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A genomic scan of porcine reproductive traits reveals possible quantitative trait loci (QTLs) for number of corpora lutea

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Abstract. Reproductive traits have low heritabilities, are expressed in only one sex, and are not measurable until sexual maturity (Avalos and Smith, Anim Prod 44:153, 1987). Using traditional methods, selection for reproductive traits is relatively less effective than selecting for growth or carcass traits. Traits most affected by a small number of genes with major effects rather than many genes with small effects are most amenable to MAS. As part of our porcine genome scan to identify quantitative trait loci (QTLs) of economic importance in marker-assisted selective (MAS) breeding programs, we examined 8 reproductive and farrowing traits in the University of Illinois (UI) Meishan × Yorkshire Resource Family. Gilts were genotyped with 119 microsatellite markers (MS) with intervals averaging 24 cM over all 18 porcine autosomes. F-ratios supporting QTL location were calculated by the least squares regression method. Results suggestive of linkage at the 5% genome-wide level were observed for the number of stillborn piglets on Chromosome (Chr) 4 (SSC4) (p-value = 0.0001), corpora lutea on SSC8 (p-value = 0.00027), and gestation length on SSC9 (p-value = 0.00019). Results for additional loci relevant to litter size, number of corpora lutea on SSC15 and 7 (p-value = 0.0029 and 0.0028 at 107 and 150 cM, respectively), gestation length on SSC15 and 1 (p-value = 0.0017 and 0.0069 at 96 and 166 cM, respectively), uterine length on SSC7 and 5 (p-value = 0.0044 and 0.0075 at 148 and 1 cM, respectively) and piglets born per litter on SSC6 (p-value = 0.0075 at 102 cM), were not statistically significant at the 5% genome-wide level. Thus, the use of a linked marker to facilitate selection for reproductive traits has considerable potential. By using linked markers, selection can be applied to both sexes before sexual maturity, making genetic selection considerably more efficient and less costly.

Introduction

QTL affecting reproductive and farrowing traits can best be identified with MS markers in crosses between pig breeds dissimilar with regard to the characteristics of interest. The recent availability of a relatively high-density linkage map has made it possible to survey the porcine genome for markers closely linked to a specific trait variant by analysis of cosegregation patterns of the marker alleles and levels of the quantitative trait in each chromosomal interval. Traits most affected by a small number of genes with major effects rather than many genes with small effects are most amenable to MAS. Rathje and associates (1997) have shown that QTL associated with porcine ovulation rate and number of fetuses are due to genes with large effects.

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Only one potential QTL for a porcine reproductive trait or clearly related locus has been identified. This QTL is a variant of the estrogen-receptor gene (Rothschild et al. 1996) and was identified using a candidate gene approach. A candidate gene approach is easy to implement, but is limited by the number of candidate genes available. Genomic scans with genetic markers require a designed population and a large number of genotypes, but offer the most complete search for QTLs across the genome. Using this approach and applying the measure of genome-wide significance defined by Kruglyak and Lander (1995), Rathje et al. (1997) reported a possible QTL on SSC8 associated with the ovulation rate in a line derived from the F_3 generation of a cross between the Large White and Landrace breeds selected for 10 generations for increased ovulation rate and embryonal survival. Here we have used the method of genomic scanning and a greater coverage of MS markers to identify intervals of the genome influencing selected reproductive and farrowing traits.

This study uses a resource population of 25 full and half-sib families (304 individuals), the descendants of 3 Meishan boars and 7 Yorkshire sows, to identify markers near the loci responsible for the reproductive differences in these breeds. Meishans are superior to Yorkshires in most reproductive traits. For example, Meishans have, on average, five more piglets per litter, 6.4 more corpora lutea, and 0.4 fewer stillborn piglets than Yorkshires (White et al. 1993). However, Yorkshires have an increased uterine length (an average of 28.4 cm), suggesting that sows with a favorable combination of characteristics from these breeds might be reproductively superior.

Materials and methods

The UI resource population. A three-generation Meishan × Yorkshire cross containing a total of 304 individuals was produced from three unrelated (for three generations) Meishan boars, each mated with either two or three Yorkshire sows (Schook and Wheeler 1994). Details of the reference family pedigree and variance of the traits are by White et al. (in preparation) and in Table 1. Examination of the variance for each of these traits in the reference family confirmed all traits are normally distributed. All individuals in the F_2 generation were characterized for growth traits, only males were characterized for carcass traits, and females (n = 122) were characterized for four reproductive and four farrowing traits.

