup> 188         X81349 / (Thieven et al. 1995)	IDVGA52	D8S21	8q23	GAATCGAACCTTTCCTTCTAC AAGCCTCACTTGACCACTGAT	(AC)21	215	X85069 / Mezzelani et al. 1995)
JAB10         D10S36         10q15         GCTGACCGTGTTAACTTCAC CTGACGACACCAAGGTTGCTG         (AC22)         270 / 290         X98445 / (Williams et al. 1996a)           HAUT30         D11S56         11q12-q13         ACGGTAGCATATGAGGGTGT CCCAACCATCCCCACTTCTG CCCAACCATCCCCACTTCTG CCCAACCATCCCCACTTCTG CCCAACCATCCCCACTTCTG CCCAACCATCCCCAAAACC AGGAAACTCCCAAAACC AAGGAACTCCCAAAACC AAGGAACTCCCAAAACC AAGGACTTACACACACAC AAGGACTTACACACACACC AAGGACTTACACACACACC AAGGACTTACACACACACC AAGGACTTACACACACACC INRA195         D11S17         11q16         CACGATAGTCACACACC AAGGACTTACACACACACC AGGACTTACACACACACC AGGACTTACACACACACC AGGACTTACACACACACC AGGACTTACACCACACC	INRA144	D9S14	9q25	TCGGTGTGGGAGGTGACTACAT TGCTGGTGGGCTCCGTCACC	(AC)20	200	X74174 / (Eggen et al. 1994)
HAUT30       D11S56       11q12-q13       ÅCGGTÄGGÅATÄTGAGGÖTGT CCCAACCATCCCCATCTCTG       (AC)2(CA)2T(CA)22       213       X89255 / (Thieven et al. 1996)         INRA177       D11S17       11q16       CAGGATATGACCTATATCA       (AC)24       195       X74201 / (Vaiman et al. 1994)         IDVGA3       D11S27       11q23       GGATAGGTATATTAAGTTTCTGGC       (AC)11       158       Z27072 / (Ferretti et al. 1994)         INRA195       D11S34       11q25       CTCCACCCTCACACC       (AC)17       220       X73945 / (Eggen et al. 1996)         JAB7       D11S38       11q26       CTCCACCCTGGCCCAGTCC       (AC)17       365/377       X98442 / (Williams et al. 1996)         JAB7       D11S38       11q26       CTCCACCGTGGTGTGTG       (AC)17       365/377       X98442 / (Williams et al. 1996)         JAB7       D12S29       12q13       ATGGGTTCTCGGTGTGTG       (AC)17       365/377       X98442 / (Williams et al. 1995)         JINRA209       D12S17       12q25       CTGCAGGCATCAGTCA       complex <sup>b</sup> 188       X81349 / (Thieven et al. 1995)         JAB3       D13S1       13q21-q22       GGAAGAGGAGCTCCTGGCCG       (AC)17       383       Z22746 / (Solinas Toldo et al. 1993)         JAB3       D13S16       13q23       GCAAGGAGGACCTGTGGCG       (AC)3N1	JAB10	D10S36	10q15	GCTGACCGTGTTAACTTCAC	(AC22)	270 / 290	X98445 / (Williams et al. 1996a)
INRA177         D11S17         11q16         CAGCAGTAGTCACCTAAAACC         (AC)24         195         X74201 / (Vaiman et al. 1994)           IDVGA3         D11S27         11q23         GGAGAGTCCCAAAACACCAGG         (AC)11         158         Z27072 / (Ferretti et al. 1994)           INRA195         D11S34         11q25         CTCCACCCTCTGCCCCAGTCC         (AC)17         220         X73945 / (Eggen et al. 1996)           JAB7         D11S38         11q26         GCGGAAGTACAGAGAGTTCTGTG         (AC)17         365/377         X98442 / (Williams et al. 1996a)           JAB7         D11S38         11q26         GCGGAAGTACAGAGAGTTCTGTG         (AC)17         365/377         X98442 / (Williams et al. 1996a)           JAB7         D12S29         12q13         ATGGGTCTCGGTGGTTGTGTG         (AC)17         365/377         X98442 / (Williams et al. 1996a)           JNRA209         D12S12         12q25         CTGCAGGTACAGTGACCT         complex <sup>b</sup> 188         X81349 / (Thieven et al. 1995)           JNRA209         D12S17         12q26         TGCTTTACTAGG         (AC)16         145         X95390 / (Bahri-Darwich et al. 1994)           JAB3         D13S16         13q21-q22         GGAGCCAGGCCCTGTCGCC         (AC)9         226/230         X98440 / (Williams et al. 1996a)           JAB3	HAUT30	D11S56	11q12-q13	ACGGTAGGAATATGAGGGTGT	(AC)2(CA)2T(CA)22	213	X89255 / (Thieven et al. 1996)
IDVGA3         D11S27         11q23         GGATAGGTATAATTAAGTTTCGGC         (AC)11         158         Z27072 / (Ferretti et al. 1994)           INRA195         D11S34         11q25         CTCCACCCTTGCCACATACTCACAC         (AC)17         220         X73945 / (Eggen et al. 1996)           JAB7         D11S38         11q26         GCGGAAGTACAGAGAGGTCCTGGG         (AC)17         365/377         X98442 / (Williams et al. 1996)           JAB7         D12S29         12q13         ATGGGTCTCTGGTGGCTGATC         (AC)17         365/377         X98442 / (Williams et al. 1996)           IDVGA57         D12S29         12q13         ATGGGTCTCTGGTGTGCTGTT         (TC)12(AC)20         196         X85072 / (Mezzelani et al. 1995)           GAGTTTGTGACAGCTGAAGTCA         complex <sup>b</sup> 188         X81349 / (Thieven et al. 1995)           INRA209         D12S17         12q25         TGCTTTACATACAGGC         (AC)16         145         X95390 / (Bahri-Darwich et al. 1994)           ETH7         D13S1         13q21-q22         GTGAGGAGCCACAGTCGTGGC         (AC)9         226/20         X98440 / (Williams et al. 1996)           JAB3         D13S16         13q23         GCAAGGGAGCCCTGTGTGC         (AC)9         226/20         X98440 / (Williams et al. 1996a)           IDVGA76         D14S18         15q14	INRA177	D11S17	11q16		(AC)24	195	X74201 / (Vaiman et al. 1994)
INRA195         D11S34         11q25         CTCCACCTCTGCCCAGTCC         (AC)17         220         X73945 / (Eggen et al. 1996)           JAB7         D11S38         11q26         CTCCACCTTCTGCCCAGTCC         (AC)17         365/377         X98442 / (Williams et al. 1996a)           JAB7         D11S38         11q26         CTCCACCTTCTGCCCAGTCC         (AC)17         365/377         X98442 / (Williams et al. 1996a)           JDVGA57         D12S29         12q13         ATGGCTCTCTCGCGTGTT         (TC)12(AC)20         196         X85072 / (Mezzelani et al. 1995)           GAGTTTGTCAAGGCTGACTT         GAGTTTGTCAAGGCTGACTT         complex <sup>b</sup> 188         X81349 / (Thieven et al. 1995)           INRA209         D12S17         12q26         TGCTTTAACAGCAGC         complex <sup>b</sup> 188         X81349 / (Thieven et al. 1995)           INRA209         D12S17         12q24         TGCAAGGCAGGTTCTTAACCAG         complex <sup>b</sup> 188         X81349 / (Thieven et al. 1994)           GTAGGCAGGTTCTTAACAGGC         (AC)16         145         X95390 / (Bahri-Darwich et al. 1994)         GTAGGCAGGAGTTCTTAACAGG           JAB3         D13S1         13q21-q22         GTGAGACCAGGTCTGTGC         (AC)17         383         Z22746 / (Solinas Toldo et al. 1993)           JAB3         D13S16         13q23	IDVGA3	D11S27	11q23	GGATAGGTATAATTAAGTTTCTGGC	(AC)11	158	Z27072 / (Ferretti et al. 1994)
