Testing for quantitative trait loci for lactation persistency in dairy cattle

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ABSTRACT

Lactational persistency has not been selected for in New Zealand dairy populations. Existing milk production data and pedigrees from New Zealand Holstein-Friesian cattle at pasture, recorded in the Livestock Improvement Corporation database were utilized with a grand-daughter design, for a genome-wide search for a quantitative trait locus for persistency of lactation. Persistency was defined as a weighted sum of test-day yields. A significant region was identified in one grandsire family on Chromosome 3 at microsatellite marker position AGLA247 (chromosomal position 67cM; $P < 0.05$). To verify this result, data from a research dairy herd defining persistency as the mean of three weekly tests of daily yield in March (late lactation) divided by the equivalent daily yields in October (early lactation)) for three seasons beginning in 1997/98 were used in a phenotypic association study on Chromosome 3 only. One allele of marker HUJ246, close to the original region appeared to show an additive effect. The maximal difference between this genotype and the others at this marker locus was 12.3 ± 4.4% of the mean persistency. The potential use of this marker in the New Zealand dairy cattle population depends on the current allele frequencies in the industry.

INTRODUCTION

Lactational persistency, the rate of fall in production during lactation, though a desired dairy performance trait, has not been directly selected for in New Zealand dairy populations. In spring-calving pasture-fed cows, peak lactation is reached soon after calving and then declines by about 10% per month (Davis *et al*., 2000) until the cows are dried off. It is known that some cows maintain their milk production through the milking season better than others. The shape of the lactation curve is of economic interest to the New Zealand dairy industry as the industry must have enough processing capacity to cope with the volume of milk at peak lactation, hence much processing plant is under-utilised at other parts of the lactation cycle (Byles, 1995), since the whole industry is seasonal in its calving pattern. Lactation persistency in New Zealand dairy cattle is heritable, although not as highly heritable as lactation milk yield. Unpublished industry estimates of heritability for persistency in New Zealand Holstein-Friesians (HF) are 0.22 for milk yield, 0.21 for fat yield and 0.16 for protein yield. Corresponding figures for New Zealand Jerseys (J) are 0.16, 0.25 and 0.16, respectively. Heritabilities for the estimated lactation yields of milk, fat and protein from the same data set gave values of 0.30 to 0.39. Phenotypic standard deviations for persistency, defined as the difference between production in late and early lactation, are 155 litres (HF) and 114 litres (J) for milk persistency, 6 kg for fat persistency and 4.5 kg for protein persistency.

Selection can be used to genetically improve persistency while maintaining constant total lactation yield (Macciotta *et al*., 2006). Several quantitative trait loci (QTL) for lactational persistency have been identified. These include an association of toll-like receptor 4 polymorphisms in Holstein bulls on chromosome 8 (Sharma *et al*., 2006), while 12 chromosomes have been shown to carry significant QTL effects for persistency of milk yield (Harder *et al*., 2006). In three U.S. Holstein families (Dairy Bull DNA Repository families), marker BL41 on chromosome 3 was associated with a decrease in milk yield during mid-lactation in one family, and marker HUJI177on chromosome 3 was associated with changes in the milk yield and protein percentage curves in another family, suggesting a QTL in the region. Interval mapping approaches detected QTLs in three possible locations on chromosome 3 including one between 62 and 72 cM (Rodriguez-Zas *et al*., 2002). The present study was designed to search for persistency

QTL in HF and J cows under New Zealand grazing conditions, and to attempt to validate any such QTL in an independent population.

MATERIALS AND METHODS

Industry study

A granddaughter experimental design (Weller *et al*., 1990) was used to evaluate marker-QTL associations in seven HF and two J families, in a 1999 Livestock Improvement Corporation study. The average number of sons per grandsire was 29, ranging from 21 to 47. The University of Liege, Belgium, provided genotype results for all individuals, with 292 microsatellite markers, mapped to the 29 bovine autosomes, an average of 10 markers per chromosome.

Three persistency traits for milk volume, fat and protein yields were investigated. Persistency was defined as the difference in production between late and early lactation. The day defining the boundary between early and late lactation was determined such that persistency was genetically independent of lactation yield. This occurred at about the 120th day in milk. A persistency phenotype for each cow was then estimated by combining test-day yields using selection index theory. Persistency breeding values were derived from production data from Sire Proving Scheme herds with first lactations commencing in the spring seasons 1987 through 1997.

QTL analysis

QTL analysis was undertaken using multimarker regression principles as developed by Knott *et al*. (1994). Confidence intervals for the QTL location were determined by bootstrapping (Visscher *et al*., 1996) from the distribution of 20,000 most likely positions.

Validation study

In order to test the above QTL findings independently in a research dairy herd, data on persistency defined as the mean of three weekly tests of daily yield in March (late lactation) divided by the equivalent daily yields in October (early lactation)) for three seasons beginning in 1997/98 were used in a phenotypic association study on chromosome 3 only, where the most significant QTL was identified. A total of 170 cows were present on 1 March 2000, when blood samples were taken for DNA analysis. There were data for 180 cows in the persistency data analysis, with a total of 316 records of persistency in 1997/98 to 1999/2000. In the final data set of those with DNA analysed, there were 142 cows with 271 persistency records. A repeated-animal restricted maximum likelihood analysis (Gilmour *et al*., 2006) was carried out, without pedigrees, to combine the "residuals" across years, so that one overall value resulted for each animal, and to fit covariates for various markers or combinations of markers. The chromosome 3 markers tested were BM4129 (at 52.459 cM), HUJ246 (67.982 cM), BM6465 (69.15 cM) and BMS2145 (93.827cM) (Ihara *et al*., 2004).