Microsatellite marker map and genomic scan. We used highly polymorphic MS markers selected from a genetic linkage map (Rohrer et al. 1996) to partition the pig genome into intervals. For each of the 18 porcine chromosomes, three to thirteen MS markers were selected for even distribution, high polymorphism content, and ease of scoring as determined by Rohrer et al. (1996). The selection resulted in an average distance of ~25 cM between markers. Genotypes for the MS markers were determined as described by Rohrer and colleagues (1994). Maximum likelihood linkage

Table 1. Reproductive and farrowing traits for the F_2 generation of the UI Resource family.

Trait	Mean	SD	Range (Min; Max)	Number of SD in Range
Reproductive traits				
Gestation length (days)	45.4	2.9	40; 54	4.8
Number of corpora lutea	14.8	3.4	3; 33	8.8
Total fetuses	11	3.6	2; 20	5.0
Uterine length (cm)	564	136	156; 951	5.8
Farrowing traits (per litter)				
Total piglets born	9	3.1	2; 19	5.5
Number of piglets born live	8.6	3.1	0; 18	5.8
Number of stillborn piglets	0.4	0.8	0; 4	5.0
Number of piglets weaned	7.9	3.0	0; 15	5.0

analysis was used for ordering the marker loci relative to each other with the CRI-MAP (version 2.4) program (Green et al. 1990). Linkage analysis using the *build* option confirmed the order of the selected markers for each chromosome and was used to calculate the genetic distance between markers in the resource family. Because scoring of some markers was far easier than of others, to minimize the possibility that genotyping errors in one marker might affect the placement of those nearby, markers that were not placed with better than 1000:1 odds by use of *build* were inserted into the fixed map with the *all* option and allowing poorer odds. The integrity of this map was checked using the chrompic option, and suspected genotyping errors involving double recombinants occurring within small map distances were investigated, corrected where necessary, and marker order and distances recalculated as described. Final marker order was checked with the *flips* option by rearranging markers in successive linear groups of two, three, and four. The best supported marker order was used in the subsequent QTL analyses. None of the *flips* calculations resulted in a change of the marker orders.

Statistical analyses and evaluation of significance. Following the recommendations of Elston (1992) and Brown et al. (1994), we chose a hierarchical search strategy, performing a genome scan for QTLs using a sparse map, then following up in interesting regions with a more dense map. The sex-averaged map was used for all evaluations of QTLs. The presence of a QTL correlating with one of the phenotypic traits was investigated separately for each chromosome with the least squares regression program for mapping developed by Haley and coworkers (1994). The program was developed for detecting QTLs in an outcross between two breeds that are dissimilar for the phenotypic traits to be considered. QTL mapping for outcrossed pedigrees is complicated by the absence of complete homozygosity in the purebred generation. Distinct from QTL mapping in inbreed lines, the mating type is defined at the locus level rather than all loci in the initial cross. The different information content of paired markers flanking an interval results in a bias in QTL detection in outcross pedigrees (Knott 1997). Thus, for outcross pedigrees, methods employing a search using all markers along a chromosome simultaneously rather than pairwise sets of markers flanking an interval are an improvement. The simultaneous search method employed reduces bias and increases the power by reducing the residual variance. The multiple regression method is more amenable to these analyses than the maximum likelihood method because parameters such as fixed effects (treatment, gender, parity, etc.) can be included, and maximum likelihood is too computationally complex to accommodate a simultaneous search (Knott 1997).

In this study, the assumption is made that the two grandparental purebreeds are fixed for all traits of interest, and estimates of the probability that each individual is either homozygous for both alleles from one of the two purebred grandparents or heterozygous, inheriting alleles from each breed, are calculated for each possible QTL position on the genetic map. The additive and dominance coefficients of each possible QTL are then calculated from these genotype probabilities, conditional on the marker genotypes for each individual, and the phenotypic measurement for an individual is regressed onto them. The cosegregation patterns of traits and marker alleles in the third generation (F_2) progeny allow the identification of QTLs related to the phenotypic differences. In all analyses of reproductive and farrowing traits, a model specifying two fixed effects, family and parity, was used. Only one covariate, litter size, was used in the analysis of gestation length and number of corpora lutea. This covariate was included in the analyses because it is hypothesized to be a causal component of the total phenotypic variance under consideration. Analyses with and without the litter size covariate were compared for gestation length and the best result reported. The remaining traits were analyzed without covariates. All of the models were evaluated assuming no genetic background effects and only one QTL per chromosome.

