Mapping of quantitative trait loci for growth and carcass traits in commercial sheep populations[1]

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ABSTRACT: Quantitative trait loci analyses were applied to data from Suffolk and Texel commercial sheep flocks in the United Kingdom. The populations comprised 489 Suffolk animals in three half-sib families and 903 Texel animals in nine half-sib families. Phenotypic data comprised measurements of live weight at 8 and 20 wk of age and ultrasonically measured fat and muscle depth at 20 wk. Lambs and their sires were genotyped across candidate regions on chromosomes 1, 2, 3, 4, 5, 6, 11, 18, and 20. Data were analyzed at the breed level, at the family level, and across extended families when families were genetically related. The breed-level analyses revealed a suggestive QTL on chromosome 1 in the Suffolk breed, between markers BM8246 and McM130, affecting muscle depth, although the effect was only significant in one of the three Suffolk families. A two-QTL analysis suggested that this effect may be due to two adjacent QTL acting in coupling. In total, 24 suggestive QTL were identified from individual family analyses. The most significant QTL affected fat depth and was segregating in a Texel family on chromosome 2, with an effect of 0.62 mm. The QTL was located around marker ILSTS030, 26 cM distal to myostatin. Two of the Suffolk and two of the Texel sires were related, and a three-generation analysis was applied across these two extended families. Seven suggestive QTL were identified in this analysis, including one that had not been detected in the individual family analysis. The most significant QTL, which affected muscle depth, was located on chromosome 18 near the callipyge and Carwell loci. Based on the phenotypic effect and location of the QTL, the data suggest that a locus similar to the Carwell locus may be segregating in the United Kingdom Texel population.

Key Words: Fat, Genome, Growth, Muscle, Suffolk, Texel

 J. Anim. Sci. 2004. 82:2234–2245

Introduction

Genomic research and the identification of quantitative trait loci help improve our understanding of the underlying biology of a specific trait. Following the successful detection of QTL in experimental populations, studies have attempted to detect QTL in commercial populations. Several studies have applied methodology to centralized dairy cattle records resulting in QTL for milk traits (e.g., Georges et al., 1995). Recently, QTL were reported in commercial pig populations (Nagamine et al., 2003). The benefits of using commercial populations are the considerable economic and time savings, in terms of the time to implementation that can be achieved. In addition, animals in national improvement schemes have good phenotypic and pedigree records suitable for analysis, and results can be used in breed improvement schemes. Additional studies may not be necessary to transfer the research findings because the results are relevant to the target population.

Compared with other livestock species, there is less published literature on QTL identification in sheep populations. Surprisingly few QTL have been published for traits of direct relevance to meat production, apart from studies of individual major genes, such as the callipyge locus (Freking et al., 2002) and other candidate genes.

In the United Kingdom, sire-referencing schemes (**SRS**) in terminal sire sheep breeds create ideal populations for QTL detection. Through widespread use of artificial insemination with semen from high-genetic-merit sires, they create large half-sib families. More-

[1]This work was funded by the Dept. for Environment, Food, and Rural Affairs (Defra), Scottish Executive Environment and Rural Affairs Dept. (SEERAD), and the Meat and Livestock Commission (MLC) through the LINK Sustainable Livestock Production Programme. We are grateful for the large inputs made by Elite Texel Sires (United Kingdom) Ltd. and Suffolk Sire Referencing Scheme Ltd. We also acknowledge assistance from MLC Signet Breeding Services and Edinburgh Genetics. Lastly, we thank the two anonymous referees for useful insights.

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Received August 25, 2003.

Accepted April 30, 2004.

Table 1. Summary of the families used in the study and the regions investigated

Breed	Family	Sire	No. of progeny	Chromosomes investigated
Suffolk	S1	25	98	1, 2, 3, 4, 5, 6, 11, 18, 20
	S2	561	276	1, 2, 3, 4, 5, 6, 11, 18, 20
	S3	1973	115	1, 3, 6, 18, 20
Texel	T1	1	130	2, 3, 4, 5, 11, 18, 20
	T2	48	199	2, 3, 4, 5, 11, 18, 20
	T3	420	75	2, 18
	T4	421	70	2, 3, 4, 18, 20
	T5	422	61	2, 3, 4, 18, 20
	T6	424	115	2, 3, 4, 5, 11, 18, 20
	T7	427	104	2, 3, 18, 20
	T8	459	87	2, 3, 4, 18, 20
	T9	525	62	2, 3, 4, 18, 20

over, live weight measures and ultrasonic measurements of muscle and fatness are standardized across farms and centrally collated. Such data provide a unique opportunity to investigate segregation of QTL relevant to meat production within commercial populations. This study aimed to determine whether there is evidence for the segregation of QTL affecting growth, muscling, or fatness in the large half-sib families of the United Kingdom terminal sire sheep SRS.

Materials and Methods

Animals

Twelve half-sib families in the Suffolk and Texel SRS were identified for investigation based on numbers of accessible progeny with breeding, live weight and ultrasonic scanning records. The twelve comprised three Suffolk (**S1** to **S3**) families and nine Texel (**T1** to **T9**) families. On average, families contained 116 offspring, with a range from 61 to 276 progeny. A summary of the families is presented in Table 1. All animals were born and reared in commercial flocks across the United Kingdom.

Each animal was weighed at 8 wk of age (**8WW**) and at ultrasonic scanning (**ScanWT**) at approximately 20 wk of age. At scanning, muscle depth (**Mus**) and fat depth (**Fat**) at the third lumbar were recorded. In the analyses, described below, both Mus and Fat traits were also phenotypically adjusted to correct for body weight (**MusWT** and **FatWT**, respectively).

Blood samples were collected at approximately 6 mo of age. For fresh samples, DNA was extracted from the blood using a standard salt extraction method and a phenol-chloroform extraction was used on blood samples that had been frozen. For sires, DNA was available from either blood or semen samples. Blood samples were not collected from dams; hence, dams were not genotyped.

Selection of Genomic Regions

Several previous studies have indicated the presence of a major gene (or genes) for production traits, relevant to growth and carcass 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 is recognized as a gene with a major influence on 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 (Walling et al., 2000) and on the homologous region in cattle (Casas et al., 2000) have highlighted a locus affecting growth on sheep chromosome 6. Associations have been shown between GH1 (located on sheep chromosome 11) and cattle growth (Taylor et al., 1998). Chromosome 18 contains the callipyge gene (Freking et al., 2002) and the Carwell longissimus muscle (ribeye) muscling locus (Nicoll et al., 1998). Finally, the major histocompatibility complex (**MHC**) is located on sheep chromosome 20. Studies in cattle (Elo et al., 1999) and pigs (Walling et al., 1998a) have found effects for growth and fatness in the homologous MHC regions of their genomes.

Each of these regions was chosen for further study. Initially, the regions described on chromosomes 2, 3, 4, 5, 11, 18 and 20, were investigated in Texel and Suffolk lambs, and subsequently the regions on chromosomes 1 and 6 were investigated in the Suffolk animals only.