RESULTS

In the industry data, the three persistency traits were highly correlated, with correlation coefficients of 0.75 between milk and fat yield persistency, 0.82 between fat and protein yield persistency and 0.85 between milk and protein yield persistency. Principal component analysis showed that a single independent trait accounted for 87% of the variation. Experiment-wise critical values were calculated assuming a single independent trait (Table 1). It should be noted that critical values differ slightly between traits.

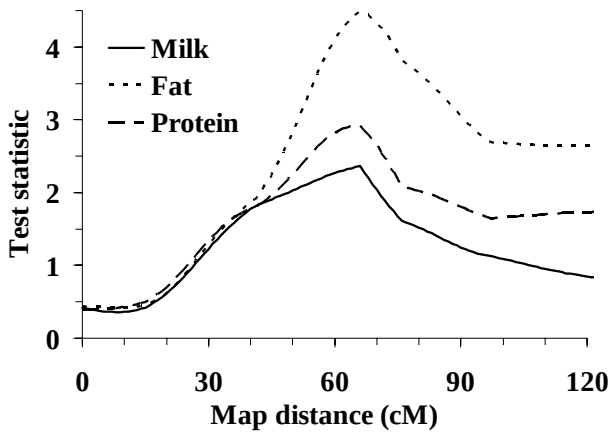
TABLE 1: Experiment-wise (error rate) threshold levels for *F* statistics for the three traits (50,000 shuffles).

Threshold level		Persistency trait		
		Milk yield	Fat yield	Protein yield
Highly significant	0.1%	4.89	4.70	5.29
Significant	5%	3.74	3.72	4.10
Suggestive	10%	2.86	2.85	3.04

The most significant QTL was identified on chromosome 3 (Figure 1), with suggestive QTL identified on two other chromosomes. Across-family QTL analysis revealed a QTL for fat yield persistency. This was significant in one grandsire family at position 67cM on chromosome 3 ($P < 0.05$), and suggestive in a similar genetic location in two other families. Note that only eight informative families were included in the analysis.

QTL positions varied slightly between families, at 64cM, 66cM and 72cM. The QTL was positioned

FIGURE 1: Test statistic versus location on chromosome 3 for three persistency traits; milk yield, fat yield and protein yield.



on the marker AGLA247 (Georges *et al*., 1995). The effects for the three families ranged from 1.52 to 2.50 kg of fat or approximately 0.25 to 0.42 phenotypic standard deviations.

Ninety-five percent confidence intervals for the QTL were calculated with bootstrapping, using all families and only the segregating families. The 95% confidence interval was 58 - 70 cM with the segregating families, with the peak occurring at 67 cM, 6.3 times higher than the nearest shoulder, while for all families it was a larger range from 44 - 86 cM.

In the research herd used for validation, the repeatability of persistency of milk yield across the three lactations was high at 0.45 ± 0.07, when genomic effects were fitted simultaneously with the repeated-animal term (see below).

Two microsatellite markers were associated significantly with persistency in this herd, i.e., HUJ246, P <0.0004; and BM6465, P <0.0086, when the two markers were fitted together. Additive and dominance effects for HUJ246 and BM6465 were fitted simultaneously with the repeated-animal model. The HUJ246 "c" allele was associated with a gene with negative effect on persistency. The additive difference between zero and two copies of the allele relative to a population mean of 0.445, for mean March milk yield/ mean October milk yield was associated with a 2 x -0.0274/ 0.445 or 12.3% difference in persistency (P <0.006, standard error (SE) 0.0196, *t*-value -2.79). There was, however, an effect of opposite sign on dominance (estimate 0.0220, SE 0.0120, *t*-value 1.83).

Significance tests for the BM6465 microsatellite ("c" allele), when tested alone, were the same sign and nearly as large in magnitude as those for HUJ246, with an estimate of the additive effect = 2 x -0.0197/ 0.445, or 8.9%, SE. 0.0214, *t* value = -1.85; dominance estimate = 0.0201, SE = 0.0138, *t*-value = 1.46, but effects for BM6465 tested in addition to HUJ246 were not significant. For the two loci combined, comparing zero to four copies of "c" as for both the HUJ246 "c" and the BM6465 "c" alleles, the significance test for the covariate effect had an *F* ratio of 7.89 (P = 0.005). The two genotypes, "cc" versus "non-c, non-c" at *both* loci had a difference in persistency of 0.1493, or 33.6% of the mean. Depending on the model fitted for persistency, the "effect" of the microsatellite(s) ranged from 7 to 14% of the phenotypic variance.

DISCUSSION

A significant QTL for fat yield persistency was identified on chromosome 3 in the industry data with the marker AGLA247 at position 67cM. This QTL was observed to be segregating in three families. The 95% confidence interval ascertained through bootstrapping was between 58 and 70 cM. The QTL identified is an interesting discovery due to the level of significance and the consistency of within-family peaks. The QTL is some 20 cM from that reported by Heyen *et al.* (1999) for fat percentage.

The markers used in the validation study of HUJ246 (68cM), BM6465 (69cM) are located very close to the marker AGLA247 used in the industry study. The observation that the results of the validation study in a different population of animals predicted a QTL at the same location as the industry study indicates that a QTL probably exists at this location.

The ranking of cows for persistency from year to year was highly repeatable (r = 0.45 ± 0.07 in the validation data). This high value was consistent with Livestock Improvement Corporation estimates of 0.40 to 0.45 (D.L. Johnson, Personal communication.).

Markers AGLA247 and HUJ246 could be used to select New Zealand cows with better lactational persistency. The potential use of these markers in the New Zealand dairy cattle population depends on the current allele frequencies in the industry.

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