To determine which intervals, indicated by the test statistic as possible locations of a trait, are genetically meaningful and which are owing solely to chance fluctuations, we examined our data, using three different measures of statistical significance, two relatively conservative, the other very liberal. The two conservative measures of statistical significance identified the same loci contributing to a particular trait with complete agreement. The 5% genome-wide significance level proposed by Lander and Kruglyak (1995) is highly stringent, making the assumption of complete saturation of the genome with markers. The criteria take into consideration the pointwise significance level of the specific threshold, the size of the genome, the rate of fluctuation of the statistic, and the threshold itself. Assuming a genome size of 2424 cM, an infinitely dense marker map, and a reproductive trait sample ($n = 122$), the 5% genome-side significance level requires an F-ratio of 10.60 and 6.59 for *significant* and *suggestive linkage*, respectively. A claim of linkage for the farrowing traits ($n = 116$) requires an F-ratio of 10.66 and suggestive linkage a 6.60 F-ratio.

We also considered a sparse-map case (Lander and Botstein 1989) that better fits our experimental design, where an adjusted threshold value takes into account the number of comparisons or intervals between markers for tests of significance. For out data set, the adjusted threshold, calculated by dividing the chosen level of Type 1 errors (α) by the number of intervals evaluated (M), was 0.05/101 or 0.0005, which results in an experiment-wise Type I error rate of $1-(1 - 0.0005)^{101}$ or 0.049 where there is independent inheritance of genetic markers. Use of the Type 1 error rate of 0.05 for declaration of significance means that $1 - (1–0.05)^{101}$ or 99.44% is the probability of a false positive occurring somewhere in the genome. The third, much more liberal, nominal (pointwise) significance value ($p < 0.05$, F-ratio >3.08) was also used to evaluate the F-ratios for evidence of a putative QTL. Because of the problem of multiple correlated traits, we expect to be wrong in identifying a QTL associated with a particular trait approximately 1 out of 18 times when using the later measure (i.e., 44 significant results observed divided by 808 tests done or 101 intervals between markers in the scan times eight reproductive traits). Therefore, we present these data only for the purpose of comparing our results with other published observations (For example, see Paszek et al. 1999). The other two measures of significance give false positive odds of ~1 out of 270 (three results *suggesting linkage* using either the Lander and Krugylak or the adjusted threshold criteria / 808 tests).

The magnitude and direction of the additive and dominance effects were also calculated with the programs of Haley and colleagues (1994). The additive genetic variance is the sum of the additive effects of an individual's alleles or the variance of breeding values of individuals in population mating randomly (Lynch and Walsh 1997). In our analyses, the magnitude of the additive effects represents the difference between homozygous individuals that received both copies of the gene from the parental Meishan boars and the mean of the two homozygous genotypes (i.e., QQ − 0.5 (QQ + qq), where the Q allele was inherited from a Meishan boar). Negative values indicate that the F_2 pigs homozygous for alleles of purebred Meishan origin have trait values less than the mean of the homozygotes receiving both alleles from either purebred Meishan or Yorkshire grandparents. Dominance effects describe one allele acting in concert with alleles at other loci to produce the trait phenotype, and the magnitude is the difference between gilts heterozygous for the putative QTL and the mean of the two homozygous genotypes (Qq − 0.5 (QQ + qq). Negative values indicate the F_2 heterozygotes receiving one allele from each purebred grandparent have trait values lower than the mean of the homozygotes receiving both alleles from either purebred grandparent.

Results

Of the eight reproductive traits examined for QTLs (Table 1), we identified chromosomal intervals associated with the number of stillborn piglets, number of corpora lutea, and gestation length (on SSC4, SSC8 and SSC9), produced F-ratios *suggestive of linkage* (Table 2, in bold) on a genome-wide basis (Lander and Kruglyak 1995). Using the adjusted threshold value of $p < 0.0005$, all three QTLs also met the criteria for a claim of linkage. The strongest evidence for a QTL involved in number of stillborn piglets was located near the telomere of SSC4 (F-ratio of 9.97 at 1 cM). The second, for a putative QTL involved in number of corpora lutea,

Table 2. Summary of QTL effects estimated for reproductive traits.

