Fine Mapping Of A QTL Affecting Backfat Thickness On SSC1 Using A Locally Congenic Backcross Design.

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Introduction

The development of molecular tools in livestock species has spurred efforts to identify QTLs for economically important traits using experimental designs. Numerous QTLs have been identified, but in most cases, mapping resolution has been rather poor. Different strategies for QTL fine mapping have been proposed (Darvasi (1998)). In mice and plants successful fine-scale recombination mapping of QTL has been obtained by using congenic strains. In pigs and in other livestock species, genetically homogenous lines do not exist. Nevertheless, recombinant backcross (BC) progeny testing has successfully been used for the fine mapping of QTL detected in experimental crosses between domestic European populations and either Wild Boar or Meishan animals. However this strategy is based on two important assumptions. First, the two breeds are supposed to be each fixed for a different QTL allele (q or Q) associated with different phenotypic values. Second, the introgressed allele is supposed to be a (co)-dominant allele. Among recently detected QTL, significant effects for growth, backfat depth, carcass, and meat quality traits have been identified in the telomeric region of the long arm of SSC1 in different F2 crosses between Meishan (MS) and Large White (LW) breeds (Rohrer and Keele (1998); De Koning et al. (1999); Paszek et al. (1999); Rohrer (2000); Bidanel et al. (2001); Milan et al. (2002)). A higher fat depth and a larger growth rate have been reported for the allele of MS origin. However, the LW allele showed partial dominance effects over the MS allele for growth and fatness traits. In this situation progeny testing between recombinant sires and pure LW dams is not optimal. The aim of the present project was to fine map SSC1 QTL, using an experimental BC design miming locally genetically identical strains used in mice.

Material and methods

Animals and measurements. A backcross (BC) QTL fine mapping program was initiated using a F1 LWxMS boar from the PorQTL mapping design (figure 1). At each generation, boars carrying a distinguishable recombinant chromosome in the confidence interval of the QTL were progeny-tested by mating them to an average of 12 sows at the INRA GEPA experimental unit. Pure LW sows were used in a first step, then a population of experimental

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dams with a 29/32 LW genetic background. These females were obtained by intercrosses between BC sires and dams and were selected to be locally homozygous in the QTL region for the MS haplotype carried by the F1 founder boar. In all cases, offspring were weighed at birth, at the beginning and at the end of the performance test. At about 120 and 140 days, ultrasound backfat thickness (BF) were measured at 4 cm from the middorsal line (on each side) at the shoulder (neck), the last rib (back), and the hip joint (rump). In total 6 different BC sires have been progeny tested using on average 100 measured offspring: 3 BC2 and 1 BC3 were used to refine the QTL interval; 2 additional BC4 were progeny tested to validate this interval. Recombinant chromosomes of these boars are presented in figure 2.
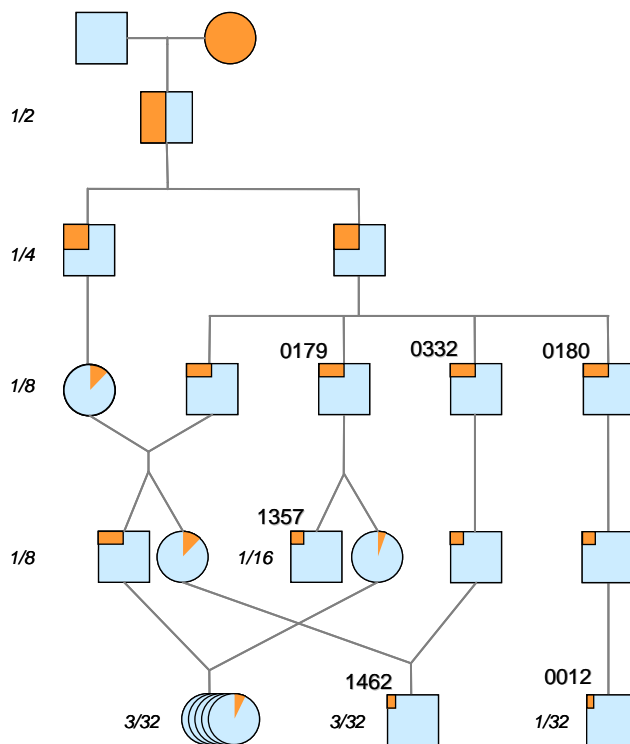

Figure 1: Structure of the SSC1 BC pedigree. The LW proportion of the genome is indicated in blue, the MS proportion in orange and by an italic numbers. Only BC females used as dams are indicated (in the other cases dams were LW females).

Genotyping and statistical analyses. Sires, dams and their offspring were genotyped for 9 to 12 microsatellite markers located from 115 to 145 cM depending on the boar family. Genotypes were obtained as described by Sanchez et al. (2004). All traits were adjusted for the fixed effects of sex and batch with the GLM procedure of SAS (SAS Inst., Inc., Cary, NC). BF was also adjusted to a constant weight by adding it as a linear covariate in the model. Adjusted data were then submitted to interval mapping analyses using the QTLMAP

software developed at INRA as previously described by Sanchez et al. (2004). At the location with the highest L-ratio, a gene substitution effect (a) was estimated within each sire family.

Results and discussion

A QTL with large effects on backfat thickness was located on chromosome 1 in a rather large initial localisation interval (between 115 and 145 cM).

