A CANDIDATE REGION APPROACH ALLOWS EFFICIENT QTL DETECTION IN UK SUFFOLK AND TEXEL POPULATIONS

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INTRODUCTION

Genomic research and the detection of quantitative trait loci (QTL) provide tools to enhance genetic progress and improve understanding of the biology of commercially important traits. In the UK, the sire referencing schemes (SRSs) in terminal sire sheep breeds establish large half-sib families and provide a unique opportunity to investigate QTL segregation within commercial populations

Several previous studies have indicated the presence of a major gene (or genes) for production traits in sheep or other mammalian species. These include growth effects on sheep chromosome 1 around the transferrin gene (Kmiec 1999), muscling effects on sheep chromosome 2 around the myostatin gene (e.g. Broad *et al.*, 2000) and growth effects around IGF1 (located on sheep chromosome 3) in cattle (Stone *et al.*, 1999). The leptin gene (located on sheep chromosome 4) has been extensively studied in numerous species and recognised as a major gene controlling fat deposition. The calpastatin gene (located on sheep chromosome 5) interacts with the callipyge gene to affect muscling (Freking *et al.*, 1999). Previous studies in sheep and of the homologous region in cattle have highlighted a locus affecting growth on sheep chromosome 6 (Walling *et al.*, 2000 and Casas *et al.* 2000 respectively). Associations have been shown between GH1 (located on sheep chromosome 11) and cattle growth (Taylor *et al.* 1998). Chromosome 18 contains the Callipyge gene (Cockett *et al.,* 1994) and the rib eye muscling locus (Nicholl *et al.*, 1998). Finally the major histocompatibility complex (MHC) is located on sheep chromosome 20. This is a very gene rich area of the sheep genome and contains many candidate genes. Studies in cattle (Elo *et al.* 1999) and pigs (Walling *et al.* 1998) have found effects for growth and fatness in homologous regions of their genomes.

The aim of this study was to determine whether there was evidence for QTL affecting growth, muscling or fatness, in these previously defined chromosomal regions, in large half-sib families of the UK terminal sire sheep SRSs.

MATERIAL AND METHODS

Animals. Eight widely used rams, three Suffolk (S1-S3) and five Texel (T1-T5) were identified for investigation based on numbers of accessible progeny with breeding, weaning and ultrasonic scanning records. All animals were born and reared in commercial flocks across the UK. Blood samples were collected at 6 months of age. DNA was extracted from the blood using a standard salt extraction method on fresh samples and a phenol-chloroform extraction on blood samples that had been frozen. Each animal was weighed at 8 weeks of age (8WW)

and at ultrasonic scanning (ScanWT) at approximately 20 weeks of age. At scanning, muscle depth (Mus) and fat depth (Fat) at the third lumbar were also recorded. Additionally, both muscle depth and fat depth traits were phenotypically adjusted to correct for body weight (MusWT and FatWT respectively).

Genotyping. All Suffolk sires were genotyped for markers across all candidate chromosomal regions outlined above, i.e. on chromosomes 1, 2, 3, 4, 5, 6, 11, 18 and 20. Texel sires were genotyped for markers across all regions except chromosomes 1 and 6. All offspring were subsequently genotyped for all markers that were heterozygous in their sire, with an average of 6.56 informative typed markers per chromosomal region per family. Linkage maps were produced using Cri-Map (Green *et al.* 1990) and were in close agreement with previous studies (Maddox *et al.*, 2001).

Statistical Analysis. Initially, the six phenotypic measurements (8WW, ScanWT, Mus, Fat, MusWT and FatWT) were corrected for fixed effects and covariates estimated using ASREML (Gilmour *et al.*, 1999) from data collected on all farms in the SRSs over the last decade. All traits were corrected for the fixed effects of flock-year, sex, birth-rearing rank and age of dam. In addition, ultrasonic scanning traits were also corrected for age at scanning. Secondly, the probability of inheriting a particular sire chromosome at a particular position was calculated for each offspring from the genotype data at 1cM intervals, using the method of Knott *et al.* (1996) along each of the chromosomes. Finally each of the adjusted phenotypes was regressed on the inheritance probabilities, at each location, on each chromosome, for each family. For each regression an F-ratio of the model including the phase probability versus the same model without the phase probability was calculated. The location with the largest F-ratio was taken to be the best estimated position for a QTL in each family, for each trait.