Trait	SSC	Position (cM)	F-ratio	*p*-value	df	F_2 mean ± S.E.	Additive effect[a]	Dominance effect[b]	% of F_2 variance[c]
Number of stillborn piglets	4	1	**9.97**	.00010	2, 98	0.59 ± 0.25	−0.31 ± 0.11**	−0.57 ± 0.18**	15.5
Number of corpora lutea	8	101	**8.89**	.00027	2, 104	17.18 ± 0.99	−1.20 ± 0.37**	−1.76 ± 0.63**	13.2
	15	107	6.20	.00290	2, 104	13.90 ± 1.09	−0.81 ± 0.58*	3.84 ± 1.13**	9.1
	7	150	6.22	.00280	2, 104	15.17 ± 0.74	2.57 ± 0.73**	1.33 ± 1.21	9.1
Gestation length (days)	9	135	**9.30**	.00019	2, 103	47.99 ± 1.34	1.52 ± 0.44**	−2.34 ± 0.75**	13.9
	15	96	6.79	.00170	2, 103	47.13 ± 1.42	1.86 ± 0.54**	1.01 ± 1.06	10.1
	1	166	5.22	.00690	2, 103	45.90 ± 1.47	1.18 ± 0.55**	0.62 ± 1.09	7.6
Uterine length (cm)	7	148	5.71	.00440	2, 104	701.50 ± 52.55	20.52 ± 33.51	−180.87 ± 57.60	8.3
	5	1	5.13	.00750	2, 104	623.57 ± 50.35	87.20 ± 27.39**	−31.54 ± 54.11	7.4
Piglets born/litter	6	102	5.15	.00750	2, 98	8.76 ± 1.08	−0.81 ± 0.49	1.94 ± 0.65**	7.8

Bold font indicates F-ratios *suggestive of linkage* according to the criteria of Lander and Krugyak (1995) and applying the adjusted threshold criteria ($p < 0.0005$).
[a] Homozygotes deviated from the mean of homozygotes.
[b] Heterozygotes deviated from the mean of homozygotes.
[c] Reduction in residual error variance owing to the presence of the QTL in the model.
* Effect was different from 0 based on the T-test (*p*-value < 0.05).
** Effect was different from 0 based on the T-test (*p*-value < 0.01).

was found on SSC8 (F-ratio of 8.89 at 101 cM), and the third, involved in gestation length, was located on SSC9 (F-ratio of 9.30 at 135 cM). A summary of the estimates for these QTL effects is presented in Table 2, with details of the chromosomal locations of each trait in Figures 1 thru 4.

The putative locus (F-ratio of 9.97 with a peak at S0227) affecting the number of stillborn piglets accounts for 15.5% of the F_2 variance (Table 2 and Fig. 1). The additive effect of −0.31 ± 0.11 indicates that sows inheriting both alleles at this locus from their Meishan grandparents produced 0.62 fewer piglets per litter than those inheriting alleles from Yorkshire grandparents, whereas heterozygous gilts produced 0.57 (±0.18) fewer piglets per litter than the gilts inheriting both alleles from the purebred Yorkshire grandams (number of stillborn piglets dominance effect; Table 2). The ratio of dominant to additive effects is 1.84, suggesting dominance or underdominance.

Suggestive evidence for a putative QTL contributing to the variance in number of corpora lutea at position 101 on SSC8 accounts for 13.2% of the F_2 variance (Table 2). The additive effect indicates that Meishans boars contributed negatively to the number of corpora lutea observed in the F_2 gilts (−1.20 ± 0.37). Marker SW444 is at the maximum of the SSC8 peak (Fig. 2), suggesting this marker is located near the QTL. White and coworkers (1993) observed that gilts of the Meishan breed had a larger number of corpora lutea than Yorkshires, prompting a simple least squares regression analysis (where the phenotype of number of corpora lutea was the dependent variable) to verity that the Meishan grandparents did, in fact, contribute a genotype that resulted in the observation of fewer corpora lutea in the F_2 gilts (*p* value of 0.0008; data not shown). With coded genotypes as the three levels of the independent variable, a one-way analysis of variance (ANOVA) also verified that Meishan grandparents contributed a genotype associated with fewer corpora lutea. The mean (±SD) number of corpora lutea observed was 14.34 ± 2.34, 13.95 ± 3.00, and 17.09 ± 3.91 for the F_2 gilts homozygous for alleles contributed by Meishan boars, heterozygotes or homozygous for alleles contributed by the Yorkshire sows, respectively (*p* value of 0.00). This analysis showed that gilts receiving both alleles from their Yorkshire grandams had an increased number of corpora lutea over heterozygotes and homozygotes receiving both alleles from Meishan grandsires. A multiple step-wise linear regression considering genotype, family, and parity in the full model showed that only genotype contributed significantly to explaining the observed variation in phenotype. (The resulting F value for the difference between the full and reduced models was 1.22, with 2,104 df, which is not significant at the 0.1 level).

