LoinMAX™ and MyoMAX™: taking DNA marker tests from the research environment to commercial reality

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ABSTRACT

Meat yield is an important component of most sheep breeding programmes. To improve meat yield, breeders use a variety of tools including marker assisted selection (MAS). Ovita funded AgResearch to develop two marker tests to assist in selection for superior meat yield. These have been branded MyoMAX™ and LoinMAX™ and originated from quantitative trait loci (QTL) studies over the past decade. In both cases the development of these commercial marker tests involved refining the QTL locations, identifying informative marker haplotypes and validating these in industry flocks. Both the LoinMAX™ and MyoMAX™ beta tests were field validated using a number of commercial breeders. The validation process showed that the phenotypic effects of both LoinMAX™ and MyoMAX™ were significant in different genetic backgrounds and that there were no negative correlations between the presence of LoinMAX™ and MyoMAX™ with other traits, such as meat quality and growth rate. Both LoinMAX™ and MyoMAX™ have now been successfully commercially launched by Catapult Genetics Ltd and are being widely used in industry.

Keywords: sheep; LoinMAX; MyoMAX; meat yield; validation.

INTRODUCTION

LoinMAX™ and MyoMAX™ are two commercial DNA tests for the presence of specific meat yield QTL in sheep that have been released by Catapult Genetics in the last two years. LoinMAX™ and MyoMAX™ are both haplotype tests and are based on the knowledge that animals inheriting a particular gene variant of interest (for example, affecting a production trait like meat yield) will also tend to inherit a broader region of chromosome surrounding that gene. DNA markers (segments of DNA for which multiple variants exist in the sheep population) are identified in this broader region and the haplotype test is formed using two or more of these markers in combination. The test is designed by identifying a specific combination of DNA marker variants (alleles), referred to as a haplotype, that is associated with inheritance of the desirable gene variant, but which is absent, or very rare, in animals which do not inherit the desirable gene variant.

Typically, a potentially informative haplotype of DNA markers will arise out of a QTL and fine-mapping research programme for a gene variant of interest. Further research will often be in process to identify the specific gene mutation responsible for the phenotypic effect observed (*e.g.* increased meat yield) but this can take many years. The Booroola and Inverdale mutations (Galloway *et al*., 2000; Wilson *et al*., 2001) took almost a decade to identify from the point at which the broad chromosomal regions containing these genes were located. The advantage of haplotype tests is that it is only necessary to know the broader chromosomal location of the gene variant, not the underlying mutation. Thus, a haplotype test allows a desirable gene variant to be utilised without having to wait for the causative gene variant to be identified.

One disadvantage of haplotype tests is that they are an indirect test (since they are not based on the causative gene variant) and are subject to a number of potential errors. Sources of error in haplotype tests include errors in the determination of an animal's haplotype, the presence of animals which carry the haplotype but do not carry the causative gene variant and the presence of animals which carry the causative gene variant but do not carry the haplotype. Therefore, haplotype tests require careful validation before being released commercially.

The process of developing a potentially useful haplotype from a research flock into a commercial test, usually begins with an assessment of the frequency of that haplotype in a sample of animals representative of the New Zealand commercial flocks. For this purpose we assembled a panel of DNA samples derived from New Zealand commercial flocks and representative of the major New Zealand sheep breeds. The second phase of the validation is to confirm and measure the size of the phenotypic effect associated with inheritance of

the desirable haplotype in commercial flocks and to check that there are no deleterious effects on other traits. This beta testing of the LoinMAX™ and MyoMAX™ was performed using a number of commercial breeders. This paper, describes the process by which the LoinMAX™ and MyoMAX™ commercial tests were developed, starting from the original QTL/fine mapping research programmes.

DEVELOPMENT OF THE 'LOINMAX™, TEST

LoinMAX™ is a test for a QTL on sheep chromosome 18 affecting rib-eye (*Longissimus dorsi*) muscling. The LoinMAX™ QTL was first reported in a 'Meat Elite' breeding programme developed among Australian Poll Dorset breeders and Landcorp in 1989/90. A small unpublished study found there was evidence for linkage with a single marker at the telomeric end of sheep chromosome 18 (Banks, 1997). Semen from two putative carrier rams was imported into New Zealand by Landcorp Farming Ltd and AgResearch and was used to inseminate 325 predominantly Romney breed ewes as part of a QTL study (Nicoll *et al.*, 1998). This study confirmed the presence of a QTL in both sires for increased L. dorsi muscling at the telomeric end of sheep chromosome 18. Carriers had 1.14 to 3.30 cm^2 larger L. dorsi area at the twelfth rib, with an overall increase in L. dorsi weight of 8%. No effects on other muscle groups or fat depots were identified. A subsequent study (Jopson *et al*., 2001) demonstrated that there was no difference in tenderness between the L. dorsi meat of carriers versus non-carriers subjected to an industry standard accelerated aging and conditioning protocol. This latter study also demonstrated that the LoinMAX™ QTL is not additive, with animals inheriting a second copy of the QTL displaying no additional increase in muscling.

Further fine-mapping work (McLaren, R.J. unpublished data) has now narrowed the location of LoinMAX™ to an approximately 350 kb region of DNA at the telomeric end of sheep chromosome 18. From this region, a potentially useful haplotype of three DNA markers spanning the region was identified and these were genotyped through a New Zealand breeds panel. A significant frequency of the haplotype was found only in the Poll Dorset breed, the breed in which LoinMAX™ QTL was first identified. This haplotype test is currently a trade secret of Ovita licensed to Catapult Genetics.