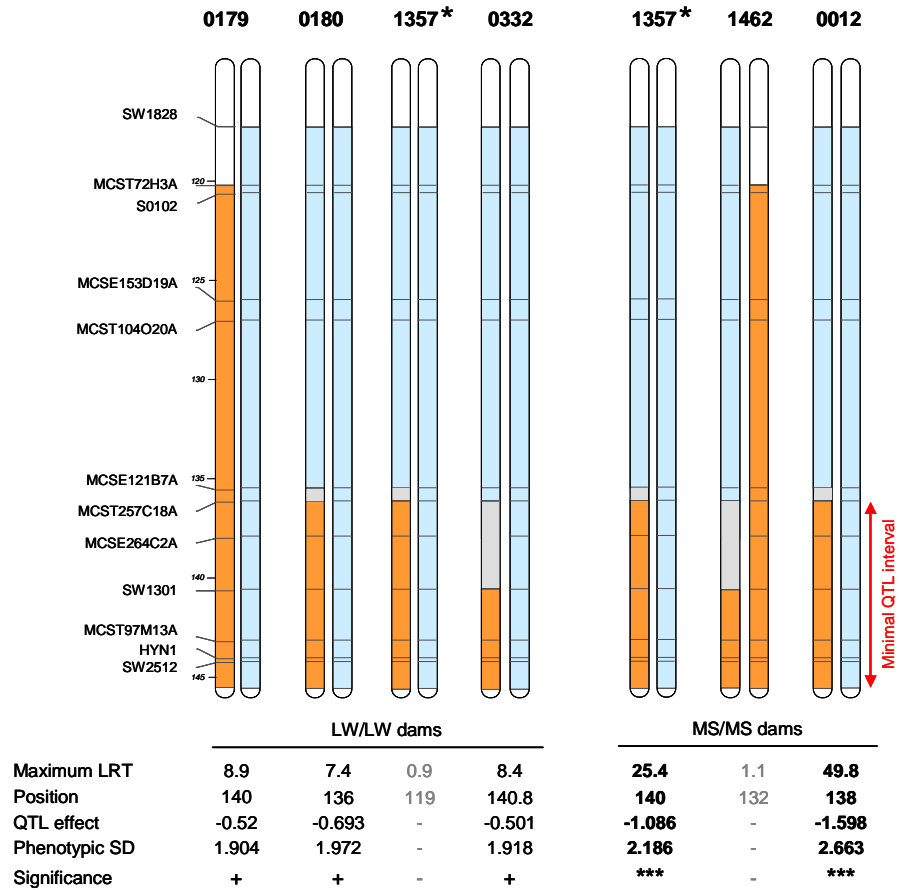|	0179	0180	1357*	0332	1357*	1462	0012
	LW/LW dams				MS/MS dams		
Maximum LRT	8.9	7.4	0.9	8.4	25.4	1.1	49.8
Position	140	136	119	140.8	140	132	138
QTL effect	-0.52	-0.693	-	-0.501	-1.086	-	-1.598
Phenotypic SD	1.904	1.972	-	1.918	2.186	-	2.663
Significance	+	+	-	+	***	-	***

Figure 2: Haplotypes of the different recombinant sires progeny tested. Breed origins are differentiated by colors (MS: orange, LW: blue, or unknown: grey). Maximum likelihood ratio test value (Maximum LRT), corresponding significance level and position (cM) are given for backfat thickness depending on the dam genotypes in the chromosomic region of interest (LWLW or MSMS). Average QTL substitution effects was estimated within sire families. *: same boar tested on different dams

Three BC2 sires (0179-0180-0332), found to be recombinant within the confidence interval of the QTL, were mated to LW dams in order to test the segregation of QTL alleles in their progeny. Significant likelihood ratio test values were obtained for the three BC2 sires at a 5% chromosome wide level for backfat trait. Additionally, sire 1357 (a BC3 boar) was also progeny tested. Conversely, test values were not significant for any trait when progeny testing was realised using pure LW dams. This result was not in accordance with results obtained with the BC2 sires, #1357 being heterozygous MS/LW in the previously defined interval. The partial dominance of LW over MS alleles, inducing a low experimental power, was a potential explanation for these inconsistent results. A much higher power would be obtained from backcrosses with MS dams, but phenotype measurement is complicated by the early puberty of crossbred MS pigs and it is more expensive. In order to overcome these difficulties, sire 1357 was progeny tested again using a population of dams with a 29/32 LW genetic background but locally MS/MS (i.e. homozygous for the recessive allele of the QTL). Highly significant results (0.1% at the genome wide level) were obtained for several traits affecting backfat depth. Based on the progeny testing data obtained, the localisation interval of the QTL affecting fatness of the animal was decreased to approximately 10 cM, limited by MCST257C18A and SW2512 markers. This minimal interval (136-145 cM) was later confirmed by progeny testing additionnal BC4 sires with locally MS/MS dams: no significant likelihood ratio tests values were obtained for a BC sire (1462) homozygous MS/MS in the QTL region, and highly significant results (0.1% at the genome wide level) were obtained with an additional heterozygous BC sire (#0012) (figure 2).

Conclusion

We have further characterized the fatness QTL on pig chromosome 1 and refined its map position to a 10 cM interval, using a locally congenic BC design. We obtained highly significant results and overcame the loss of power due to the dominance of the LW allele. Additional advanced BC families are being produced to further refine the QTL localization using the same design.

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