Quantitative Trait Loci Related To Wool Quality In Merino Sheep

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Introduction

An analysis of QTL detection was realized in a Merino sheep resource population. The aim of this work was the search for loci influencing wool quality, important goals in Merino breeding programs. The purpose of this paper is to confirm and improve the identification of QTLs that affect wool traits and fleece characteristic in this sheep population resource. Here, we extended the analysis performed by Bidinost et al. (2008) by two additional families and genotypes markers on chromosomes ovine (OAR) 3 and 11, and we analyzed these new data by testing several hypotheses.

Material And Methods

Experimental design. The data resource comprised 10 paternal half-sib families of Merino breed which were part of a sire-reference genetic evaluation. Neither sires nor dams were related to each other. Lambs were born on three farms. Family size averaged 61.7 offspring ranging from 30 to 88 per sire. Wool production and wool quality traits were recorded on sire's progeny. At the age of 11 and 23 months old (first and second shearing, respectively) the following traits were recorded : clean fleece weight (CFW; kg), greasy fleece weight (GFW; kg), clean wool yield (YLD; %), mean fibre diameter (FD; µm), coefficient of variation of FD (CVFD; %), average curvature of fibre (CF; ^{0}/mm), staple length (SL; mm) and staple strength (SS; N/ktex).

Our linkage analysis covered specific regions on OAR3 (13 microsatellites from 179.4 to 215.9 cM region) and OAR11 (9 markers covering from 66.6 cM to 77.8 cM), with an average distance between markers of 2.8 and 1.2 cM, respectively. All information about this discovery of the markers, including the references, are given after the website URL http://www.ncbi.nlm.nih.gov/genome/guide/sheep/. DNA extraction and PCR conditions have been early described (Bidinost et al. 2008).

Statistical analyses. The phenotypic data were pre-corrected by three fixed effect (flock, sex and litter size). Residuals from this analysis were the data vectors used for QTL mapping. The QTL analyses were performed by multi-marker regression (Elsen et al. 1999) using INRA QTLMap software with the following model: $y_{ij} = s_i + (2p_{ij} - 1)a_i + e_{ij}$

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where y was the individual phenotype adjusted as described above, s is the sire, p was the probability of inheriting one defined QTL allele from sire i for the daughter j given the marker information, a 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 between sire heterogenous variance σ^2_{ei}. Significance thresholds were determined through 1,000 within-family phenotype permutations (Churchill and Doerge, 1994). The conservative genome-wide thresholds were derived from chromosome-wide significance levels, following Bonferroni procedure. The 95% confidence intervals of the QTL locations were estimated by LOD drop-off (Lander and Botstein, 1989).

We tested three hypotheses: a single QTL controlling a single trait, two linked QTLs controlling a single trait, and a single QTL controlling more than one trait. To test the existence of a pleiotropic QTL, traits were grouped according to two criteria. The first assembled traits showing significant evidence of a QTL in a single QTL analysis (hereafter referred to as BSQ). The second criterion, hereafter referred to as BPC, assembled traits that were phenotypically correlated.

Results And Discussion

Single Trait methods. QTLs for three traits were detected on OAR11 (Table 1). However, no QTL was identified on OAR3. Results of the tests of the "one QTL" vs. "two QTLs" hypothesis for the single trait analysis are summarized in Table 1. Three situations were observed with varying levels of significance in three tests (no QTL vs. one QTL, one QTL vs two linked QTL, no QTL vs. two-linked-QTL). In the first situation, there was a significant effect of QTL in both the one and two-linked-QTL vs. 0 QTL tests but results of the one-QTL vs. two-QTL test were not significant (SS_1). In the second situation, occurring for $CVFD_2$, the results of the 0 vs. one-QTL, one vs. two-QTL and 0 vs. two-QTL hypotheses were all significant. However (i) the location of one of the two QTLs was identical to the location of the QTL in the single-QTL analysis; (ii) the most significant families (no shown) displayed QTL effects of opposite signs. These two observations suggest that the second QTL could be a statistical artefact. In the last situation two putative were revealed QTLs (for GFW_1 and for YLD_2, test 0 QTL vs. 2 QTL) while no QTL was detected in the single-QTL analysis. QTL substitution effects with opposite signs were observed for seven of 10 families (no shown), suggesting that the QTL effects were masked in the single-QTL analysis. Nevertheless, the same two-QTL analysis in YLD_2 excluding family 10, rejected the null hypothesis (there are no QTLs) at the 5% chromosome-wise level.