JAB7D11S3811q26GCGGAGTACGAGAGATCTCTGTG GCAGATACGTGGATGACCCTGATC(AC)17365/377X9842 / (Williams et al. 1996a) $IDVGA57$ D12S2912q13ATGGGTCTCTCGGTGCTGTT GAGTTTGTGCAAGGCTGACTT(TC)12(AC)20196X85072 / (Mezzelani et al. 1995) $HAUT1$ D12S2212q25CTGCAGGGCTACAGTCA GACGGGAGTTGTTAACCAGcomplexb188X81349 / (Thieven et al. 1995) $INRA209$ D12S1712q26TGCTTTACATATCAAGGCAGC GACAGGGAGTTCTTTACTGAG(AC)16145X95390 / (Bahri-Darwich et al. 1994) $ETH7$ D13S113q21-q22GTGAGACCCAGCTCTGGCG CGGACCCAGCTCCTGCCGG(CA)3N15(CA)17383Z22746 / (Solinas Toldo et al. 1993) $JAB3$ D13S1613q23GCAAGGAGGACCCTGTGCG CCTAGTATCCAGTGTGTCC(AC)9226/230X98440 / (Williams et al. 1996a) $IDVGA76$ D14S3514q14CACGGGACCACTGGGACTCT CACCTTGCGAAAGATGAACATSSCP188Z73083 / Williams et al. 1996a) $JAB1$ D15S1815q14CAGCCATTAAGGCTGGGATTCC GGTCACCAGCGCTCACCAGCAG(AC)15221/225X98443 / (Williams et al. 1996a) $JAB8$ D15S2015q21CACGTCACCCGCTTTCTTG GAGGAGGCAGCCCAGGGGG(AC)17200U51937 / (Eggen et al. 1996)	INRA195	D11S34	11q25		(AC)17	220	X73945 / (Eggen et al. 1996)
IDVGA57 D12S29 D12S19 D12S2 D12S29 D12S17 D12S2 D12S2 D12S17 D12S2 D12S17 D12S2 D12S17 D12S1 D12S17 D12S1 D12 D12S1 D	JAB7	D11S38	11q <b>26</b>	GCGGAAGTACAGAGAGTTCTGTG	(AC)17	365/377	X98442 / (Williams et al. 1996a)
HAUT1D12S2212q25CTGCAGGCTACAGTCA GACAGGGCTACAGGCAcomplexb188X81349 / (Thieven et al. 1995)INRA209D12S1712q26TGCTTTACATATCAAGGCAGC GTAGGCAGATTCTTTACTGAG(AC)16145X95390 / (Bahri-Darwich et al. 1994) GTAGGCAGATTCTTACTGAGETH7D13S113q21-q22GTGAGACCCAGCTCCTGCCTGG GGAGCAGGGCTGCAGGGCG(CA)3N15(CA)17383Z22746 / (Solinas Toldo et al. 1993) CGAAGGAGGCCGJAB3D13S1613q23GCAAGGAGCCCTGTGCG CCTAGTATCCAGTCTGTGTGCC(AC)9226/230X98440 / (Williams et al. 1996a)IDVGA76D14S3514q14ACACGGGACCACTGGGACCT CACCTTGCGGAATGAACATSSCP188Z73083 / Williams et al. 1996c)JAB1D15S1815q14CAGCCATTAGGGCTGGGGATTCC GGTCACAGGGCGCTCACAGGGAGGCTCACAGGA GATTTCTGGGGAGGGCTCACAGCGG GGTGAGTGTAACACCTGTGTGCG(AC)15221/225X98443 / (Williams et al. 1996a)JAB8D15S2015q21AGAGGCAGTCCAGTGGAA GGTGAGTCAACACCTGTGGGGG(AC)17200U51937 / (Eggen et al. 1996)	IDVGA57	D12S29	12q13	ATGGGTCTCTCGGTGCTGTT	(TC)12(AC)20	196	X85072 / (Mezzelani et al. 1995)
INRA209D12S1712q26TGCTTTACATATCAAGGCAGC GTAGGCAGATTCTTTACTGAG(AC)16145X95390 / (Bahri-Darwich et al. 1994)ETH7D13S113q21-q22GTGAGACCCAGCTCCTGCCTGG CGGAGCAGGGTGTCAGAGCCG(CA)3N15(CA)17383Z22746 / (Solinas Toldo et al. 1993) CGGAGCAGGGTGTCAGAGCCGJAB3D13S1613q23GCAAGGAGGACCCTGTGCG CTAGTATCCAGTCTGTGCG(AC)9226/230X98440 / (Williams et al. 1996a) CTAGTATCCAGTCTGTGCGIDVGA76D14S3514q14ACAGGGACCATTGGGACCT CACCTTGCGAAAGATGAACATSSCP188Z73083 / Williams et al. 1996c) CACCTTGCGGAACGATTCCAGGGCTGCGGATTCC (AC)18224/240X98434 / (Williams et al. 1996a) GATTTCTGGAGGGCTGAGGGCTJAB1D15S1815q14CAGCCATTAGGGCTGGGATTCC GGTGAGGCTAACACCG GGTGAGGTAACACCTGTGTGCG(AC)15221/225X98443 / (Williams et al. 1996a)JAB8D15S2015q21CACGTCACCGCTTTCTTG GGTGAGGTAACACCTGTGGCG(AC)17200U51937 / (Eggen et al. 1996)	HAUTI	D12S22	12q25	CTGCAGGGCTACAGTCA	complex <sup>b</sup>	188	X81349 / (Thieven et al. 1995)
ETH7D13S113q21-q22GTGAGACCCAGCTCTCTGCCTGG CGGAGCAGGGTGTCAGAGCCG(CA)3N15(CA)17383Z22746 / (Solinas Toldo et al. 1993)JAB3D13S1613q23GCAAGGAGGACCCTGTGCG CTAGTATCCAGTCTGTGTCC(AC)9226/230X98440 / (Williams et al. 1996a)JAB3D13S1613q23GCAAGGAGCACCTGTGGCC CTAGTATCCAGTCTGTGTCCSSCP188Z73083 / Williams et al. 1996a)IDVGA76D14S3514q14ACACGGGACCACTGGGACCT CACCTTGCGAAAGATGAACATSSCP188Z73083 / Williams et al. 1996c)JAB1D15S1815q14CAGCCATTAGGGATGGGATTCC GATTCTGGGAGGCTCACAGCAG(AC)18224/240X98434 / (Williams et al. 1996a)JAB8D15S2015q21CACGTCACCGCTTTCTTG GTGTAACACCTGTGTGCG(AC)15221/225X98443 / (Williams et al. 1996a)INRA224D15S2215q21AGAGGCAGTCCAGTGAAAGG(CA)17200U51937 / (Eggen et al. 1996)	INRA209	D12S17	12q26	TGCTTTACATATCAAGGCAGC	(AC)16	145	X95390 / (Bahri-Darwich et al. 1994)
JAB3D13S1613q23GCAAGAGGAGCCTGTGGCG CCTAGTATCCAGTCTGTGTCCACCG226/230X98440 / (Williams et al. 1996a)IDVGA76D14S3514q14ACACGGACCATGGGACCT CGCTTGCGAAAGATGAACATSSCP188Z73083 / Williams et al. 1996c)JAB1D15S1815q14CAGCGCATTAGGGACTGCGGATTCC GATTCTGGAGGAGCTCACAGGCAGG(AC)18224/240X98434 / (Williams et al. 1996a)JAB8D15S2015q21CACGTCACCGCTTTCTTG GGTGAGTGTAACACTGTGGGGA(AC)15221/225X98443 / (Williams et al. 1996a)INRA224D15S2215q21AGAGGCAGTCCAGTGAAAGG GGTCAGTCACAGTGAAAGG(CA)17200U51937 / (Eggen et al. 1996)	ETH7	D13S1	13q21-q22	GTAGGCAGATTCTTTACTGAG	(CA)3N15(CA)17	383	Z22746 / (Solinas Toldo et al. 1993)
IDVGA76D14S3514q14ACACGGGACCACTGGGACCTSSCP188Z73083 / Williams et al. 1996c) CACCTTGCGAAGAGTGGAACATJAB1D15S1815q14CAGCCATTAAGGGCTGGGATTCC(AC)18224/240X98434 / (Williams et al. 1996a)JAB8D15S2015q21CACGTCACCGCTTTCTCTG(AC)15221/225X98443 / (Williams et al. 1996a)JNRA224D15S2215q21AGAGGCAGTCCAGTGAAAGG(CA)17200U51937 / (Eggen et al. 1996)	JAB3	D13S16	13q23	GCAAGGAGGAGGCCTGTGCG	(AC)9	226/230	X98440 / (Williams et al. 1996a)
CACCTTGCGAAAGATGAACAT       CACCTTGCGAAAGATGAACAT         JAB1       D15S18       15q14       CAGCCATTAGGGCTGGGATTCC       (AC)18       224/240       X98434 / (Williams et al. 1996a)         JAB8       D15S20       15q21       CACGTCACCGCTTTCTGTG       (AC)15       221/225       X98443 / (Williams et al. 1996a)         INRA224       D15S22       15q21       AGAGGCAGTCCAGTGAAAGG       (CA)17       200       U51937 / (Eggen et al. 1996)	IDVGA76	D14S35	14q14	CUTAGTATCCAGTCTGTGTCC ACACGGGACCACTGGGACCT	SSCP	188	Z73083 / Williams et al. 1996c)
GATTTCTGGAGGAGGCTCACAGCAG JAB8 D15S20 15q21 CACGTCACCGCTTTCTCTTG (AC)15 221/225 X98443 / (Williams et al. 1996a) GGTGAGTGTAACACCTGTGTGCG INRA224 D15S22 15q21 AGAGGCAGTCCAGTGAAAGG (CA)17 200 U51937 / (Eggen et al. 1996)	JAB1	D15S18	15q14	CAUCTTGCGAAAGATGAACAT CAGCCATTAAGGGCTGGGATTCC	(AC)18	224/240	X98434 / (Williams et al. 1996a)
GGTGAGTGTAACACCTGTGTGCG INRA224 D15S22 15q21 AGAGGCAGTCCAGTGAAAGG (CA)17 200 U51937 / (Eggen et al. 1996)	JAB8	D15S20	15q21	GATTTCTGGAGGAGGCTCACAGCAG CACGTCACCCGCTTTCTCTTG	(AC)15	221/225	X98443 / (Williams et al. 1996a)
	INRA224	D15S22	15q21	GGTGAGTGTAACACCTGTGTGCG AGAGGCAGTCCAGTGAAAGG	(CA)17	200	U51937 / (Eggen et al. 1996)