Genotyping Strategy

Informative marker panels were developed separately for each sire, in up to nine of the selected candidate regions of the sheep genome (Table 1). This was achieved by initially genotyping each sire for all available microsatellite markers across each candidate region and then selecting heterozygous markers at approximately 10-cM intervals wherever possible. All offspring were subsequently genotyped for selected markers that were heterozygous in their sire. Relative marker locations were verified by producing linkage maps using Cri-Map (Green et al., 1990). These were

in close agreement with previous studies (Maddox et al., 2001), indicating accurate genotype data. Given the comprehensive nature of the Maddox et al. (2001) data set, marker orders and their relative positions were those published by Maddox et al. (2001).

Data Analysis

Preparation of Phenotypic Data. The six phenotypic measurements (8WW, ScanWT, Mus, Fat, MusWT, and FatWT) were precorrected for known fixed effects and covariates. To achieve this, data collected on all farms in the SRS over the last decade were used, giving data sets of approximately 87,000 and 75,000 records for the Suffolk and Texel SRS, respectively. From these complete data sets, fixed effects for flock-year, sex, birth-rearing rank, and age of dam were estimated for each trait using ASREML (Gilmour et al., 1999), and these estimates were then used to precorrect the phenotypic records on genotyped animals, including ultrasonic scanning traits corrected for age at scanning. Animals without a complete phenotypic record were removed from the analysis.

Information Content. Information content was calculated at 1-cM intervals across all the regions under investigation in each population in this study for each analysis. The information content of an individual marker is the proportion of animals in which the allele inherited from the sire can be unambiguously identified. Information content at genome position i was calculated as var(pi)/0.25, where pi is the inheritance probability for each offspring included in the analysis and 0.25 is the expected variance of inheritance probabilities for a fully informative marker.

Estimation of QTL Position and Effects for a Single-QTL Model. The probability of inheriting a particular sire chromosome at a particular position was calculated for each offspring from the genotype data at 1-cM intervals along each chromosome, using the method of Knott et al. (1996). A small number of uninformative offspring (i.e., offspring with genotypes identical to their sire) were removed from the analysis for that particular genomic region. Each of the adjusted phenotypes was then regressed on the inheritance probabilities, at each location on each chromosome. For each regression, an F-ratio of the full model including the inheritance probability vs. the same model without the inheritance probability was calculated. The location with the largest F-ratio was taken to be the best estimated position for a QTL for each trait.

The QTL effects were estimated using two models, within family and breed level. First, in recognition of the fact that the samples were from outbred populations and only a small number of families were available, analyses were applied to each individual sire family. The test within each family produces an F-ratio with 1 df in the numerator and $(m - 2)$ df in the denominator, where m is the number of progeny in the analyzed family. Second, breed-level QTL were investigated. To remove scale differences between families arising from between-family heterogeneity of variance, the variance of each phenotypic measurement was standardized within each family to one. Standardized adjusted phenotypes were regressed on the inheritance probabilities for all animals genotyped for the genomic region under investigation at each location. Estimates of the putative QTL effect were calculated for each sire included in the analysis for the genomic region under investigation. This test produces an F-ratio with n df in the numerator, where n is the number of sires analyzed and $(\Sigma m) - 2n$ df in the denominator.

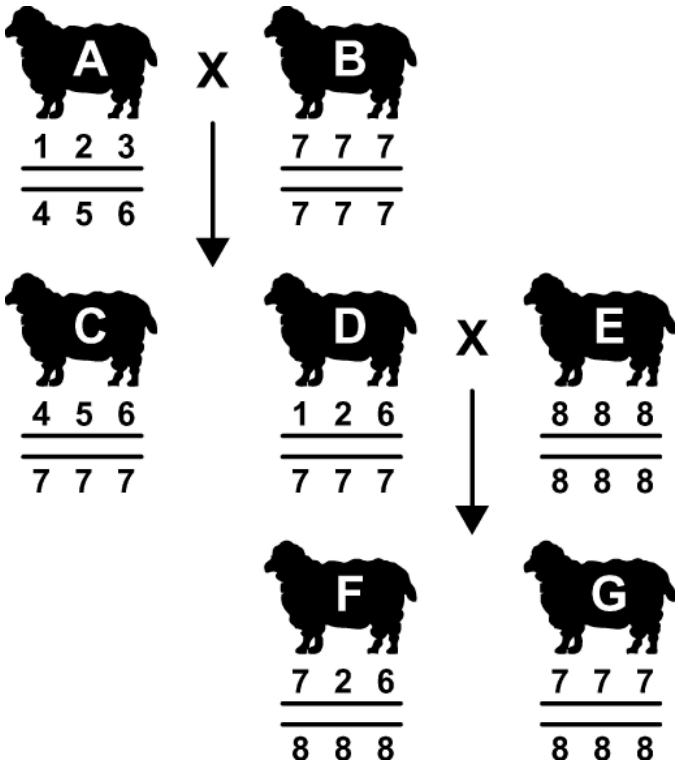

Figure 1. Simplified diagram of family structure used for the three-generation analyses.

Three-Generation Analysis. Our data set comprised two extended families: in the Suffolk breed, Sire 1973 was the progeny of Sire 561, and in the Texel breed, Sire 424 was the progeny of Sire 1. Additional analyses were performed on these related families. In genomic regions where the two sires had at least one informative marker in common, inheritance probabilities for the progeny of the older sire were calculated for the paternal haplotype inherited by the younger sire using an extension of the method of Knott et al. (1996). Inheritance probabilities for the same allele were also calculated in the progeny of the younger sire also using the method of Knott et al. (1996). Inheritance probabilities of the alleles shared by the two sires in the younger family were calculated by multiplying inheritance probabilities of the sire with those of the progeny. Adjusted phenotypes were subsequently regressed onto the hap-

Table 2. Summary of measured traits

	Suffolk			Texel		
Trait[a]	Mean	SD	RSD[b]	Mean	SD	RSD[b]
8WW, kg	31.8	6.89	3.48	22.4	4.32	2.93
ScanWT, kg	53.9	9.60	5.19	45.5	8.76	5.02
Mus, mm	31.3	4.66	2.65	28.7	3.28	2.44
Fat, mm	3.50	1.70	0.91	2.47	1.40	1.06
MusWT, kg	—	—	2.21	—	—	2.05
FatWT, kg	—	—	0.85	—	—	0.95

[a]The traits are defined as follows: 8WW = live weight at 8 wk of age; ScanWT = live weight at scanning (approximately 20 wk of age); Mus = muscle depth at the third lumbar; Fat = fat depth at the third lumbar; MusWT = muscle depth corrected for live weight; FatWT = fat depth corrected for live weight.
[b]Standard deviation of the phenotypic residual values after correcting for fixed effects and covariates included in the model.

lotype inheritance probabilities. A single-QTL effect was estimated for the entire family.

