



Functional investigation of a QTL affecting resistance to *Haemonchus contortus* in sheep

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Loci underlying variation in nematode resistance in three sheep populations: a joint analysisV. Riggio¹, R. Pong-Wong¹, G. Sallé², M.G. Usai³, S. Casu³, C. Moreno², O. Matika¹ and S.C. Bishop¹¹The Roslin Institute and R(D)SVS, University of Edinburgh, Easter Bush, EH25 9RG Midlothian, United Kingdom, ²INRA, SAGA, BP 27, 31326 Castanet-Tolosan, France, ³SGB, AGRIS Sardegna, loc. Bonassai, 07040 Olmedo, Sassari, Italy; oswald.matika@roslin.ed.ac.uk

Gastrointestinal nematode infections are one of the main health/economic issues in the sheep industry. Indicator traits for resistance such as faecal egg count (FEC) are commonly used in genomic studies; however published results are inconsistent among breeds. Meta (or joint) analysis is a tool for aggregating information from multiple independent studies. The aim of this study was to identify loci underlying variation in FEC, as indicator of nematode resistance, in a joint analysis, using data from three populations (Scottish Blackface, Sarda × Lacaune, and Martinik Black-Belly × Romane), genotyped with the ovine 50k SNP chip. The trait analysed was the average animal effect for Strongyles FEC data (estimated by fitting a repeatability model across different time points for each population). Analyses were performed using two different software packages: Regional Heritability Mapping (RHM) and QTLMap, which make contrasting assumptions when identifying QTL. After editing, a total of 4,123 and 2,644 animals were available for RHM and QTLMap analyses respectively; after quality control 38,991 SNPs (common across breeds) were available for both analyses. RHM identified significant regions on OAR4, 19 and 20, with the latter being the most significant. The OAR20 region is close to the Major Histocompatibility Complex, which has often been proposed as a functional candidate for nematode resistance. This region was significant in only one of the three individual breeds. The most significant region identified from QTLMap was on OAR12. This result was confirmed by RHM, when using the reduced dataset edited for QTLMap. These regions will be investigated in other breeds to validate the results. These results are obtained through the EC-funded FP7 Project 3SR-245140.

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In previous studies using the ovine 50K SNP chip, a region of OAR12 was found to be associated with variation in fecal egg count (FEC) during *Haemonchus contortus* infection in both naive and primed back-cross (BC) lambs. Our study aimed at investigating the functional properties of this QTL region. Based on the linkage analysis results, BC sheep were selected for different haplotypes in a 20-Mbp-wide QTL region, and mated together to produce BC×BC progenies carrying two favorable or unfavorable QTL alleles. Subsequent to this marker-assisted mating, an association analysis pinpointed a 4-SNP haplotype. Based on the 4-SNP region, 61 BC×BC lambs were selected and experimentally infected with 10,000 *H. contortus* larvae. FEC were determined every three days from 18 days post-infection (dpi) and blood cell populations were determined at 0, 14 and 27 dpi. After one month, sheep were sacrificed to determine worm burden and worm fertility and to sample tissues for gene expression analysis. Significant differences in FEC and hematocrit drop were found. In addition, the female worms recovered from predicted resistant sheep were less fecund (250 eggs/female less). A gene expression analysis was performed between carriers of two 4-SNP alleles with opposite effects. A 4-fold over-expression was found for IL4 and IL13 genes in the abomasal mucosa of the sheep carrying the favorable QTL allele, indicating a higher Th2-biased cytokinic environment. Still, no functional candidate gene underlying the QTL region has been validated so far.

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