Marker	D No.	FISH	Primers (5' to 3')	Repeat	Size (BP)	Acc. No./Reference
JAB4	D15S19	15q21-q22	GCCAGTTTTGTGTACATTATGG GATTACCAGGTACACCTCCAAGC	(AC)13	190/196	X98438 / (Williams et al. 1996a)
INRA046	D15S5	15q25	CAAGIGICIICIAAGGIACIGG CAGCTCATGTGTTTACATGGC	(AC)10	115	X71495 / (Vaiman et al. 1993)
IDVGA10	D15S13	15q25	AGTCTCTGGAACTTCTCCTAC GGCTAAGACCAGCACCTTGAATA	(AC)23	190	Z27076 / (Ferretti et al. 1994)
IDVGA32	D15S16	15q25	GGGACCTTGAGGAGGAGACAG CAGGTGTTGGAGGATGAGAAAGG	(AC)15	205	X85051 / (Mezzelani et al. 1995)
HAUT23	D15829	15027	CAGTCTATGGCACAGTCTTGGTA	complex <sup>b</sup>	451	X89249 / (Thieven et al. 1996)
IORT395	D15823	15027-028	TCTTACACCGCAGTCCTGAA ACAACAGGAAAGCTCTGCCA	(CA)17	92	X95067 / (Niiman et al. 1996)
IDVGA66	D16834	16a12	ACATGTAGCTGTTGATACAGAT	SSCP	209	773082 / (Williams et al. 1996c)
IDVCAC	D10004	16-16	GGCAGTCCTCATCAGACACTC	(AC)14	200	2/5002/ (Winnams et al. 1990c)
IDVGA08	D16523	10010	GAGGGACTTGGGAGGGAAAC	(AC)14	200	X85081 / (Mezzelani et al. 1995)
ETH11	D1685	16q21	GGCGGACAAGAGTTCTCAGAGAC CTTGACACATCATTGAGCACATGC	(GT)16GC(GT)4	214	Z22740/(Solinas Toldo et al. 1993)
JAB9	D16S25	16q17	CAACTTCAAATTCTCTTGG CAGACCTGCAGGCAGCCTGCATC	(AC)10 INTERRUPTED	192/200	X98444 / (Williams et al. 1996a)
IDVGA40	D17S24	17q23	TTCAGTCAAGCAAACAGGACAT TGGGTTCAGTTGCTGGGTGGAGA	(AC)16	240	X85055 / (Mezzelani et al. 1995)
IOZARA975	D17S29	17q26	TGAAGGAGATGTGGGTTTG TTGACGAGAGACCCAAGAAGTAAACC	(CA)2TG(CA)10(CT)2	233	U47616 / (Martin-Burriel et al. 1996a)
IDVGA31	D18S10	18q12-q13		(AC32)	214	X85050 / (Mezzelani et al. 1995)
HAUT14	D18S17	18 <b>q21</b>	TGACCTTCACTCATGTTATTAA	complex <sup>b</sup>	161	X81350 / (Thieven et al. 1995)
IDVGA55	D18S16	18q24	GTGACTGTATTTGTGAACACCTA	(AC)12	199	X85071 / (Mezzelani et al. 1995)
IDVGA46	D19S18	19q16	AAATCCTTTCAAGTATGTTTTCA	(AC)11	205	X85062 / (Mezzelani et al. 1995)
IOBT34	D1984	19q22	TCCTTCCATCAATGTGTCAGTCC	(GC)12AT(GT)20	190	U31002 / (Olsaker et al. 1996b)
IDVGA44	D19S17	19q22	TTGGGATTCGGTGGTCAGICIG GGGAGAATGGATGGAACCAAAT	(AC)19	211	X85059 / (Mezzelani et al. 1995)
ETH3	D19S2	19q23	TTCGAAGACGGGCAGACAGG GAACCTGCCTCTCCTGCATTGG	(CA)26	122	Z22744 / (Solinas Toldo et al. 1993)
COSMAP1B	MAP1B	20q14-q15	ACTTCGCCTGTGGCCAAGTAGG TACTGATTCTGACTGATTATGTCT	(CA)12	280	(Eggen 1992)
IDVGA45	D21S25	21a15	GGCTGTGCCTCGATAGATGGTGCT GTGGTGGCAAAGAGTCAGA	(AC)12	149	X85060 / (Mezzelani et al. 1995)
HAUT28	D21S46	21015-016	AACAGCCCTGATTTCCATA GCTTGTTCCTATCACCCATTTA	(AC)5G(CA)2TTCC(CA)7	359	X89253 / (Thieven et al. 1996)
IDVGA 39	D21S24	21a23nrox	GAAGGCTTGGAAAATGTAATC	(AC)17	191	X85054 / (Mezzelani et al. 1995)
IDVGA30	D21523	21a23		(AC)40	240	X85049 / (Mezzelani et al. 1995)
INDA 10A	D21025	27912	TTGTAAAACTCGGGGCATAAGCA	(AC)12	195	X73044 / (Eggen et al. 1996)
INKA194	D22519	22415	ACAGTTCTGGAGTTGCTTGATG	(AC)12	105	X73944 / (Eggen et al. 1996)
IOB1528	D23827	23q14	AACTCCAGGAGTTGGTGATGG	(01)16	182	X95068 / (Nijman et al. 1996)
R11479	D23S38	23qter	CLAGIAACGATCIAGCAAAGGGATC	SSCP	111	U59512/(Olsaker et al. 1996c)
IOBT1401	D24S13	24q21	CATAGGTTTGTTTAATGCTCAG TGATAGATAGCATTTGATTGGC	(CA)14	110	X95069 / (Nijman et al. 1996)
IDVGA71	D25S12	25q12-q13	GCTAGGCATCTGGCAAATAG CCTCAGAGAAGCCTGGTCAT	(AC)2AG(AC)16	192	X85053 / (Zhang et al. 1995)
HAUT39	D25S19	25q17	GCTTGAAAGAAAAATGCCAAAGA GAGCCTCCCACCCCAACAG	CTN(AC)11GC	145	X89260 / (Thieven et al. 1996)
INRA222	D25S11	25q19	GTGGAGGAGTCTTACAGTCCAG GATCTCTGGATTGAAGGAAC	(GT)11	160	U51935 / (Eggen et al. 1996)
IDVGA59	D26S14	26q22	AACCCAAATATCCATCAATAG	(AC)23	256	X85074 / (Mezzelani et al. 1995)
IOBT313	D27S14	27q23	GAATCAATAAAGAAGATGCAGCACG	(CA)12	124	U59511 / (Olsaker et al. 1996a)
INRA201	D28S17	28q12	CTCACTAAGCTGACTCAGGC	(AC)11	148	U51932 / (Eggen et al. 1996)
IDVGA29	D28S12	28q13	CCCACAAGGTTATCTATCTCCAG	(AC)23	149	X85048 / (Mezzelani et al. 1995)
IDVGA43	D28S18	28q17	GGGGGGTTGGAAGTATTATCTG	(AC)14	171	X85058 / (Mezzelani et al. 1995)
IDVGA8	D28S10	28q18-q19		(AC)19	224	Z27074 / (Ferretti et al. 1994)
IDVGA7	D2982	29q19	IAGCAGAAAGCACAGGAGTC GGGTGGGCTTCATTTCTATG	(AC)9	195	Z27073 / (Ferretti et al. 1994)
JAB5	D29812	29q12	CAGCCACTGTCTCCTCCCAC CCTAGCGATCTCTTGATTGAG	(AC)17	292/302	X98440 / (Williams et al. 1996a)
INRA211	D29S24	29q16	CCTTTGGAACTTCTGATGGTGC TTACAGGTGCAGCTCAGATGCTG	(CA)8C(CA)4	206	U51933 / (Eggen et al. 1996)