In order to confirm the association between the inheritance of the LoinMAX™ test haplotype and the increased L. dorsi muscling, we examined data from three breeding groups found to be using sires with the haplotype; MegaMeat, Southern Poll Dorset and Rissington Breedline. The association between the LoinMAX™ haplotype and increased rib-eye muscling was confirmed in all three flocks using several different methods of phenotypic measurement (including both live animal and carcase data). Data from Rissington Breedline confirmed that progeny inheriting a single copy of LoinMAX™ had on average 7.8% greater boneless loin weight than the non-carriers.

DEVELOPMENT OF THE 'MYOMAX™, TEST

A QTL affecting muscle and fat traits was identified in New Zealand Texels on sheep chromosome 2, in the region of the *GDF8*-myostatin gene (Broad *et al*., 2000). The presence of this QTL was confirmed as part of a Ph.D. programme (Johnson, 2003) and shown to increase leg muscle (only legs and loins were analysed at this point) and decrease fat in the leg (Johnson *et al*., 2005). A potentially useful haplotype (MyoMAX™, Trade secret of Ovita licensed to Catapult Genetics), consisting of two markers, was identified in the QTL region and these two markers were genotyped through a New Zealand breed panel consisting of: Dorset, Texel, Romney, Peredale and Coopworth animals. A high frequency of the haplotype was observed in Texels, the breed in which this QTL was identified but the haplotype was either not detected or detected at only a low frequency in the other tested breeds.

Validation and beta testing of the MyoMAX™ test occurred during 2005-2006. The effect of MyoMAX™ was examined in different genetic backgrounds and in composite sheep (sourced from Mt Linton and Texel sire reference group's progeny test). Viascan phenotypic data was collected from three slaughters and additional carcass measurements were also taken. There were no significant differences between these slaughters so the data was pooled for analysis purposes. Analysis showed that the MyoMAX™ haplotype significantly increased weight of lean loin, leg and shoulder and significantly decreased GR. Therefore we were confident that the MyoMAX™ haplotype has the same effect in different genetic backgrounds. The additive model best fitted our data and it is therefore probable that two copies of MyoMAX™ have double the effect of one copy.

We also examined the effect of MyoMAX™ on lamb survival, by looking at the MyoMAX™ status of lambs in one flock and the MyoMAX™ status of ewes in another flock. There was no significant effect of MyoMAX™ on lamb survival.

SUMMARY

LoinMAX™ and MyoMAX™ were both validated before commercial release. The validation process consisted of confirming the phenotype in commercial flocks and evaluating correlations of the phenotype with other important traits such as lamb survival. The effects of LoinMAX™ and MyoMAX™ were consistent in different genetic backgrounds and had no negative effect on other production traits. Therefore we were confident that the tests would be useful in selection for meat yield in a commercial sheep breeding programme. The strategies by which commercial breeding programmes are now using LoinMAX™ and MyoMAX™ depends on the frequency of the alleles in their flocks.

ACKNOWLEDGEMENTS

The authors acknowledge this report is part of a much larger body of work with many people contributing. Particularly we thank the breeders for supply of phenotypic information and assistance from AgResearch staff, Gordon Greer, Benoit Auvray, Mary Wallace and staff at GenomNZ.

Prospects for genome wide selection in the New Zealand livestock industries

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ABSTRACT

A number of DNA marker applications aid genetic selection within subpopulations of New Zealand's livestock farming industries. Advances in genomic tools have opened the possibility of genotyping individuals for a large set of markers, enough to have a large proportion of the genome 'tagged' with at least one of these markers. This creates opportunities for genome wide selection (GWS) – the selection of breeding animals based on very large numbers of marker genotypes. There are a number of requirements for GWS to work, including the existence of genotyping technology to provide genotypes at a sufficient density, tractable statistical methods for estimating marker effects to use in prediction models and suitably phenotyped populations which when genotyped allow estimation of relationships between markers and traits. Additional phenotyped populations are required to validate their predictive ability and demonstrate economic benefits to breeders and to the commercial tier. A high density genotyping platform is available for cattle, but may need further developments. Plans are underway for a similar platform for sheep, but not for deer. The dairy industry is well placed to implement GWS, as there is a large bank of DNA samples of industry sires representing most of the genome variation present. Genotyping these animals will allow marker effects to be estimated and the reduced generation interval arising from selecting young sires should easily overcome any decrease in selection accuracy compared with progeny testing. Industry structures are likely to evolve to allow a return to entities undertaking GWS. GWS will be most beneficial for traits with low heritability, that cannot be measured on selected parents and/or that are expensive to measure. Genes of known effect may continue to be accounted for individually, particularly if they exhibit non-additive effects.

Keywords: genome; marker assisted selection; dairy; beef; sheep; deer.

INTRODUCTION

The last century saw selection in livestock breeding programmes advance from ad-hoc visual and family-based procedures to the use of quantitative information to provide selection rankings based on predictions of genetic merit. New statistical models that incorporate information from multiple relatives and multiple traits and their implementation using computers have enabled this development. The focus has been on a relatively small number of highly heritable traits that are relatively simple to record. