

Detection on OAR7 of QTL affecting fat and protein yields in dairy sheep

Sara Casu¹, Maria Colombino¹, Giuliana Mulas¹, Stefania Sechi¹,
François Barillet², Antonello Carta¹

¹Settore Genetica e Biotecnologie - DIRPA, AGRIS-Sardegna, Olmedo, Italy

²Station d'Amélioration Génétique des Animaux-INRA-Toulouse, France

Corresponding author: Sara Casu. Settore Genetica e Biotecnologie - DIRPA, AGRIS-Sardegna. Loc. Bonasai SS291 km 18.6, 07040 Olmedo (SS), Italy - Tel. +39 079 3750367 - Fax: +39 079 389450 - Email: scasu@agrisricerca.it

ABSTRACT - The objective of this paper was identifying QTL that affect fat and protein yields in dairy sheep independently of milk yield. Data were collected in an experimental flock of 887 ewes organized in a daughter design. QTL detection focused on OAR7 where 13 microsatellites were available. The genetic abilities to produce fat and protein independently from the ability to produce milk were estimated as the residuals of the regression of EBV for fat and protein yields on EBV for milk yield. One QTL affecting fat yield (CWP=0.00009) and one QTL affecting protein yield (CWP=0.006) were detected. The most probable QTL location was 115.3 cM in the Sheep Best Position Linkage Map Version 4.7 for both traits. No QTL affecting milk yield was detected. The analysis of fat and protein yields independently of milk yield is an effective strategy to identify chromosomal regions affecting milk composition with no detrimental effect on milk yield.

Key words: QTL, Fat yield, Protein yield.

Introduction - Fat and protein are of great importance in determining sheep cheese production. Fat and protein yields affect the total amount of cheese production and are strongly related to milk yield, whereas the corresponding concentrations affect cheese yield. The classical quantitative selection for improving milk composition is difficult due to the negative genetic correlations with milk yield and the high cost of phenotypes recording, especially in sheep. For this reason many efforts have been made in identifying genes responsible of milk composition to implement in MAS or GAS programs. Whatever the selection approach the target, traits to increase cheese yield have always been the fat and protein percentages. These phenotypes resulted from ratios of two basic traits: fat or protein yield and milk yield. This fact explains the classical genetic antagonism between percentages and milk yield. In QTL detection studies, the use of percentages makes difficult to distinguish the effects of found QTL on the basic traits. From a selection point of view, it is a crucial point to increase the fat and protein yields independently from milk yield to avoid any negative consequences on the fat and protein concentrations. The objective of this paper was to find QTL with such feature on ovine chromosome 7.

Material and methods - Data were collected during 4 lactations on 887 Sardinian x Lacaune backcross ewes organized in a daughter design of 10 half-sib families of around 100 ewes/sire (Carta *et al.*, 2008). Milk yield and milk fat and protein yields, measured twice a month during each lactation, were analyzed on a lactation basis. Five milk production traits were computed: milk yield (MY, Kg), fat yield (FY, Kg), protein yield (PY, Kg), fat content (FC, %) and protein content (PC, %). Breeding values for these traits were estimated including in the analysis all available lactation records of ewes related to the target population: backcross ewes, their dams and daughters. In a first step, data of the 3 populations were analyzed separately to adjust for the specific environmental effects (Usai, 2008). Thus 16,437 residuals per trait, corresponding to the original lactations adjusted for fixed effects, were produced. The pedigree

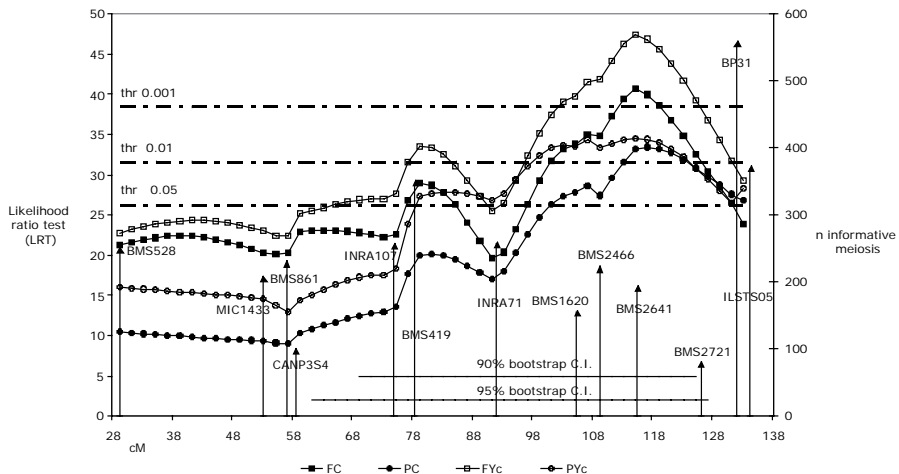
file included 11,927 individuals, born between 1960 and 2004. EBV estimation was performed by a pentaitrait repeatability animal model. EBV of BC ewes were then adjusted for ½ the EBV of their dams. In a daughter design this is equivalent to adjust for the “family effect” as proposed by Aulchenko *et al.* (2007). Finally, EBV for FY and PY were regressed on the EBV for MY. Residuals of such regressions (FYc and PYc) represented the genetic ability to produce fat and protein in milk independently from the genetic ability to produce milk. These residuals, as well as EBV adjusted for dams’ EBV for the 5 original traits, were used as phenotypes for the QTL detection analysis on OAR7.