Thresholds. Following the recommendations of Lander and Kruglyak (1995) it is worth reporting all regions with a nominal P value of $P<0.05$, but without any claims of linkage. To reduce the number of false positive results, whilst reporting information that may be of benefit to other researchers this study reports all regions with a nominal P value of $P<0.01$.

Confidence Intervals. Confidence intervals (CIs) were constructed for each significant effect using the bootstrap approach (Visscher *et al*. 1996). One thousand bootstrap resamples were used. Removing the top and bottom 2.5% of the resampled positional estimates created the estimated 95% confidence interval for each QTL in each family.

RESULTS AND DISCUSSION

Significant effects were detected in 6 different chromosomal regions. These are summarised in Table 1. Effects varied between 0.5-0.8 phenotypic standard deviations. Four of the regions contained effects only detected in individual sires. In contrast, the effects on chromosomes 2, 18 and 20 were supported by data from 2 or 3 different sires. The data on chromosome 2 suggests 2 QTL (results not shown), one at ~60cM affecting muscle growth and another at ~170cM affecting fat growth. The fat QTL on chromosome 2 corresponds to the region of the myostatin locus responsible for double muscling in cattle. The effects on chromosome 18 on muscle in T2 are almost identical to the description of the rib eye muscling locus, both in size

of effect and chromosomal position, possibly suggesting that a similar allele to the Carwell allele at the rib eye muscling locus is segregating in UK Texel sheep. It also coincides with the known Callipyge location.

In agreement with previous studies in other mammalian species, a family from both the Texel and Suffolk populations produced evidence for a locus close to, or within, the MHC region affecting fatness. In other regions one Texel family produced evidence for a locus affecting fatness on sheep chromosome 4 near the leptin gene, an effect on muscularity was observed in a Suffolk sire on chromosome 1 and effects on both fat and muscle were seen in Suffolk and Texel families respectively.

Table 1. Summary of significant effects at the 1% nominal level[A]

Chromosome	Sire	Progeny	Trait	Position	95% CI	Effect (se)	F
1	S1	95	Mus	200	32-224	2.28 (0.729)	9.77
		95	MusWT	200	6-299	1.62 (0.547)	8.78
2	T1	115	Fat	169	53-170	0.62 (0.170)	12.85
		115	FatWT	169	5-260	0.51 (0.160)	9.94
	T2	105	MusWT	59	5-141	1.24 (0.463)	7.14
	T4	70	8Week	167	5-169	1.93 (0.617)	9.78
		70	ScanWT	165	8-169	2.74 (0.997)	7.53
3	S2	228	FatWT	80	2-85	0.44 (0.154)	7.96
	T3	186	Mus	13	0-83	1.54 (0.433)	12.64
		186	MusWT	18	5-69	1.25 (0.340)	13.47
4	T2	104	FatWT	43	0-43	0.75 (0.221)	11.44
18	S1	95	8Week	98	67-98	2.59 (0.759)	11.69
	S2	229	8Week	38	0-81	1.52 (0.572)	7.05
		229	ScanWT	44	0-77	2.10 (0.754)	7.77
	T2	96	Mus	85	34-98	1.82 (0.554)	10.79
		96	MusWT	88	15-98	1.26 (0.441)	8.16
	T5	94	Fat	75	47-92	0.77 (0.289)	7.02
		94	FatWT	76	20-92	0.69 (0.241)	8.19
20	S1	89	Fat	54	2-88	0.45 (0.159)	7.97
	T1	115	Fat	49	1-71	0.59 (0.182)	10.44

[A]Effects given in kg for weight traits and mm for fat and muscle traits

CONCLUSION

The study has been successful in detecting QTL in large half-sib commercial populations and has highlighted segregation of several major loci segregating in UK terminal sire sheep sire referencing schemes. The chromosomal candidate region approach is useful in populations where traditional QTL mapping methods e.g. diverse breed or line crosses, may not be feasible or appropriate. Importantly, it shows the generic applicability of results across breeds. Future work aims to improve the resolution of position and effects detected in this study.

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