Table	1.	Continued.

Marker	D No.	FISH	Primers (5' to 3')	Repeat	Size (BP)	Acc. No./Reference
	DX830		CCTACTGTTTAGCACAGGGAACTC	(AC)12		V96912 / (7bang at al. 1005)
IDV GA62	DA350	лцэч	GGCAAACCATTCCAGTATTC	(AC)12	191	A00015 / (Zhang et al. 1995)
IOZARA1489	DXYS4	Xq42-q43	ATGCACCAGACACGGAATGG TGCGGCAGCGTCAAAATGAC	complex <sup>b</sup>	292	U47615 / (Martin-Burriel et al. 1996b)

Symbols for anonymous DNA loci: *IDVGA*, Istituto per la Difesa e la Valorizzazione del Germoplasma Animale, CNR, Italy; *JAB* and *RI*, Roslin Institute (Edinburgh), Roslin, Scotland; *INRA*, Institute National pour la Recherche Agronomique, Jouy en Josas, France; *HAUT*, Department of Animal Breeding and Genetics, Hannover School of Veterinary Sciences, Hannover, Germany; *ETH*, Swiss Federal Institute of Technology, Zurich, Switzerland; *IOZARA*, Norwegian College of Veterinary Medicine, Oslo, and Facultad de Veterinaria, Zaragoza, Spain; *IOBT*, Norwegian College of Veterinary Medicine, Oslo, Norway.

mosomes, covering 40.6% of the whole genome (Table 2). In some cases, the size of the interval used was increased by including genes for which physical and linkage mapping data were available. Thus, on Chr 6 the interval was between D6S24 and CAS@ (Gallagher et al. 1994), on Chr 7 between D7S30 and RASA (Eggen et al. 1992), on Chr 11 between D11S56 and LGB (Hayes and Petit 1993), and on Chr 17 between D17S29 and FGG (Johnson et al. 1993). An interval could be defined on Chr 9 despite the presence of only one cosmid-derived, physically anchored locus: connexin 43 (GJA1) was used as a second marker, which was recently mapped by FISH to 9q15-q16 (Castiglioni et al. 1996).

