

DETECTION OF QUANTITATIVE TRAIT LOCI AFFECTING MILK PRODUCTION OR SOMATIC CELL SCORE IN THREE FRENCH DAIRY SHEEP BREEDS BY PARTIAL GENOTYPING ON SEVEN CHROMOSOMES

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INTRODUCTION

In countries as France, Italy or Spain, dairy sheep have been efficiently selected on milk traits. Last decade, important efforts in research and extension have concentrated on selection of new traits, as milkability, resistance to mastitis or longevity of the animals, for which the economic importance has increased rapidly in the last years. To face these new goal, on-going researches are combining classical quantitative approaches and QTL detection. Indeed QTL information could be particularly appropriate for traits difficult or expensive to measure. Therefore two complementary QTL detection projects have been implemented in France and Italy : one based on crossbreeding between Sarda and Lacaune breeds in an experimental flock in Sardinia with numerous phenotypic measurements combined with a genome scan (Carta *et al.*, 2002), and the other based on purebred granddaughter families of French dairy sheep breeds (Barillet, 1999). In a first step, seven chromosomal regions previously associated in dairy cattle with milk production traits or somatic cell score (SCS) were tested in 13 granddaughter French dairy sheep families. The purpose of this paper is to present preliminary results of this limited QTL detection project.

MATERIAL AND METHODS

Experimental design. The QTL design was a granddaughter design (Weller *et al.*, 1990) including 419 AI rams distributed in 13 families (10 in Lacaune, 1 in Basco-Bearnaise, 2 in Manech blond faced breeds). Family size averaged 32 sons per sire and ranged from 21 to 48. The sons were born from 1992 to 1999 and were progeny tested with 88 daughters on average.

Phenotypic data. Six traits were considered in the analysis: milk yield, fat and protein yield, fat and protein content, and somatic cell score (SCS) for mastitis resistance. The phenotypic unit of measurement was daughter yield deviations (DYD) (VanRaden and Wiggans, 1992), *i.e.* the average of the phenotypes of the daughters of each son adjusted for the environmental effects and additive genetic merit of the daughters' dams, obtained from the French dairy sheep evaluation system (Barillet *et al.*, 1996 ; Rupp *et al.*, 2002).

Marker data. As described in table 1, 20 microsatellite markers were chosen on 7 candidate chromosomes. These regions were selected because their homologous regions in bovine were shown to carry QTL affecting milk traits or SCS. Accordingly, the equivalence between ovine

and bovine chromosome numbers (according to ISCNDA 1989) is indicated in table 1. In fact, as shown by the location and the number of markers per chromosome (2 to 4) with respect to the total length of the chromosomes, only regions of interest and not whole chromosomes were analysed.

Table 1. Description of the partial genotyping

Sheep chromosome (Bovine chromosome)	Total length of the chromosome (cM Haldane)	Number of markers used in the analysis	Length of analysed linkage group (cM Haldane)
OAR5 (BTA7)	150	4	125
OAR6 (BTA6)	154	2	35
OAR9 (BTA14)	126	2	55
OAR11 (BTA19)	118	3	55
OAR15 (BTA15)	111	3	65
OAR16 (BTA20)	80	4	70
OAR22 (BTA26)	85	2	20

Statistical methods. The QTL detection was carried out according to the methodology proposed by Knott *et al.* (1996) and Elsen *et al.* (1999) by within-sire linear regression using the following model :

$$DYD_{ij} = s_i + (2p_{ij}-1)a_i + e_{ij}$$

where s_i was the fixed effect of the sire i , p_{ij} was the probability of inheriting one arbitrarily defined QTL allele from sire i for son j given the marker information, a_i was half the substitution effect of the putative QTL carried by the sire i , and e_{ij} was the residual assumed to be normally distributed with a zero expectation and a heterogeneous variance $\sigma_{e_{ij}}^2/CD_{ij}$. CD_{ij} was the reliability of the proof of the son j , depending of the number of granddaughters and the heritability of the trait.

For each chromosome, the probability for each possible phase of the sires was estimated from progeny marker information. The most likely phase was retained and the probability that each progeny received one or the other chromosomal segment was estimated at every position, given this phase, using a 1 cM step.