To illustrate the methodology to derive inheritance probabilities, consider the simplified pedigree in Figure 1, in which Sire A is mated to Dam B, producing offspring C and D, and Male offspring D is mated to Dam E, producing Offspring F and G. Each animal is genotyped for three linked markers as shown. For simplicity, the dam genotypes are included and are unique. Probabilities for inheriting alleles 1, 2 and 6 from Animal A are all 1 because these are the alleles inherited by the younger sire (Animal D) regardless of the phase in Sire A. Likewise, probabilities of inheriting alleles 4, 5, and 3 from Animal A are all 0. Inheritance probabilities between markers are calculated using the recombination fractions between flanking markers and the current position. Assigning probabilities of 1 to common haplotype alleles in the younger sire avoids all offspring with 0 inheritance probabilities in the final generation and hence not contributing to the analysis. Likewise, the inheritance probability for alleles 2 and 6 in Animal F is also 1, and the inheritance probability of allele 7 at these marker locations is 0. Final inheritance probabilities for Animals F and G are obtained by multiplying their inheritance probabilities with those of Animal D at each location along the chromosome.

The analysis contrasts the allele in the younger sire inherited from the grandsire with the allele not inherited from the grandsire and that inherited from the dam. In Figure 1 for the first marker, allele 1 is contrasted with alleles 4 and 7 and for the second marker, allele 6 is contrasted with alleles 3 and 7.

Significance Thresholds. The 5% regional thresholds were calculated for each trait, for each analysis using permutation testing (Churchill and Doerge, 1994). This threshold was adjusted using a Bonferroni correction to produce a 5% genomewide threshold, assuming the total genome size of the sheep to be 3,500 cM (Maddox et al., 2001). For example, the 5% genomewide *P*-value from a 100-cM chromosome would be equivalent to the (100/3,500) × 0.05 = 0.0014 chromosomal *P*-value. In addition, a suggestive threshold (Lander and Kruglyak, 1995) equivalent to one false-positive result per genome scan was calculated with a *P*-value 20 times larger than the genomewide *P*-value.

Confidence Intervals. If the largest F-ratio indicated a QTL at the suggestive level, one- and two-LOD (LOD, logarithm of the odds of linkage) support intervals were produced by taking the region of the chromosome encompassed when reducing the largest *F*-ratio by the equivalent of an LOD score of either 1 or 2 (Lander and Botstein, 1989). This calculation was preferred to the bootstrap method (Visscher et al., 1996), which produces conservative intervals around areas of higher marker density (Walling et al., 1998b, 2002) and typically produced intervals covering the whole chromosome with these data owing to the shallow pedigree structure and consequently low numbers of recombination events (results not shown).

Two-QTL Model. When the largest *F*-ratio indicated a QTL at the suggestive level, a two-QTL analysis was applied. A grid search was performed at 1-cM intervals along the same chromosome. The best-fitting two-QTL model was tested against the model fitting only one QTL using an *F*-ratio with 1 df in the numerator.

The program QTL Express (Seaton et al., 2002) was used for the breed and family-level analyses for single- and two-QTL models and the calculation of the thresholds.

Results

Summary statistics for the two breeds are presented in Table 2. In terms of the QTL analyses, this study was successful in detecting QTL at the suggestive level for all families investigated, and for all traits considered, in seven of the nine chromosomal regions. However, the strength of evidence varied according to the approach used to analyze the data.

Breed-Level Analyses

One genomic region produced suggestive evidence for a QTL in the breed-level analyses. This QTL was in the Suffolk breed and located on chromosome 1 with the estimated position at 227 cM lying between markers

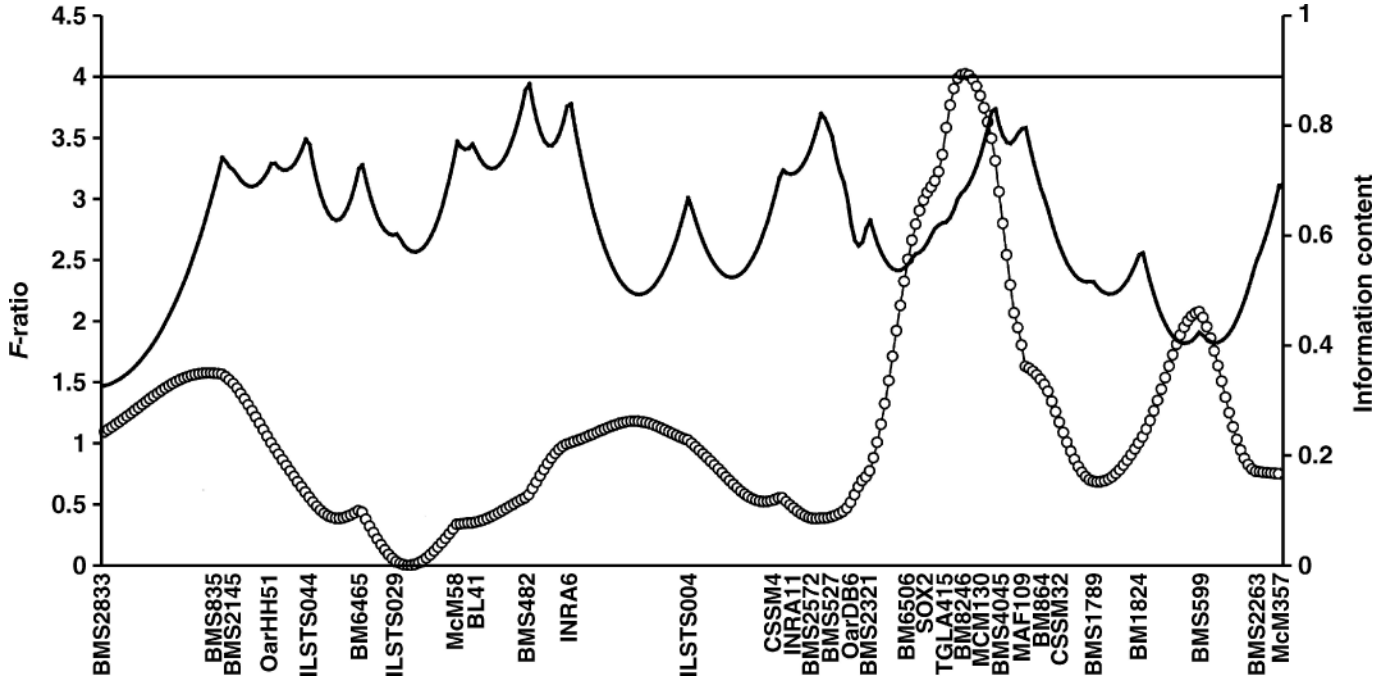

Figure 2. *F*-ratio profile (○) for muscle depth and information content (thick continuous line) along chromosome 1 from the population analyses. Approximate marker positions are indicated on the x-axis, and the suggestive threshold is marked by the thin horizontal line.