Results for a chromosomal region affecting the number of corpora lutea (on SSC8) were sufficiently interesting for further analysis, with the maximum peak closest to marker SW444 in the interval flanked by SW905 to S0086 (Table 2 and Figure 2). Smaller peaks were also observed near SW764 on SSC7 and in the interval between SW906 and SWR2121 on SSC15 with F-ratios of 6.22 (*p*-value = 0.00029) and 6.20 (*p*-value = 0.0028) respectively. Although the latter small peaks were not significant by either of the criteria we used, the three peaks together account for 31.4% of the F_2 variance in number of corpora lutea.

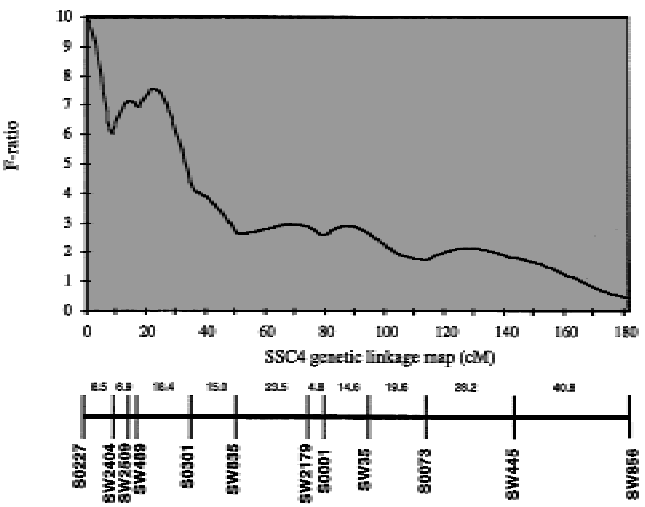

Fig. 1. Evidence for a possible QTL associated with number of stillborn piglets on SSC4.

The additive effect for the putative QTL on SSC8 affecting number of corpora lutea (Table 2) indicated that gilts receiving both alleles from their Meishan grandsires ovulated 2.4 eggs fewer than gilts receiving both alleles from their Yorkshire grandams, whereas heterozygous gilts ovulated an average of 1.76 fewer eggs than the gilts inheriting both alleles from the purebred Yorkshire grandams (number of corpora lutea dominance effect, Table 2). The ratio of the dominance effect to the additive effect was 1.46, again suggesting underdominance.

The third putative QTL detected affects gestation length, is supported by an F-ratio of 9.30 (significant at the $p < 0.0005$ level), and is located on SSC9 with a peak between markers SW174 and SW1651 (Fig. 3). The additive effect of gestation length on SSC9 was observed to be 1.52 days (Table 2). Thus, gilts that inherited both alleles from the Meishan founding boars were observed to have a gestation length of 3.04 days longer than gilts that received both copies from the Yorkshire sow founders. The dominance effect of the QTL involved in gestation length on SSC9 was −2.34 days. From this observation, we can say that heterozygous gilts are