Genotypings of the BC animals for 13 microsatellites on OAR7 were carried out in multiplex using an ABI PRISM® 3100- Avant Genetic Analyzer (Applied Biosystems, Foster City, CA). To reduce the number of molecular analyses, informative families to genotype for further markers were chosen on the basis of a preliminary QTL detection analysis with only 5 microsatellite genotypes. A specific genetic map was built using all available genotypes with CRI-MAP 2.4 software (Green *et al.*, 1990). QTL detection analysis was carried out following Elsen *et al.* (1999), by a within-sire linear regression of phenotypes on the probability of inheriting one defined QTL allele from the sire given the marker information using a 1 cM step. The chromosome-wise (CW) rejection thresholds were estimated by within-family permutations for each trait using 100,000 permutations (Churchill and Doerge, 1994). The confidence interval of the QTL location was estimated by 10,000 bootstraps (Visscher *et al.*, 1996). Analyses were performed using the QTLmap software of INRA-France.

Results and conclusions - Preliminary analyses carried using 5 markers suggested the presence of a QTL affecting FYc and/or PYc on 6 families, which were then genotyped for the other markers. The 13 analyzed microsatellites are reported in Figure 1.

The found order of markers was the same as the Sheep Best Position Linkage Map Version 4.7, which, finally, was retained for the QTL analysis. The length of the analyzed segment corresponded to 78% of the whole chromosome (148.4 cM). The average distance between markers was 8.7 cM with the longest gap (23.9 cM) at beginning of the explored segment. The number of informative meioses ranged from 556 for BP31 to 78 for BMS2721.

Figure 1. OAR7 map, n. informative meiosis (height of marker arrows), CW significance thresholds; likelihood ratio test profile (LRT) for fat (FYc) and protein (PYc) yields adjusted for milk yield, fat (FC) and protein (PC) contents, bootstrap confidence intervals.



The final analyses produced a significant LRT value either for FYc and PYc or FC and PC (Table 1). The most significant result was found for FYc (CWP=0.00009). The most probable location was at 115.3 from the beginning of the chromosome. The QTL segregates in 5 families with estimated allelic substitution effects ranging from 0.18 to 0.43 Kg (Table 1). Also the analysis of FC indicates the presence of a QTL in the same position, although with a lower significance level (CWP=0.0005). This QTL was significant in 4 out of the 5 families. The LRT of PYc and PC showed a significant peak in approximately the same position. The QTL for PYc segregates in 4 families, 3 out of which were also significant for FYc. Available data only gave limited resolution of the QTL locations with 90% bootstrap confidence interval spanning 56 cM for the most significant one. Fifty-seven percent of bootstrap locations however fell in a 20 cM interval.

No QTL were detected on MY, FY, and PY. In particular the LRT peak for milk yield was far from significance (CWP=0.63), supporting that the found QTL only affects milk composition. These results confirm that the analysis of fat and protein yields adjusted for milk yield is an effective strategy to identify chromosomal regions affecting the ability to produce fat and protein independently from the ability to produce milk.

Table 1. Likelihood ratio test (LRT), Chromosome Wise significance level (CWP), most probable locations from the origin of the chromosome (pos) and QTL effects.

Trait	LRT	CWP	pos	Informative families	Allelic substitution effect (range)			
					raw values	p.s.d. units	raw mean units	raw s.d. units
FYc	47.38	.00009	115.3	1-4-6-8-9	0.18-0.43 kg	0.24-0.59	0.01-0.02	0.03-0.08
PYc	34.44	.006	115.3	1-4-5-6	0.13-0.18 kg	0.34-0.48	0.01-0.01	0.03-0.04
FC	40.74	.0005	115.3	1-6-8-9	0.08-0.21%	0.26-0.68	0.01-0.03	0.15-0.38
PC	33.44	.005	117.3	4-5-6	0.06-0.10%	0.36-0.55	0.01-0.02	0.17-0.27

FYc: fat yield adjusted for milk yield; PYc: protein yield adjusted for milk yield; FC: fat content; PC: protein content; p.s.d.: standard deviation of the phenotype used for the QTL detection.

The potential selection for this QTL is expected to enhance fat and protein yields and concentrations with no detrimental effect on milk yield.

This work was funded by MIPAF in the framework of the «SelMol» project.

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