BM8246 and McM130. The QTL affected muscle depth with the effect significantly different from 0 only in Family S1 despite the inclusion of Families S2 and S3 in the analysis. The one- and two-LOD support intervals were between 211 to 239 cM and 203 to 256 cM, respectively, between markers BMS2321 and BMS1789. The distribution of the *F*-ratio and the information content across the chromosome is presented in Figure 2. Interestingly, the inclusion of a second QTL was statistically significant at a nominal level (P = 0.042). The location of the first QTL remained almost identical to the single-QTL model located at 228 cM. The second was located at 285 cM around the location of BMS599, where a second smaller peak can be seen in Figure 2, albeit in an area of lower information content. The QTL effects are in repulsion, with the size of the first QTL significantly different from 0 only in Family S1 and the second QTL significant only in Family S3.

Individual Family Analyses

All families produced evidence for suggestive QTL in one or more regions, although none produced evidence for a QTL at the 5% genomewide level. The suggestive QTL are summarized in Table 3 in decreasing order of statistical significance. As would be expected, the results differed between families, with only three genomic regions containing suggestive QTL in two separate families for a similar trait (growth, muscularity or fatness) with best-estimated positions separated by less than 100 cM. These three regions were growth traits (8WW and ScanWT) in Families S1 and S2 on chromosome 18 at positions 101 and 58 cM, respectively; Fat on chromosome 20 in Families T1 and S1 at 48 and 57 cM, respectively; and fat traits (Fat and FatWT) on chromosome 3 in Families T8 and S2 at 248 and 254 cM, respectively.

The sizes of effects for the suggestive QTL varied from approximately 0.4 to 1.0 residual phenotypic standard deviation (**RSD**). The largest effect of 1 RSD was the effect on fatness on chromosome 3 in Family T8. The size of effects for the majority of suggestive QTL was between 0.6 and 0.8 RSD. The one- and two-LOD support intervals were on average 43 and 139 cM long, respectively. Smaller LOD support intervals were typically obtained when the maximum *F*-ratio was high. Two LOD support intervals covered the entire chromosome when the maximum *F*-ratio was less than 8.5.

Three-Generation Analysis

Two regions in each of the joint families produced suggestive statistical evidence for a QTL. The regions were chromosome 3 and 6 in the Suffolk family and chromosomes 2 and 18 in the joint Texel family (T1/6), and the results are summarized in Table 4. The distribution of the test statistics for those traits with significant evidence for a QTL and the information content across the chromosome are shown in Figures 3 through 6. The most statistically significant effects were for muscle depth (both unadjusted and adjusted for weight) on chromosome 18 in the Texel family. The estimated position was at 87 cM between markers OB2 and CSSM18, 6 cM proximal in comparison to the indi-

Table 3. Summary of suggestive QTL from the individual family analyses, presented in order of decreasing significance

Chr[a]	Trait[b]	Family[c]	*F*-ratio	Position, cM[d]	Effect ± SE	1 LOD CI[e]	2 LOD CI[e]
2	Fat, mm	T1	12.84	132	0.62 ± 0.17	125 to 139	120 to 156
18	8WW, kg	S1	12.08	101	2.58 ± 0.74	95 to 101	87 to 101
4	FatWT, mm	T6	11.69	137	0.78 ± 0.23	121 to 138	112 to 138
20	Fat, mm	T1	10.37	48	0.57 ± 0.18	30 to 64	0 to 80
2	FatWT, mm	T1	10.10	132	0.51 ± 0.16	124 to 144	112 to 173
3	Mus, mm	T9	9.97	0	1.90 ± 0.60	0 to 40	0 to 89
2	8WW, kg	T3	9.74	131	1.89 ± 0.61	109 to 138	61 to 144
1	Mus, mm	S1	9.22	226	1.85 ± 0.61	212 to 238	199 to 257
1	MusWT, mm	S1	8.91	223	1.38 ± 0.46	210 to 240	190 to 261
18	Mus, mm	T6	8.85	93	1.56 ± 0.52	79 to 101	63 to 101
3	Fat, mm	T8	8.77	248	0.97 ± 0.33	205 to 282	181 to 282
3	MusWT, mm	T2	8.73	222	1.45 ± 0.49	189 to 250	0 to 282
2	Mus, mm	T9	8.56	61	2.00 ± 0.69	0 to 98	0 to 179
1	ScanWT, kg	S1	8.17	306	3.42 ± 1.20	297 to 310	0 to 310
4	ScanWT, kg	T5	7.90	0	3.56 ± 1.27	0 to 17	0 to 138
18	ScanWT, kg	S2	7.79	58	2.12 ± 0.76	32 to 75	0 to 105
18	FatWT, mm	T7	7.83	84	0.68 ± 0.24	64 to 91	0 to 105
20	Fat, mm	S1	7.74	57	0.44 ± 0.16	0 to 80	0 to 80
3	FatWT, mm	S2	7.71	254	0.44 ± 0.16	225 to 282	0 to 282
2	ScanWT, kg	T3	7.61	127	2.80 ± 1.02	100 to 138	0 to 254
6	MusWT, mm	S2	7.35	4	0.96 ± 0.35	0 to 53	0 to 150
3	FatWT, mm	T8	6.99	234	0.84 ± 0.32	205 to 282	0 to 282
3	MusWT, mm	T9	6.93	0	1.36 ± 0.52	0 to 89	0 to 282
2	8WW, kg	T4	5.67	248	2.28 ± 0.96	188 to 254	0 to 254

[a]Chr = chromosome number.
[b]The traits are defined as follows: 8WW = live weight at 8 wk of age; ScanWT = live weight at scanning (approximately 20 wk of age); Mus = muscle depth at the third lumbar; Fat = fat depth at the third lumbar; MusWT = muscle depth corrected for live weight; FatWT = fat depth corrected for live weight.
[c]Family is the family code as defined in Table 1.
[d]Positions are given in centimorgans from the marker closest to the distal end of the p arm on metacentric chromosomes or the marker closest to the centromere on telocentric chromosomes.
[e]1 LOD CI and 2 LOD CI are the 1 and 2 LOD drop score confidence intervals, respectively, expressed in centimorgans from the marker closest to the distal end of the p arm on metacentric chromosomes or the marker closest to the centromere on telocentric chromosomes.

vidual analysis for T6, with the two-LOD support including all locations distal to marker TGLA122. The information content across the two-LOD support region remained above 0.65 at all locations. The effect of the QTL was 1.31 mm and 1.15 mm for Mus and MusWT, respectively, slightly smaller than, although not significantly different from, the result from the single-QTL analysis. The other significant effect in the Texel family

Table 4. Summary of suggestive QTL from the three-generation analyses, presented in order of decreasing significance

Chr[a]	Trait[b]	Family[c]	*F*-ratio	Position, cM[d]	Effect ± SE	1 LOD CI[e]	2 LOD CI[e]
18	MusWT, mm	T1/6	11.40	87	1.15 ± 0.33	67 to 101	76 to 99
6	MusWT, mm	S2/3	11.35	0	1.21 ± 0.36	0 to 60	0 to 84
18	Mus, mm	T1/6	10.03	87	1.31 ± 0.40	64 to 101	74 to 101
6	Mus, mm	S2/3	9.93	0	1.29 ± 0.40	0 to 26	0 to 84
3	8WW, kg	S2/3	8.04	282	2.33 ± 0.81	242 to 282	0 to 282
2	Fat, mm	T1/6	7.99	235	0.61 ± 0.21	228 to 254	0 to 254
3	FatWT, mm	S2/3	7.29	255	0.40 ± 0.15	222 to 282	0 to 282

[a]Chr = chromosome number.
[b]The traits are defined as follows: 8WW = live weight at 8 wk of age; Mus = muscle depth at the third lumbar; Fat = fat depth at the third lumbar; MusWT = muscle depth corrected for live weight; FatWT = fat depth corrected for live weight.
[c]Family codes are defined in Table 1. T1/6 refers to the joint family comprising T1 and T6, and S2/3 refers to the joint family comprising S2 and S3.
[d]Positions are given in centimorgans from the marker closest to the distal end of the p arm on metacentric chromosomes or the marker closest to the centromere on telocentric chromosomes.
[e]1 LOD CI and 2 LOD CI are the 1 and 2 LOD drop score confidence intervals, respectively, expressed in centimorgans from the marker closest to the distal end of the p arm on metacentric chromosomes or the marker closest to the centromere on telocentric chromosomes.