The total genetic distance covered by the intervals between markers in 1666 cM, giving an estimate for the whole genome of 4176 cM, which is much larger than the size of 2800 cM predicated on the basis of chiasmata counts (Jagiello et al. 1974; Logue and Harvey 1978) and larger still than 3540 cM suggested in the current version of the CGD linkage map from the distribution of markers (Barendse et al. 1996). Table 2 presents estimates of genome size obtained from the evaluation of individual intervals. There is great variation in the estimated values, which is reflected in a mean of 4176 cM with a standard error of 1360 cM. However, it is interesting to inspect the data for some of the intervals. D3S29 and D3S24 span 3.10% of the genome, or more significantly about 66% of Chr 3, which, when extrapolated, gives an estimate of 5063 cM for the total genome. From other chromosomes that are well covered by the anchored markers, generally high values are obtained for the inferred genome size: Chr 18 (61.5%) 6519 cM; Chr 28 and Chr 29 (both 70%), 5868 cM and 7132 cM respectively. On the other hand, the intervals for Chr 1 (68.5% coverage) and for Chr 25 (67.5%) suggest a size for the genome that is much below the average and close to the estimate based on chiasmata counts.

#### Discussion

In this study cosmid and phage clones have been used as a tool to add to the developing linkage map of the bovine genome by providing cytogenetically mapped anchored loci. Microsatellite markers were derived from 74 cosmid and 9 phage clones and have been placed on the genetic map, while the parent clones have been localized on the bovine chromosomes by FISH. All 30 bovine chromosomes have at least one microsatellite marker mapped by FISH.

The use of microsatellite markers derived from cosmid and phage clones as anchor loci for the genetic map has highlighted the usefulness of combining genetic and physical mapping. Compared with the first published linkage map (Barendse et al. 1994), the set of markers described here allowed the anchoring and orientation of the linkage groups corresponding to Chrs 3, 4, 7, 8, 9, 10, 12, 13, 16, 18, 19, and Chr 28 and led to the identification of four unassigned linkage groups, corresponding to Chrs 9, 12, 16, and <sup>b</sup>The complete repeats are as follows: HAUT1, (TG)3Å(GT)5A(TG)5A(GT)5A(TG) 10(CG)2; HAUT23, (AC)6CAGT(AC)6CAGTTCAGGA(AC)9CCCAGTT(CA)17C GNGTTCAGG(AC)13CCCAGTT(CA)17CGNGTTCAGG(AC)18CCCAGTTCT (CA)14CGNGTTCAGG(AC)13CGAGTTCAGG(AC)17; HAUT14, (AT)2(GT)15G(GT)9G(GT)13C(TG)2; IOZARA1489, (CT)5GA(GT) 2(CT)5(CA)12CT(CA)4.

29, formerly Chr 25 (ISCNDA 1989, 1990; Popescu et al. 1996). The linkage group for Chr 27 with marker *D27S14*, 27q23, was assigned and orientated by a single appropriately placed marker, confirming the recent identification of the last unassigned linkage group (Masabanda et al., 1996). Similarly, cosmid MAP1B, 20q14-q15, confirmed the orientation of the linkage group for Chr 20.

Another interesting feature of cytogenetically mapped loci, anchored to the linkage map by means of highly polymorphic STS markers, is that they allow an evaluation of the alignment between the physical map and genetic maps (Bray-Ward et al. 1996). This, in turn, gives an estimate of the rate of genetic recombination and, ultimately, of genome size.

In this study, 83 markers defined intervals on 20 chromosomes, representing 40.6% of the total karyotype, and a genetic distance of 1666 cM. An estimate of 4176 cM was thus obtained for the whole genome, which is greatly in excess of the 2800 cM predicted by chiasmata counts (Jagiello et al. 1974; Logue and Harvey 1978). Our data would also suggest a larger size than the 3540 cM proposed from data in the present bovine linkage map, based on the Cattle Genotype Database (CGD; Barendse et al. 1996). However, care must be taken when considering these estimates as they have large errors associated with them and do overlap at the extremes of the range. Typing errors inflate the estimate of genetic length, particularly considering the relatively low number of markers presented in this study. Nevertheless, our finding is of special interest in light of the results published in a similar study on the porcine genome (Ellegren et al. 1994b). Ellegren and colleagues estimated a genome size of about 2000 cM with a set of markers covering 37.5% of the pig karyotype, a figure much closer to mouse, 1600 cM, than to human, 3800-4000 cM, despite a very similar length of 2.8 billion base pairs (bp). Conversely, our estimate for the size of bovine genome of 4176 cM suggests it might be closer to human and possibly even larger.

The data cannot be taken as conclusive. In particular, it is known from extensive and careful studies in human (Bray-Ward et al. 1996) that the relationship between physical and genetic maps is not uniform along the length of a chromosome. Genetic recombination is generally suppressed in the centromere-proximal regions. Moreover, the striking reduction of genetic recombination in the human acrocentric chromosomes (Bray-Ward et al. 1996) has a special significance here, since all the chromosomes in the bovine karyotype are acrocentric. Conversely, the telomeric regions of several human chromosomes cover very long distances in recombination map units, cM, if compared with the short spacing of the physically mapped markers. Special care should also be taken in evaluating FISH data for telomeric markers, since it is often difficult to interpret the ends of chromosomes by fluorescence microscopy. However, imprecise FISH assignments of telomeric markers cannot alone explain the large estimated genome size presented in this paper. This could be the case for two of the



Fig. 1. Integration of the physical and genetic map of the bovine genome Idiograms of G-banded chromosomes (Popescu et al. 1996) are flanked by the corresponding linkage groups. Linkage data are based on the CGD (W. Barendse, CSIRO). Distances are in cM and the linkage groups are drawn to scale, with the total length given at the bottom. D numbers identify anonymous DNA loci. The physically mapped loci anchored on the genetic map are shown in between the idiograms and the linkage groups with the corresponding chromosomal band assignments. For Chr 15 the linkage

group is drawn at a 200% scale and only some markers are shown (see Table 1 for a complete list). D numbers without a physical assignment are of markers that define the current ends of the linkage groups. For the following genes the physical mapping information was incorporated in the figure: *CAS@* (Gallagher et al. 1994), *RASA* (Eggen et al. 1992), *LGB* (Hayes and Petit 1993), *FGG* (Johnson et al. 1993), *GJA1* (Castiglioni et al. 1996).