The rejection thresholds were estimated by within-family permutations as proposed by Churchill and Doerge (1994), for each trait and each chromosome using 10,000 permutations. QTL findings were reported in three ways, by listing (i) all locations significant at the chromosome-wise $\alpha_c = 0.05$ type-I error level (with 2.1 (= 6 x 7 x 0.05) type-I errors expected by chance under the null hypothesis of no QTL segregating), (ii) all locations of suggestive significance (with one type-I error expected by chance; thus $\alpha_c = 0.036$), and (iii) all locations of experiment-wise significance, the type-I error (α_c) for each chromosome x trait being determined from $\alpha_{exp} = 1 - (1 - \alpha_c)^n$ where α_{exp} represented the experiment-wise significance (0,10) and n was the number of the independent subexperiments. Assuming that n was equal to 4 x 7=28 resulted in $\alpha_c = 0,0036$ which corresponded to about 0.025 false positive per trait, and about 0.70 false positive expected for all traits.

RESULTS AND DISCUSSION

As described in table 2, the 20 microsatellites were informative enough, since the number of heterozygous sires was 8.8 in average for a total of 13 families. The percentage of informative meioses was 70 % in average and ranged from 31 to 90 % according to the markers (table 2).

Table 2. Characteristics of the microsatellites

Sheep chromosome	No. of markers	Markers (distance, cM Haldane)	# heterozygous sires	# genotypes	% informative meioses
OAR5	4	RM6 (25)	10	407	57
		BMS2258 (90)	6	406	31
		MCM527 (120)	6	375	43
		BMS1247 (150)	7	208	79
OAR6	2	BM9058 (10)	11	335	66
		OARAE101 (45)	6	185	76
OAR9	2	CSSM66 (25)	10	292	88
		MCM42 (80)	9	278	50
OAR11	3	HEL10 (20)	4	108	90
		IDVGA46 (55)	9	257	70
		BM17132 (75)	10	336	64
OAR15	3	MAF65 (30)	10	278	45
		BMS2684 (50)	12	364	76
		BMS820 (95)	9	286	62
OAR16	4	BM1225 (10)	9	265	90
		MAF214 (40)	7	210	73
		LS08 (50)	12	378	83
		MCM150 (80)	9	295	79
OAR22	2	BM4505 (40)	10	259	87
		BMS882 (60)	11	338	81
Mean			8.8		70

In agreement with dairy cattle results (Georges *et al.*, 1995 ; Spelman *et al.*, 1996 ; Coppieters *et al.*, 1998 ; Zhang *et al.*, 1998 ; Heyen *et al.*, 1999 ; Boichard *et al.*, 2002), QTL for fat content, protein content and protein yield were found on sheep chromosome 9, 5 and 6, respectively (table 3). The more significant QTL was found for fat content on chromosome 9. On the homologous chromosome 14 in bovine, this QTL was recently identified as a mutation in DGAT1 gene (Grisart *et al.*, 2002). Conversely, two QTL for SCS were detected on chromosomes 6 and 16 in regions differing (to our knowledge) from homologous ones associated with SCS in dairy cattle. Each detected QTL segregated in 2 or 3 families only. The estimated substitution effects ranged from 0.6 to 2 genetic standard deviations, but the highest values were likely overestimated due to limited power of the design.

Table 3. Experiment-wise, Suggestive and chromosome-wise significant QTL

Trait	Chromosome	Closest marker	P	Number of informative families
<i>Experiment-wise significant QTL</i>				
Fat content	OAR9	CSSM66	0.004	3
<i>Suggestive significant QTL</i>				
SCS	OAR6	OARAE101	0.023	2
SCS	OAR16	BM1225	0.009	2
<i>Chromosome-wise significant QTL</i>				
Protein content	OAR5	BMS2258	0.043	3
Protein yield	OAR6	OARAE101	0.039	3
<i>Close to chromosome-wise significant QTL</i>				
Fat yield	OAR6	OARAE101	0.085	3
Protein content	OAR22	BMS882	0.079	2

CONCLUSION

These preliminary results are promising, especially when comparing QTL detection for milk production traits in cattle and sheep. This project is being continued in the two next years by considering other chromosomal regions, new traits (udder score, longevity) and by adding new bigger families, as DNA of 4,000 AI progeny tested rams will be available next year for the French dairy sheep breeds.

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