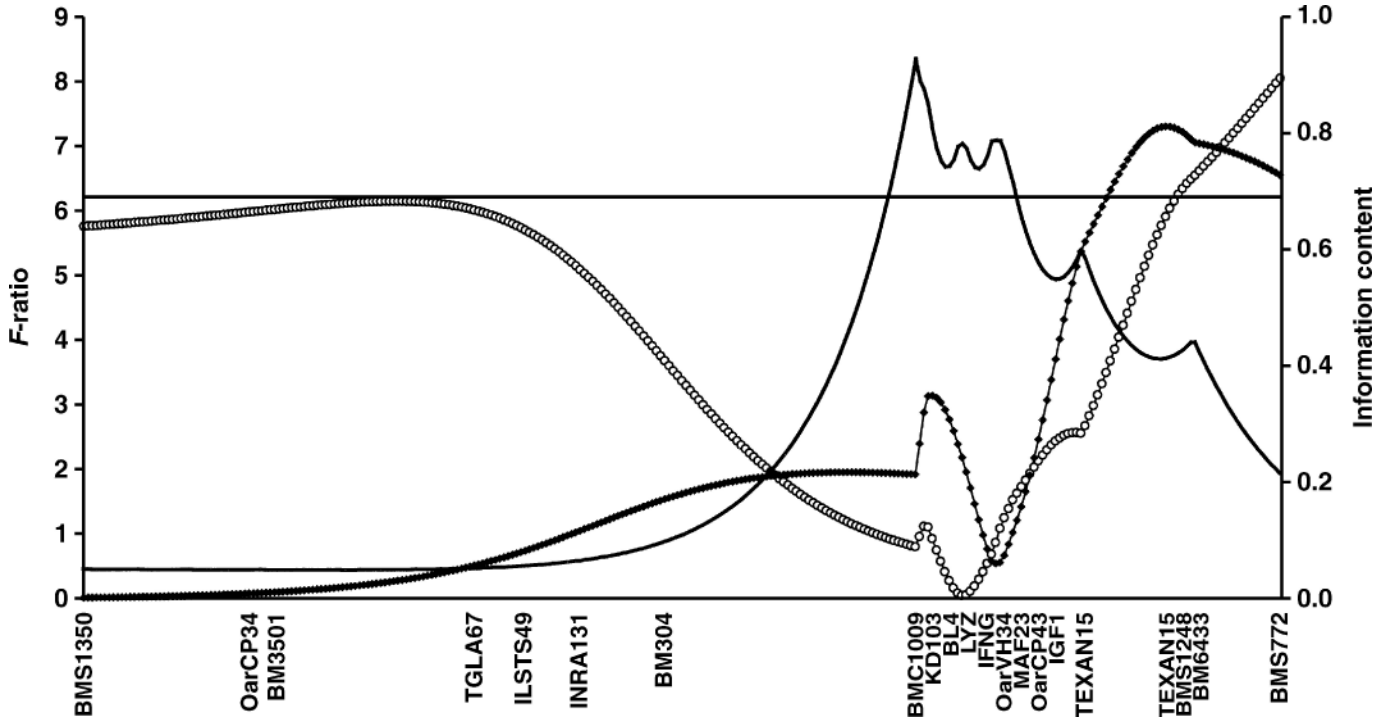

Figure 3. *F*-ratio profile for live weight 8 wk of age (○) and fat depth corrected for live weight (◆), and information content (thick continuous line), along chromosome 3 from the analysis of joint families S2 and S3. Approximate marker positions are indicated on the x-axis, and the suggestive threshold is marked by the thin horizontal line.

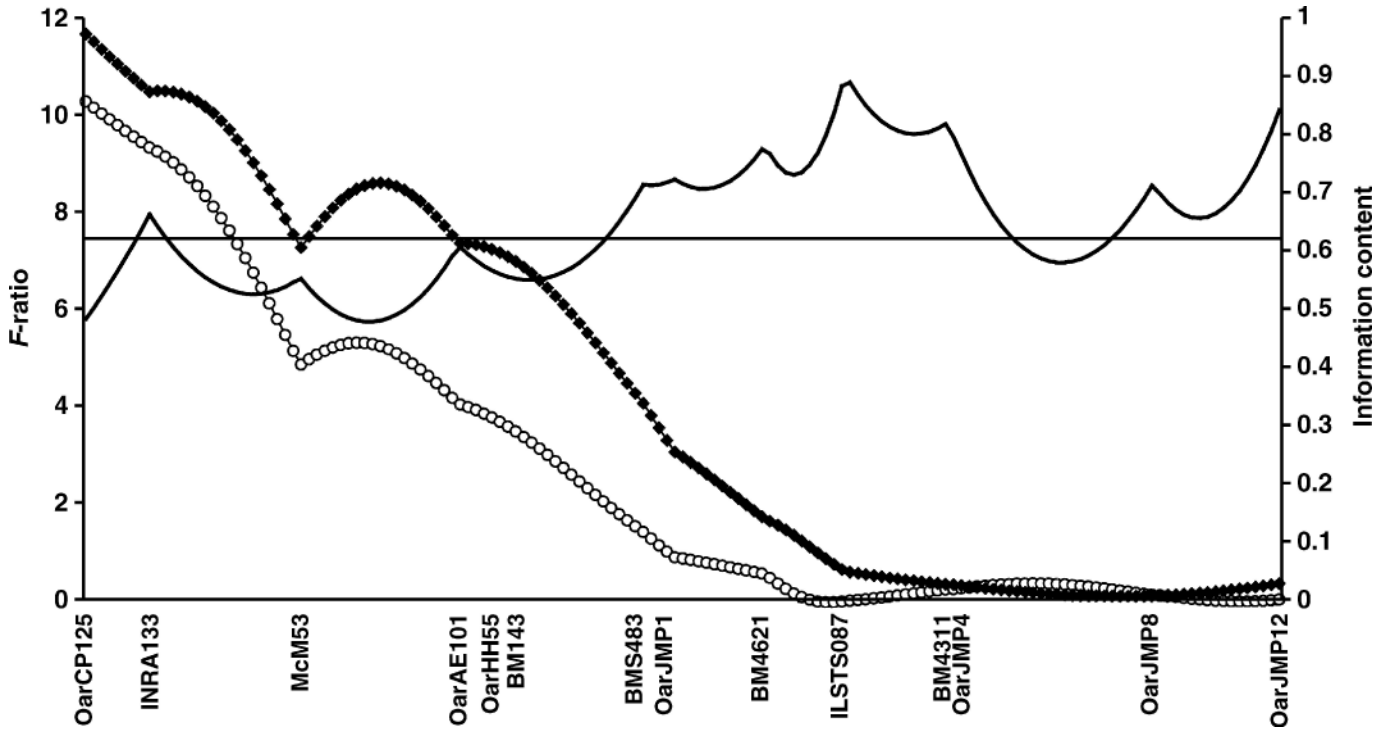

Figure 4. *F*-ratio profile for muscle depth (○) and muscle depth corrected for live weight (◆), and information content (thick continuous line), along chromosome 6 from the analysis of joint families S2 and S3. Approximate marker positions are indicated on the x-axis, and the suggestive threshold is marked by the thin horizontal line.

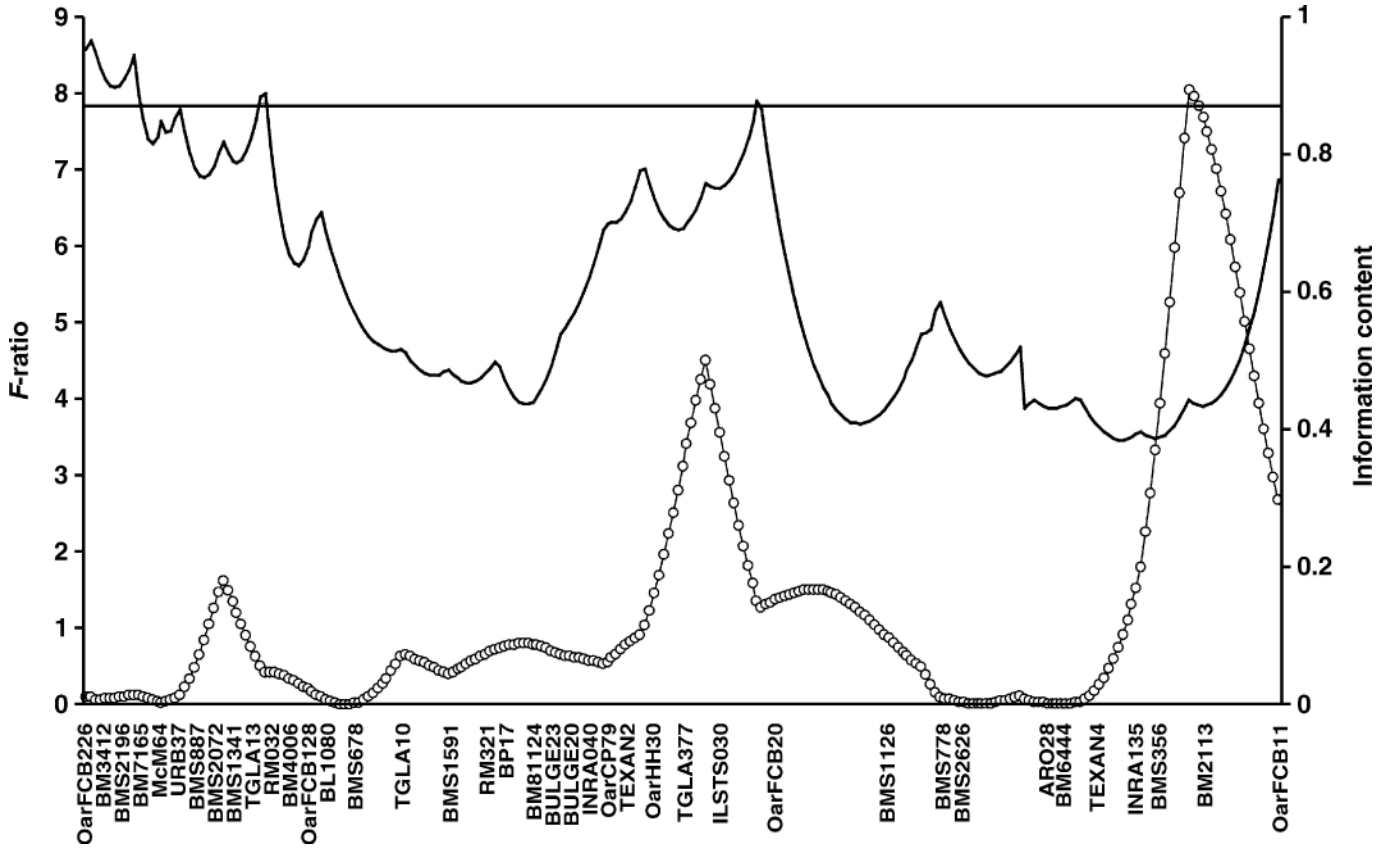

Figure 5. *F*-ratio profile for fat depth (○) and information content along chromosome 2 from the analysis of joint families T1 and T6. Approximate marker positions are indicated on the x-axis, and the suggestive threshold is marked by the thin horizontal line.

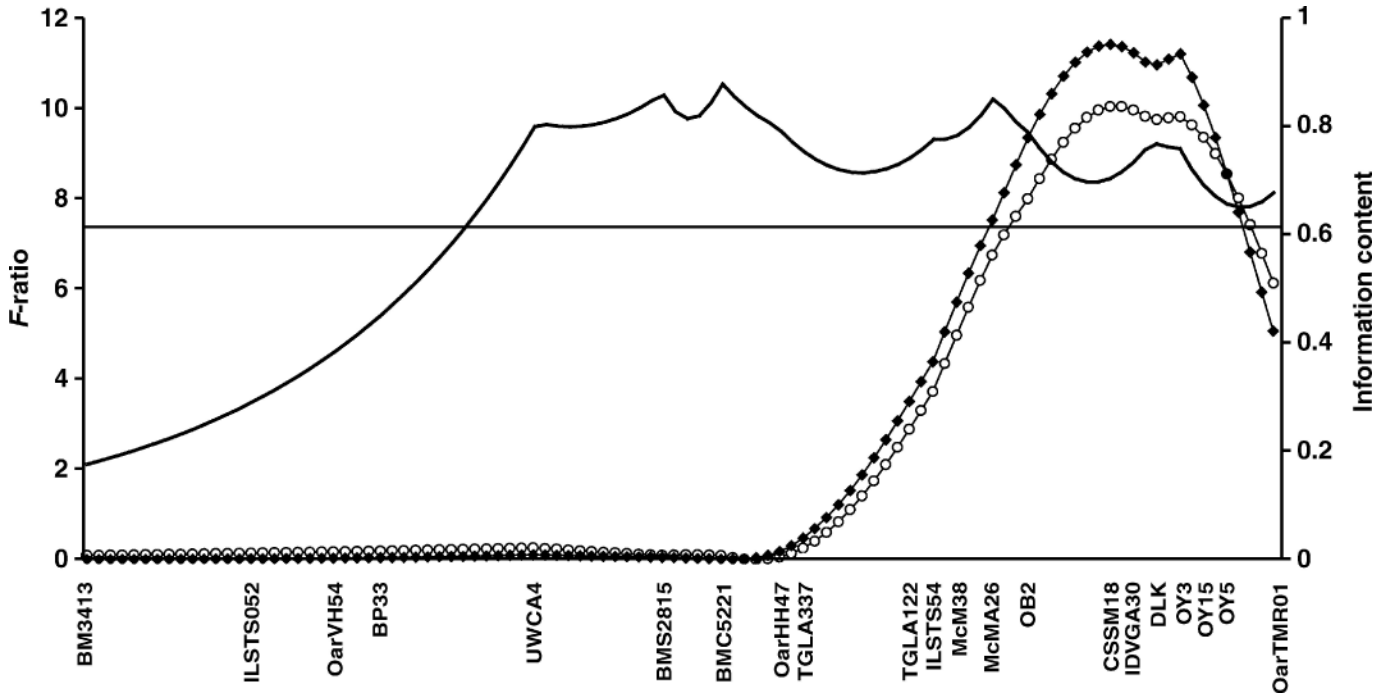

Figure 6. *F*-ratio profile for muscle depth (○) and muscle depth corrected for live weight (◆), and information content (thick continuous line), along chromosome 18 for the analysis of joint families T1 and T6. Approximate marker positions are indicated on the x-axis, and the suggestive threshold is marked by the thin horizontal line.

Table 5. Results for two-QTL models that gave a significantly better fit to the data at a 5% nominal level than the equivalent single-QTL model, presented in order of decreasing significance

Chr[a]	Trait[b]	Family[c]	*P* (2v1)	Position 1[d]	Effect ± SE	Position 2[d]	Effect ± SE
1	Mus, mm	S1	0.007	222	0.73 ± 0.21	295	0.75 ± 0.27
2	8WW, kg	T3	0.011	134	–1.55 ± 0.37	189	2.27 ± 0.79
18	Mus, mm	T1/6	0.015	54	–1.22 ± 0.44	82	2.00 ± 0.49
18	MusWT, mm	T1/6	0.018	54	–0.97 ± 0.37	82	1.70 ± 0.40
18	Mus, mm	T6	0.032	53	–1.47 ± 0.57	84	2.29 ± 0.64

[a]Chr = chromosome number.
[b]The traits are defined as follows: 8WW = live weight at 8 wk of age; Mus = muscle depth at the third lumbar; MusWT = muscle depth corrected for live weight.
[c]Family is the family code as defined in Table 1. T1/6 refers to joint family comprising T1 and T6.
[d]Pos 1 and Pos 2 are the positions of the 1st and 2nd QTL, respectively, given in centimorgans from the marker closest to the distal end of the p arm on metacentric chromosomes or the marker closest to the centromere on telocentric chromosomes.

on chromosome 2 affected fat depth with an effect of 0.61 mm at 235 cM around the location of marker BM2113, with an information content of 0.43. The *F*-ratio was 7.99. This represented a decrease in significance in comparison to the individual analysis for T1; the size of effect remained the same but the estimated positions differed by over 100 cM.

The effect on muscle depth (both unadjusted and adjusted for weight) on chromosome 6 in the Suffolk family remained at the end of the chromosomal region, in agreement with the analysis in Family S2. Effects of 1.29 mm and 1.21 mm for Mus and MusWT, respectively, were larger than that from the individual family analysis of S2. The effects on chromosome 3 were toward the distal end of the chromosome, where information content was low because markers where initially scattered around the IGF1 and IFNG. For all the results for the three-generation analysis, with the exception of the Fat effect on chromosome 2 for joint family T1/6, the strength of evidence for a QTL was greater than that from the individual family analyses.

Two-QTL Model

When the data presented evidence of a suggestive QTL, a two-QTL model was applied to the data. The results presented in Table 5 represent cases where the inclusion of a second QTL was statistically significant at a nominal 5% level, and the two putative QTL were located in different marker intervals. Owing to the collinearity between locations within the same interval, estimates of two QTL locations within the same interval are unreliable and highly variable, generally being inflated. This typically leads to an optimal two-QTL model with the loci closely positioned and acting in repulsion, with large effects that are similar but opposite. As observed by Whittaker et al. (1996), it is impossible to map nonisolated QTL; hence, such results have not been included.

The strongest statistical evidence for a second QTL was on chromosome 1 for muscle depth in Family S1. The original single QTL for muscle depth on chromosome 1 appears, from this analysis, to be a result of two separate QTL acting in coupling with almost equal effect (0.73 mm and 0.75 mm). The position of the first QTL (222 cM) is almost identical to that from the single-QTL analyses (226 cM) with the second QTL approximately 70 cM distal at 295 cM. The coupling of the two QTL effects is in contrast to the result for the population analysis where the effects, in different families, were in repulsion. The inclusion of a second QTL was not statistically significant for the similar trait of muscle depth adjusted for body weight in the same family.

The data for 8-wk weight on chromosome 2 for Family T3 also produced nominal statistical evidence for a second QTL. The two QTL acted in repulsion, with the first QTL with similar position (134 cM) and effect (1.55 kg) to the single-QTL model. The second and larger QTL is 55 cM distal at 189 cM, with an opposing effect of 2.27 kg. The only other analysis to produce statistical evidence for a second QTL in the region under investigation was that of the muscle data for Family T6 and joint Family T1/6. In this result, both the QTL from the two-QTL model were proximal to that from the single-QTL model located at approximately 54 cM and 82 cM. The effects were in repulsion, and the more distal QTL having the larger effect.

Whereas the second QTL were statistically significant at a nominal level for all the results in Table 5, none were significant at the suggestive significance level that was applied to the single-QTL models.

Discussion

This study has provided one of the first accounts of a QTL study for growth and carcass conformation traits in domesticated sheep covering several genome regions. In contrast to many other QTL studies, the analysis has been applied to commercial animals rather than to experimental populations comprising diverse crosses. There are many reasons for using this approach. Primarily, the animals and data used in this study are routinely produced and collected through the established genetic improvement schemes for the Texel and

Suffolk breeds in the United Kingdom. As this information is readily available, this considerably decreases the costs of the experiment. Given the relatively long generation interval in the larger domestic livestock species, the use of existing population structures also substantially decreases the time taken to create a population with the numbers of animals necessary for appropriate power for QTL detection. Further, results from commercial animals are immediately relevant for use in marker-assisted selection schemes that aim to optimize the response to selection by the inclusion of molecular data into breeding value estimation (e.g., Goddard and Hayes, 2002). Significant findings from the study can, in principle, be included into breeding value estimation immediately for the next round of matings. Indeed, the repetition of analysis and consequent generations undergoing marker-assisted selection can significantly improve the estimated position and effect of the desired locus (Pong-Wong et al., 2002), and thus contribute to the benefits of marker-assisted selection.