Table 2. Comparison of the bovine physical and genetic maps

Chr	Markers (Longest interval)	Length (% genome)	Genetic distance sex aver. (cM)	Inferred genome size sex aver. (cM)
1	D1S42-D1S40	4.02	121	3010
2	D2S32–D2S7	3.516	112	3185
3	D3S29-D3S24	3.114	159	5063
5	D5S3–D5S30	1.635	81	4954
6	D6S24–CAS@	2.62	90	3435
7	D7S30-RASA	2.27	91	4009
8	D8S20D8S21	2.13	107	5023
9	GJA1-D9S14	1.79	28	1564
11	D11S17LGB	3.09	123	3980
12	D12S29-D12S17	2.19	83	3790
15	D15S18D15S23	1.63	47	2883
16	D16S34D16S5	1.63	59	3620
17	D17S29-FGG	2.00	91	4550
18	D18S10-D18S16	1.58	103	6519
19	D19S18D19S2	1.14	64	5614
21	D21S25D21S23	1.22	47	3852
23	D23S27-D23S38	1.07	55	5121
25	D25S12-D25S11	1.33	37	2782
28	D28S17-D28S10	1.21	71	5868
29	D29S12D29S2	1.36	97	7132
	Total interval	40.66	1666	4176

small chromosomes, namely Chr 28 and Chr 29, and for Chr 5, which has several markers mapped to the telomere, but not for other chromosomes, such as Chrs 17, 18, 7, and 3. As an example, the interval spanning markers D7S30 and RASA on Chr 7 does not include the telomeric region, yet predicts a genome size above 4000 cM. On Chr 3, one could argue that the large estimate of 5063 cM is due to the inclusion of four telomeric markers in the interval; however, a large estimate is still obtained (4980 cM) when considering the interval between markers D3S29 and D3S25, which do not include the telomere region of the chromosome. A chromosome size-dependent control of recombination has been hypothesized in organisms as distant as yeast and human (Kaback 1996). Thus, small human chromosomes.

Curiously, there seems to be a better correlation between the genetic size of a genome and the number of chromosomes, rather than size in base pairs. At least, this is what emerges from the analysis of the few mammalian species where substantial linkage data are available: thus, for the mouse (19 autosomes, X and Y) and pig (18, X and Y), the genome size is small (1600 and 2000 cM respectively), compared with human (23, X and Y) with 3800-4000 cM and now for the cattle genome (29, X and Y), which is apparently above 4100 cM (Davisson and Roderick 1989; Weissenbach et al. 1992; Dietrich et al. 1994; Archibald et al. 1995; Hudson et al. 1995). It will be interesting to see the result for sheep, when the linkage map is more complete (Crawford et al. 1995). In chicken the available maps are incomplete (Bumstead and Palyga 1992; Levin et al. 1994); however, the 33 known linkage groups in the chicken seem to cover as much as 3200 cM, and possibly more, despite the genome being  $1 \times 10^9$  bp, about one-third the size of the mammalian species mentioned above. Indeed, there is evidence that the rate of genetic recombination might be three times higher in chicken than in human.

A better coverage of physically mapped markers is required to assess the alignment of the physical and genetic maps of cattle. However, based on a 40.6% physical coverage of the genome, the data presented in this paper suggest that the current linkage map of the bovine genome may cover less than the hypothesized 95% (Barendse et al. 1996).

The availability of physical resources, such as YAC and BAC libraries (Cai et al. 1995), as well as the development of tools like radiation hybrid somatic cell lines (Gyapay et al. 1996), will be crucial to a better integration of the physical and genetic maps of

cattle, and ultimately for the accurate localization of trait loci as a prelude to finding the genes themselves.

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## References

- Anastassiadis, C., Leyhe, B., Olsaker, I., Friedl, R., Rottmann, O., Hiendleder, S., Erhardt, G. (1996). Three polymorphic microsatellites for bovine chromosomes 7, 12, and 19. Anim. Genet. 27, 125–126.
- Archibald, A.L., Haley, C.S., Brown, J.F., Couperwhite, S., McQueen, H.A., Nicholson, D., Coppieters, W., Van de Weghe, A., Stratil, A., Winterø, A.K. Freedholm, M., Larsen, N.J., Nielsen, V.H., Milan, D., Woloszyn, N., Robic, A., Dalens, M., Riquet, J., Gellin, J., Caritez, J.-C., Burgaud, G., Ollivier, L., Bidanel, J.-P., Vaiman, M., Renard, C., Geldermann, H., Davoli, R., Ruyter, D., Verstege, E.J.M., Groenen, M.A.M., Davies, W., Høyheim, B., Keiserud, A., Andersson, L., Ellegren, H., Johansson, M., Marklund, L., Miller, J.R., Andersson Dear, D.V., Signer, E., Jeffreys, A.J., Moran, C., Le Tissier, P., Muladno, Rothschild, M.F., Tuggle, C.K., Vaske, D., Helm, J., Liu, H.-C., Rahman, A., Yu, T.-P., Larson, R.G., Schmitz, C.B. (1995). The PiGMaP consortium linkage map of the pig (*Sus scrofa*). Mamm. Genome 6, 157–175.
- Bahri-Darwich, I., Vaiman, D., Olsaker, IL., Oustry, A., Cribiu, E.P. (1994). Assignment of bovine synteny groups U27 and U8 to R-banded Chromosomes 12 and 27, respectively. Hereditas 120, 261–265.
- Barendse, W., Vaiman, D., Kemp, S.J., Sugimoto, Y., Armitage, S., Williams, J.L., Sun, H.S., Eggen, A., Agaba, M., Aleyasin, S.A., Band, M., Bishop, M.D., Buitkamp, J., Byrne, K., Collins, F., Cooper, L., Coppettiers, W., Denys, B., Drinkwater, R.D., Easterday, K., Elduque, C., Ennis, S., Erhardt, G., Ferretti, L., Flavin, N., Gao, Q., Georges, M., Gurung, R., Harlizius, B., Hawkins, G., Hetzel, D.J.S., Hirano, T., Hulme, D., Joergensen, C.,, Kessler, M., Kirkpatrick, B.W., Konfortov, B., Kostia, S., Kuhn, C., Lenstra, J., Leveziel, H., Lewin, H.A., Leyhe, B., Li, L., Martin Buriel, I., McGraw, R.A., Miller, J.R., Moody, D.E., Moore, S.S., Nakane, S., Nijman, I., Olsaker, I., Pomp, D., Rando, A., Ron, M., Shalom, A., Soller, M., Teale, A.J., Thieven, I., Urquhart, B., Vage, D-I., Van de Weghe, A., Varvio, S., Velmala, R., Vilkki, J., Weikard, R., Woodside, C., Womack, J.E., Zanotti, M., and Zaragoza, P. (1996). A medium density genetic linkage map of the bovine genome. Mamm. Genome, 8, 21–28.
- Barendse, W., Armitage, S.M., Kossarek, L.M., Shalom, AL., Kirkpatrick, B.W., Ryan, A.M., Clayton, D., Li, L., Neiberg, H.L., Zhang, N., Grosse, W.M., Weiss, J., Creighton, P., McCarthy, F., Ron, M., Teale, A.J., Fries, R., McGraw, R.A., Moore, S.S., Georges, M., Soller, M., Womack, J.E., Hetzel, D.J.S. (1994a). A genetic linkage map of the bovine genome. Nature Genet. 6, 227–235.
- Bishop, M.D., Kappes, S.M., Keele, J.W., Stone, R.T., Sunden, S.L.F., Hawkins, G.A., Solinas-Toldo, S., Fries, R., Grosz, M.D., Yoo, J., Beattie, C.W. (1994). A genetic linkage map for cattle. Genetics 136, 619–639.
- Bray-Ward, P., Menninger, J., Lieman, J., Desai, T., Mokady, N., Banks, A., Ward, D.C. (1996). Integration of the cytogenetic, genetic, and physical maps of the human genome by FISH mapping of the CEPH YAC clones. Genomics 32, 1–14.
- Bumstead, N., Palyga, Y. (1992). A preliminary linkage map of the chicken genome. Genomics 13, 690–697.
- Cai, L., Taylor, J.F., Wing, R.A., Gallagher, D.S., Woo, S.-S., Davis, S.K. (1995). Construction and characterization of a bovine bacterial artificial chromosome library. Genomics 29, 413–425.
- Castiglioni, B., Ferretti, L., Tenchini, M.L., Mezzelani, A., Simonic, T., Duga, S. (1996). Physical mapping of connexin 32 (GJB1) and 43 (GJA1) genes to bovine chromosomes Xq22 and 9q15-16 by fluorescence in situ hybridization. Mamm. Genome 7, 634-635.
- Crawford, A.M., Dodds, K.G., Ede, A.J., Pearson, C.A., Montgomery, G.W., Garmonsway, H.G., Beattie, A.E., Davies, K., Maddox, J.F., Kappes, S.W., Stone, R.T., Nguyen, T.C., Penty, J.M., Lord, E.A., Broom, J.E., Buitkamp, J., Schwaiger, W., Epplen, J.T., Matthew, P.,