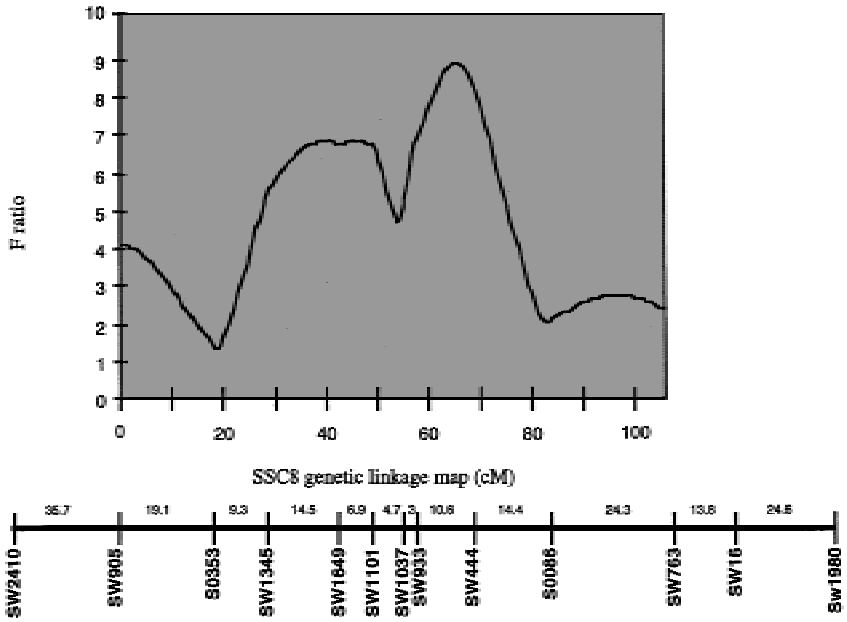

Fig. 2. Evidence for a possible QTL associated with number of corpora lutea on SSC8.

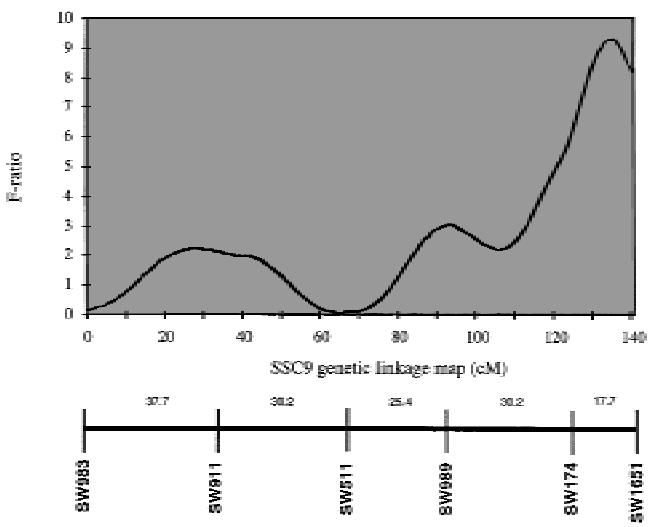

Fig. 3. Evidence for a possible QTL associated with gestation length on SSC9.

expected to have a gestation length of 2.34 less than the average of the homozygotes. The dominance to additive effect ration was −1.54, again evidence for dominance or underdominance. The F-ratios for the remainder of the traits included in Table 2 and presented in Figure 4 (bars) were significant with only nominal (pointwise) criteria (p-value < 0.05), but are included here because they are biologically relevant to litter size, a characteristic of economic importance that has been frequently selected for by breeders.

Discussion

The least squares regression analysis method requires the assumption that the parental breeds crossed in producing the reference family were fixed for alternative alleles for all loci involved in a particular trait. The assumption is necessary to ensure that the estimate of additive and dominance effects can be interpreted as the effect of a single QTL locus. If the assumption is violated, the interpretation of these effects may not be owing to a single locus, and the power to detect a QTL will be increasingly reduced and its effect underestimated (Haley et al. 1994). Lack of fixation of QTL alleles in the purebred parental lines could lead to erroneous estimates of additivity and dominance, but can not account for our observation of effects that are contrary to expectation. There may be other major loci affecting these traits that we have not detected with the markers used in this scan. Also, in our study, it is not possible to differentiate between QTL effects that are due to a single QTL with a large effect or to multiple genes in close proximity that affect the same trait. Georges and associates (1995) point out the risk that a QTL study with low or moderate power may overestimate QTL effects because only QTLs with inflated estimates that are due to chance will reach the significance threshold. Our study may suffer from this problem owing to the limited sample sizes, especially since most of the traits examined show low heritability (Beavis 1998). The samples sizes are limited by the costs of producing, collecting phenotypic measurements, and genotyping the study kindred. Thus, we expect inflated estimates of the effect of the QTLs detected and that confirmatory studies in other pedigrees will be necessary to verify the presence of the putative QTL detected. Of greater concern is the possibility that real QTLs will be missed (high rate of Type II errors; van Ooijen, 1992), and for this reason we report results that are *suggestive* (Lander and Kruglyak, 1995) and of nominal significance to promote the possibility that data may be summarized across several similar studies in the future as suggested by Andersson and coworkers (1998).