From the breed-level analyses, statistical evidence was found for a QTL on chromosome 1. The effect on muscle depth was located 50 cM proximal to the transferrin gene, one of a family of metal-binding proteins with an in-vivo preference for ferric iron. Interestingly, the second significant QTL in the two-QTL analysis was located only 12 cM distal to location of the transferrin gene. Given the confidence intervals for the QTL positional estimate and the sparse knowledge of the sheep genome, the transferrin gene is a possible candidate gene for the observed effects. However, there are many genes in this region and further experiments are required to test specific hypotheses regarding candidate genes.

From the analysis of individual families, the most significant result affected fatness on chromosome 2. The estimated position was 26 cM distal to the myostatin locus, responsible for the double muscling phenotype in cattle. The position and the effects are in good agreement with a previous Texel study in New Zealand (Broad et al., 2000). The locus detected by Broad et al. (2000) not only affects muscle measurements, but also is reported to affect fat depth, in agreement with this study. One other study in sheep has also reported effects on fatness around the myostatin region (Marcq et al., 2002). Although a causative effect of the myostatin locus on fatness in these populations cannot be unambiguously inferred, these results are consistent with the observation that double-muscled animals have carcasses with a low percentage of fat. Therefore, mounting evidence suggests that this region of the sheep genome contains a gene or number of genes with significant effects on carcass composition.

Another significant region detected in the three-generation analysis was on chromosome 18. Effects on muscle depth traits were detected in Texel Family T1 and more significantly in the joint Family T1/6. The position of this effect corresponds to the region of the chromosome containing both the callipyge gene and the Carwell locus. Given the complex nature of callipyge inheritance, in which only heterozygous offspring inheriting the mutation from their sire express the phenotype, as well as the observation that the callipyge effect is localized to the loin region, it is unlikely that the effect in this population is attributable to the segregation of callipyge alleles; however, the effect is similar to the description of the Carwell gene. The position of the Carwell gene was reported to be around 2 to 6 cM telomeric of CSSM18 (Nicoll et al. 1998); this study found an effect approximately 2 cM telomeric of CSSM18 in the individual family analysis and 5 to 10 cM centromeric of CSSM18 in the joint family analyses. The effect was on muscle depth of the longissimus dorsi, with no effect on fatness, although the position of the phenotypic measurement differs slightly between this and the previous study, namely, 3rd lumbar vs. 12th rib, respectively. The size of the effect in the work of Nicoll et al. (1998) indicated an increase of muscle depth adjusted for live weight in Australian Poll Dorset sheep of 2.97 mm (SE = 0.82), which is not significantly different from the effect in the Texel families in this study (1.15 to 2.00 mm). The effect was more significant in the three-generation analysis when the model was adjusted for live weight, which is also in agreement with previous findings (Nicoll et al. 1998), and suggests that the primary effect is an alteration in muscle shape. Given the similarities between the previous description of the Carwell locus and these results, it can be hypothesized that the same or similar locus was also segregating in the flocks used in this study.

The use of a candidate region approach seems to be successful in finding QTL in the chosen regions; however, it is not possible to estimate the number of effects that were not detected as a result of being outside of the genomic regions in this study. Nonetheless, the strategy of selecting genomic regions that have previously been reported to contain QTL affecting similar traits seems to be effective. This study suggests that where constraints that prevent a full genome scan exist, selection of genomic regions previously reported to be associated with effects on traits similar to those being studied is a valid strategy to detect QTL.

The analysis of these data used three differing methodologies assuming different underlying models of any putative gene. The breedwide analyses are most powerful when a QTL is segregating in all the sires. Given the outbred nature of the population and assuming a biallelic QTL, even in the most favorable scenario with QTL allele frequencies of 0.5 on average, the effect will not be segregating in 50% of the sires. Given the limited number of sires likely to be heterozygous for the QTL, the residual variation from other nonsegregating families is such that only very large QTL will be found to be statistically significant. Despite this, for any given significance level, the breed-level analyses are likely to be the most powerful. This study found a greater number of suggestive QTL from the within-family analyses,

but the results were not adjusted for the number of families tested.

The mapping of QTL involves substantial multiple testing. This study encompassed 42.3% of the sheep genome and attempted to correct for multiple positional testing using the suggestive and 5% genomewide thresholds suggested by Lander and Kruglyak (1995). The study did not adjust for the 12 families tested in the within-family analyses, nor did it correct for the three traits analyzed (growth, muscle, and fat). Given the number of tests, it is likely that some results are Type 1 errors (identifying a QTL when there is not one present); however, increasing the stringency of the thresholds would increase the frequency of Type 2 errors (failing to identify a QTL present in the region studied). Adjusting thresholds for the number of traits analyzed seems to be too stringent, as results previously declared significant at a suggestive or 5% genomewide level may no longer reach the new required threshold simply because of the addition of a new trait in the analyses. Such a result would be discarded despite no change in the evidence for a putative QTL. Hence, in accordance with the recommendation of Lander and Botstein (1995), suggestive results are reported with the recognition of the potential for false positive results.

The three-generation analysis has a number of advantages over the individual family analyses. The analyses benefit from increased power owing to the additional numbers of progeny. Furthermore, the extra generation can generate additional recombination events, which leads to improved resolution of the mapped position. This is usually a better means of improving the positional estimate of the QTL than increasing marker density, which generally adds little precision (Visscher et al., 1996). In other words, additional recombination events are more likely to be observed through the addition of more animals rather than more markers.

If confined to a single generation, Meuwissen et al. (2002) demonstrated through work on a QTL for twinning rate that a combination of both linkage and linkage disequilibrium mapping is able to fine-map QTL. Using only large half-sib families, similar to the data analyzed in this study, Meuwissen et al. (2002) were able to map a QTL within a region less than 1 cM. Application of this methodology to the sheep data used in this study may improve the current resolution of the identified QTL.

In summary, this study has identified at least three regions of the genome associated with muscle and fat depth in both Suffolk and Texel sheep that lend themselves to additional studies as well as exploitation possibilities. As described earlier, tracking haplotypes defining these chromosomal regions through subsequent generations in the same families can, in principle, enhance short-term genetic progress if no recombination occurs within the defined region, or improve the precision of the estimated QTL effect if observed recombinations do occur. In other words, our results present an opportunity to simultaneously improve the estimated position and effect of the desired locus as well as contribute to the benefits of marker-assisted selection.

Implications

Several regions of the sheep genome have produced suggestive evidence for quantitative trait loci with significant effects on carcass traits, when analyzed within half-sib families. Four of these regions, affecting muscle depth (chromosomes 1 and 18) and fat depth (chromosomes 2 and 3) contained evidence that was consistent across families or generations. These results have been found in sheep in commercial breeding programs and, hence, the results can potentially be included directly within these breeding programs. Such inclusion would have the benefit of simultaneously increasing genetic progress in the measured traits (marker-assisted selection) as well as allowing a refinement of the mapped position of the quantitative trait locus.

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