Table 1. Results of QTL analyses under a single QTL and two-linked-QTL hypotheses for body weight and wool traits in the single trait model for OAR11.

Trait[1]	Hypothesis test					
	0 QTL vs. 1 QTL		1 QTL vs. 2 QTL		0 QTL vs. 2 QTL	
	Position (cM)	LRT^{x}_{l} [2]	Positions (cM)	LRT^{x}_{l} [2]	Positions (cM)	LRT^{x}_{l} [2]
SS_1	66.60	28.80 [a**]	67.60-71.60	13.54	67.60 - 71.60	42.37 [a*]
$CVFD_2$	67.60	31.20 [a**]	66.60-67.60	17.95 [a*]	66.60 - 67.60	49.15 [a***]
GFW_1	66.60	14.10	68.60-77.60	23.83 [a**]	68.60 - 77.60	37.92 [a**]
YLD_2	68.60	14.30	68.60-70.60	21.95 [a**]	68.60 - 70.60	36.21 [a*]

[1] SS=staple strength; CVFD=coefficient of variation of FD; GFW=greasy fleece weight; YLD=clean wool yield. Indexes: 1=first shearing; 2=second shearing.
[2] LRT^{x}_{l}: Maximum likelihood ratio for each trait *l* for *x* locus. Significance level: [a] **c**hromosome-wide. * p<0.05**;** ** p<0.01; *** p<0.001

Multiple Trait method. Table 2 shows the results of the single pleiotropic QTL test. One QTL was detected that simultaneously affected SS_1 and $CVFD_2$ (under significant one-QTL results), and FD_2, $CVFD_2$ and CF_2 (under BPC assembling) at 67.60 cM. The likelihood profile for the joint analysis was similar to the $CVFD_2$ profile for which a QTL was detected at position 67.60 cM with both models single and multitrait models.

Confidence interval for those QTLs were flanked by the LSCV36 (at 66.6 cM) and BMS501 (at 77.7 cM) markers. Keratine and keratine-associated proteins genes (KRT1, KAP1 and KAP3) have been mapped from 71.9 cM to 73.4 cM on OAR11 (McLaren et al. 1997). Those genes could be responsible for these detected QTL signals. Consistently, in the Romney breed, Rogers et al. (1994) identified a QTL for KAP1.1, KAP1.3 (formerly known as B2A and B2C, respectively) and KRT1.2 using a candidate gene approach. We found that the same QTL that primarily affects certain traits also likely exerts secondary effects on other related traits.

Table 2. Results of QTL analyses in the multiple trait model for the significant traits detected in single trait analysis and for the correlated traits.

Traits[1]	Trait selection criteria[2]	Position (cM) [3]	LRT^{x}_{1}	LRT^{x}_{1} Threshold[4]
SS_1 $CVFD_2$	BSQ	67.60 [66.60-68.76]	62.55	59.81 [a**]
FD_2 $CVFD_2$ CF_2	BPC	67.60 [66.95-68.85]	54.25	51.33 [a*]

[1] SS=staple strength; FD= Fibre diameter; CVFD=coefficient of variation of FD; CF= average curvature of fibre. Indexes: 1=first shearing; 2=second shearing.
[2] Criteria to select the traits for the multitrait analysis. BSQ: Based on the significant QTL analysis; BPC: Based on the phenotypic correlation.
[3] Position and interval confidence in cM.
[4] Significance level: [a] chromosome-wide; * $p<0.05$; ** $p<0.01$

Conclusion

One-QTL hypothesis is the most common strategy for QTL mapping. This was the case for experiments dealing with several breeds and commercial sheep crosses based on the single wool trait,. The present study demonstrates the utility of other strategies. Although selection for wool traits (decrease in fibre diameter and increase in fibre length and strength) has been applied in the experimental lines used in this work, the QTL detected for several wool traits, tends to show that part of the genetic variance can be explained by the segregation of QTL of medium to large effects.

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