Matthews, M.E., Hulme, D.J., Beh, K.E., McGraw, R.A., Beattie, C.W. (1995). An autosomal genetic linkage map of the sheep genome. Genetics 140, 703–724.

- Davisson, M.T., Roderick, T.H. (1989). Linkage Map. In Genetic Variants and Strains of the Laboratory Mouse, ed. 2, M.F. Lyon, A.G. Searles, eds. (Stuttgart: Gustay Fischer Verlag), pp. 416–427.
- Dietrich, W.F., Miller, J.C., Steen, R.G., Merchant, M., Damron, D., Nahf, R., Gross, A., Joyce, D.C., Wessel, M., Dredge, R.D., Marquis, A., Stein, L.D., Goodman, N., Page, D.C., Lander, E.S. (1994). A genetic map of the mouse with 4006 simple sequence length polymorphisms. Nature Genet. 7, 220-245.
- Eggen, A. (1992). Recherche de marqueurs polymorphes dans la région putative de l'Amyotrophie spinale bovine a l'aide de la cartographie comparée. Ph.D. Thesis ETHZ Nr. 9835.
- Eggen, A., Fries, R. (1995). An integrated cytogenetic and meiotic map of the bovine genome. Anim. Genet. 26, 215-236.
- Eggen, A., Solinas-Toldo, S., Dietz, A.B., Womack, J., Stranzinger, G., Fries, R. (1992). RASA contains a polymorphic microsatellite and maps to bovine syntenic group U22 on chromosome 7q24-qter. Mamm. Genome 3, 559–563.
- Eggen, A., Bahri-Darwich, I., Mercier, D., Vaiman, D., Cribiu, E.P. (1994). Assignment of bovine synteny group U2 to chromosome 9. Anim. Genet. 25, 183-185.
- Eggen, A., Bahri-Darwich, I., Elduque, C., Petit, E., Oustry, A., Cribiu, E.P., Leveziel, H. (1996). New cosmids containing microsatellites for mapping the bovine genome. Mamm. Genome, in preparation.
- Ellegren, H., Chowdhary, B., Johansson, M., Andersson, L. (1994a). Integrating the porcine physical and linkage map using cosmid-derived markers. Anim. Genet. 25, 155-164.
- Ellegren, H., Chowdhary, B.P., Johansson, M., Marklund, L., Fredholm, M., Gustavsson, I., Andersson, L. (1994b). A primary linkage map of the porcine genome reveals a low rate of genetic recombination. Genetics 137, 1089-1100.
- Ferretti, L., Leone, P., Pilla, F., Zhang, Y., Nocart, M., Guérin, G. (1994). Direct characterization of bovine microsatellites from cosmids: polymorphism and synteny mapping. Anim. Genet. 25, 209–214.
- Fries, R., Eggen, A., Womack, J.E. (1993). A bovine genome map. Mamm. Genome 4, 405–428.
- Gallagher, D.S., Schelling, C., Groenen, M.A.M., Womack, J. (1994). Confirmation that the casein gene cluster resides on cattle chromosome 6. Mamm. Genome 5, 524.
- Green, P., Falls, K., Crooks, S. (1990). Documentation for CRI-MAP, version 2.4 (ST Louis: Washington University School of Medicien).
- Gyapay, G., Schmitt, K., Fizames, C., Jones, H., Vega-Czarny, N., Spillet, D., Muselet, D., Prud'Homme, J.-F., Dib, C., Auffray, C., Morissette, J., Weissenbach, J., Goodfellow, P.N. (1996). A radiation hybrid map of the human genome. Hum. Mol. Genet. 5, 339–346.
- Harlizius, B., Guérin, G., Ferretti, L. (1996). IDVGA65 (D6S29), an SSCP marker assigned to BTA6 by means of FISH, genetic and synteny mapping. Anim. Genet., in press.
- Hayes, H., Petit, E.J. (1993). Mapping of the  $\beta$ -lactoglobulin gene and of an immunoglobulin M heavy chain-like sequence to homoeologous cattle, sheep and goat chromosomes. Mamm. Genome 4, 207–210.
- Hudson, T.J., Stein, L.D., Gerety, S.S., Ma, J., Castle, A.B., Silva, J., Slonim, D.K., Baptista, R., Kruglyak, L., Xu, S.H., et al. (1995). An STS-based map of the human genome. Science 270, 1945–1954.
- ISCNDA 1989 (1990). International system for cytogenetic nomenclature of domestic animals (Di Berardino D, Hayes H, Fries R, Long S, eds.). Cytogenet. Cell Genet. 53, 65–79.
- Jagiello, G.M., Miller, W.A., Ducayen, M.B., Lin, J.S. (1974). Chiasma frequency and disjunctional behaviour of ewe and cow oocytes matured in vitro. Biol. Reprod. 10, 354–363.
- Johnson, S.E., Barendse, W., Hetzel, D.J. (1993). The gamma fibrinogen gene (FGG) maps to chromosome 17 in both cattle and sheep. Cytogenet. Cell Genet. 62, 176–180.
- Kaback, D.B. (1996). Chromosome-size dependent control of meiotic recombination in humans. Nature Genet. 13, 20-21.
- Kühn, C., Weikard, R., Goldammer, T., Olsaker, I. (1996). FBN3 (D6S24)—a bovine microsatellite derived from cosmid clOBT475 at chromosome 6q12-14. Anim. Genet. 27, 61.
- Levin, I., Santangelo, L., Cheng, H., Crittenden, L.B., Dodgson, J.B. (1994). An autosomal genetic linkage map of the chicken. J. Hered. 85, 79–85.
- Lichter, P., Tang, C.-J., Call, K., Hermanson, G., Evans, G.A., Housman, D., Ward, D.C. (1990). High resolution mapping of human chromosome 11 by in situ hybridization with cosmid clones. Science 247, 64–69.