Our results for SSC8 (Fig. 2) are consistent with those published by Rathje and colleagues (1997) suggesting a QTL for ovulation rate on SSC8, but are not consistent with their more weakly supported locations on SSC13, 15, and 4. Comparison of the two studies for a possible QTL on SSC15 indicates that the intervals with the maximal F-ratio are adjacent rather than overlapping. It is possible that we may be detecting the same QTL affecting number of corpora lutea or ovulation rate on SSC8 in the two study populations comprised of different pig breeds; the Meishan and Yorkshire breeds in this study and the White Landrace lines selected for increased ovulation rate and embryonal survival studied by Rathje and coworkers (1997) and Casey et al. (1994).

Like Rathje and associates (1997), we did not find a significant QTL affecting the number of piglets born per litter, although we did observe an additive effect that suggested F_2 gilts homozygous for Meishan alleles produced 1.62 (±0.49) fewer piglets pet litter,

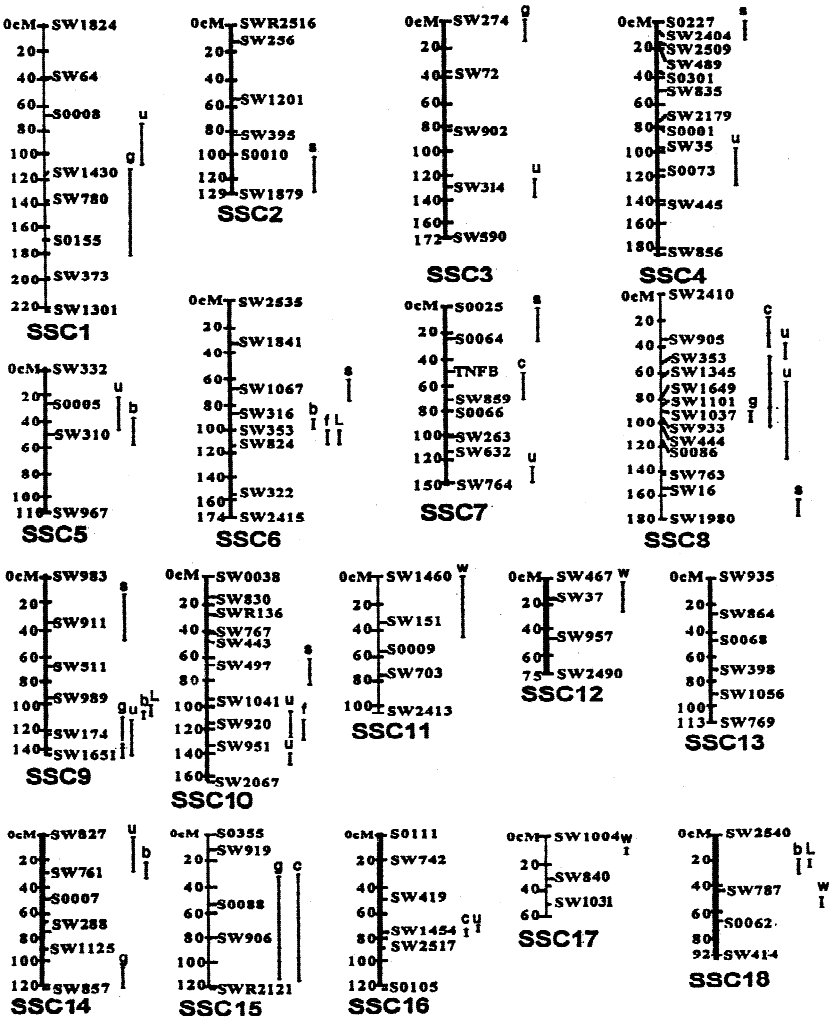

Fig. 4. Chromosomal location of traits detected by genomic scan. Vertical bars show traits associated with regions of F-ratio >3.08, corresponding to a nominal *p*-value < 0.05, and b = piglets born, c = number of corpora lutea, f = total fetuses, g = gestation length, l = piglet born live, s = stillborn piglets, u = uterine length, and w = piglets weaned.

and heterozygous gilts produced 1.94 more piglets per litter than the mean of the homozygotes. We also found a linkage suggestive of a QTL affecting the number of stillborn piglets on SSC4 closest to marker S0227 (Table 2 and Fig. 1). It is interesting to note that the observation of suggestive QTLs for corpora lutea and stillborn piglets (Table 2 on SSC8 and 4, respectively) did not singly or cumulatively have a significant effect on the observed number of piglets born per litter (F-ratio 5.15, *p*-value = 0.0075); although this does not exclude the possibility that these loci contribute, along with other loci, to the number of piglets born.

Bennett and Leymaster (1989) proposed and extended (Bennett and Leymaster 1990) a model for litter size at birth that was dependent on ovulation rate, embryonic viability, and uterine capacity. They concluded, from simulations based on experimental results, that independent selection for either ovulation rate or uterine capacity will not result in large changes in litter size. Their simulations support the idea that selection for genes affecting multiple traits is required to increase litter size. Our scan of the swine genome for markers linked to these traits in the Meishan and Yorkshire breeds that differ in litter size found suggestive evidence for QTLs involved in ovulation rate (number of corpora lutea), embryonic viability (number of stillborn piglets), and gestation length. If uterine capacity is taken to be a function of number of corpora lutea divided by the uterine length, then the F_2 mean on SSC8 associated with number of corpora lutea divided by the F_2 mean on SSC7 associated with uterine length would be, 17.18 / 701.50 or 0.024 corpora lutea per centimeter of uterine length. Our results are consistent with Bennett and Leymaster's model in that we detected suggestive evidence for differences in multiple traits associated with the documented differences in the litter size between Meishan and Yorkshire breeds. Our results imply a causal relationship between selection for a single characteristic (litter size) and changes in the genes specifying the traits of at least ovulation rate and embryonic viability (as well as gestation length in our study).

The traits we have studied here are life history traits that clearly affect viability and fertility and therefore the contribution of offspring to the next generation. Thus, they are tied very closely to fitness of the individual or its adaptive value that is subject to selection and results in the change in frequency of the genes and genotype we observed in the next generation. Falconer and McKay (1996) propose a hierarchy of the causes of variation in fitness with the number of offspring produced or fertility (here litter size) and quality of offspring (here number weaned) representing primary components of fitness. Individual fitness is difficult to measure directly and to separate from parental fitness, so they suggest that it can be estimated by combining values of the components at contributing levels (here we measured litter size and viability or number born live or stillborn). Of the traits measured in this study, number of corpora lutea, uterine length, and gestation length influence the litter size. The variation in these characters and other, less direct and less obvious influences, not measured in this study or even identified as relevant, may be associated with the variation of fitness. If thought of as selection on single traits, the estimated magnitude and direction of selection may be misleading, since selection acts on fitness and simultaneously on many phenotypi-

cally correlated traits (Falconer and McKay 1996). Thus, it is not particularly disturbing that all three QTLs suggested by this study result in effects that are in conflict with the parental phenotypes.

Since all three putative QTLs appear to be dominant or underdominant, the genetic variation for fitness may be at the selection limit rather than increasing, as would be predicted by Fisher's fundamental theorem if the variance of fitness were additive and, therefore, likely to be owing to deleterious recessives maintained at low frequency by mutation balancing selection. Also, Falconer and McKay (1996) point out that inbreeding is known to reduce characters connected with reproductive capacity and is expected when selection is applied to a metric character that is not fitness itself, as was done in this study. This results in reduced fitness unless the character is controlled entirely by genes with no effect on fitness. Also, the relative fitness of a genotype is not necessarily the same in all individuals and presumably is reduced when coadpated gene complexes are broken up by between-breed crosses.

In summary, the experimental method used in this study appears to have potential for identifying QTLs affecting traits of economic importance and is essential for selection with multiple markers simultaneously to enhance the response to selection for desired characteristics in marker-assisted selection (MAS) procedures that are biologically related, such as reproductive traits. However, it is apparent that extending this study to include larger resource families will probably be necessary to increase the power to verify our current observations. Studies of additional crosses will be necessary to confirm the validity of our suggestive observations. A radiation hybrid map that includes the markers used in this study is currently in progress, and these physical measurements will improve our estimates of the distances between markers (derived in this study by linkage analysis) to facilitate narrowing of the intervals associated with the putative QTL identified by us. High-resolution maps of regions surrounding QTLs will be important for identifying additional markers more tightly linked to traits of interest, and that can be assembled into marker multiplexes for efficient MAS in large production herds.

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