- Logue, D.N., Harvey, M.J.A. (1978). Meiosis and spermatogenesis in bulls heterozygous for a presumptive 1/29 Robertsonian translocation. J. Reprod. Fertil. 54, 159–165.
- Martin-Burriel, I., Chowdhary, B.P., Prakash, B., Zaragoza, P., Olsaker, I. (1996a). A polymorphic bovine dinucleotide repeat D17S29 (IOZARA975) at chromosome 17q26. Anim. Genet. 27, 287.
- Martin-Burriel, I., Chowdhary, B.P., Prakash, B., Zaragoza, P., Olsaker, I. (1996b). A polymorphic bovine dinucleotide repeat DXYS4 (IOZARA1489) located at Xq42-q43 and linked within the pseudoautosomal region of the sex chromosomes. Anim. Genet. 27, 287.
- Masabanda, J., Kappes, S.M., Smith, T.P.L., Beattie, C.W., Fries, R. (1996). Mapping of a linkage group to the last bovine chromosome (BTA27) without an assignment. Mamm. Genome 7, 229–230.
- Mezzelani, A., Solinas Toldo, A., Nocart, M., Guérin, G., Ferretti, L., Fries, R. (1994). Mapping of syntenic groups U7 and U27 to bovine chromosomes 25 and 12, respectively. Mamm. Genome 5, 574–576.
- Mezzelani, A., Zhang, Y., Redaelli, L., Castiglioni, B., Leone, P., Williams, J., Solinas-Toldo, S., Wigger, G., Fries, R., Ferretti, L. (1995). Chromosomal localization and molecular characterization of 53 cosmidderived bovine microsatellites. Mamm. Genome 6, 629-635.
- Nijman, I.J., Lenstra, J.A., Schwerin, M., Olsaker, I. (1996). Polymorphisms and physical locations of three bovine microsatellite loci: IOBT395, IOBT528 and IOBT1401. Anim. Genet. 27, 221–222.
- O'Brien, S.J., Womack, J.E., Lyons, L.A., Moore, K.J., Jenkins, N.A., Copeland, N.G. (1993). Anchored reference loci for comparative genome mapping in mammals. Nature Genet. 3, 103–112.
- Olsaker, I., Prakash, B., Guttersrud, O.A., Chowdhary, B.P. (1996a). D27S14 (IOBT313): a genetic and physical marker for bovine chromosome 27, submitted.
- Olsaker, I., Solinas Toldo, S., Fries, R. (1996b). A highly polymorphic bovine dinucleotide repeat D19S4 (IOBT 34) at chromosome 19q21. Anim. Genet. 27, 58-59.
- Olsaker, I., Williams, J.L., Chowdhary, B.P., Karlsson, L., Urquhart, B.G.D., Prakash, B. (1996c). Physical and genetic mapping of a bovine dinucleotide repeat marker D23S38 (IOBT1479) to chromosome 23q24-25. Anim. Genet., in press.
- Olson, M., Hood, L., Cantor, C., Botstein, D. (1989). A common language for physical mapping of the human genome. Science 245, 1434–1435.
- Popescu, C.P., Long, S., Riggs, P., Womack, J.E., Schmutz, S., Fries, R. (1996). Standardization of cattle karyotype nomenclature. Report of the committee for the standardization of the cattle karyotype. Cytogenet. Cell Genet., in press.
- Solinas Toldo, S., Fries, R., Steffen, P., Neibergs, N.H., Barendse, W., Womack, J.E., Hetzel, D.J.S. (1993). Physically mapped, cosmidderived microsatellite markers as anchor loci on bovine chromosomes. Mamm. Genome 4, 720–727.
- Thieven, U., Harlizius, B., Simon, D. (1995). Dinucleotide repeat polymorphism at the bovine HAUT1 and HAUT14 loci. Anim. Genet. 26, 123.
- Thieven, U., Solinas Toldo, S., Fries, R., Barendse, W., Simon, D., Harlizius, B. (1996). Polymorphic CA-microsatellites for the integration of the bovine genetic and physical map. Mamm. Genome, in press.
- Vaiman, D., Bahri-Darwich, I., Mercier, D., Yerle, M., Eggen, A., Leveziel, H., Guérin, G., Gellin, J., Cribiu, E.P. (1993). Mapping of new bovine microsatellites on cattle chromosome 15 with somatic cell hybrids, linkage analysis, and fluorescence in situ hybridization. Mamm. Genome 4, 676–679.
- Vaiman, D., Mercier, D., A., E., Bahri-Darwich, I., Grosh, C., Cribiu, E.P., Dolf, G., Oustry, A., Guérin, G., Levéziel, H. (1994). A genetic and physical map of bovine chromosome 11. Mamm. Genome 5, 553–556.
- Weissenbach, J., Gyapay, G., Dib, C., Vignal, A., Morissette, J., Millasseau, P., Vaysseix, G., Lathrop, M. (1992). A second-generation linkage map of the human genome. Nature 359, 794–801.
- Williams, J.L., Morgan, A.L.G., Guérin, G., Urquhart, B.G.D. (1996a). Ten cosmid-derived bovine microsatellite markers. Anim. Genet. 27, 380.
- Williams, J.L., Olsaker, I., Teres, V.M. (1996b). Using SSCP to facilitate mapping microsatellite loci. Mamm. Genome 8, in press.
- Williams, J.L., Urquhart, B.G., Castiglioni, B., Ferretti, L. (1996c). Mapping two microsatellite loci, IDVGA66 (D16S34) and IDVGA76 (D14S35), by SSCP. Anim. Genet., in press.
- Womack, J.E., Moll, Y.D. (1986). Gene map of the cow: conservation of linkage with mouse and man. J. Hered. 77, 2-7.
- Zhang, Y., Redaelli, L., Castiglioni, B., Mezzelani, A., Ferretti, L. (1995). Five polymorphic bovine microsatellite loci, IDVGA-62A, IDVGA-71, IDVGA-82, IDVGA-88, IDVGA-90. Anim. Genet. 26, 365–366.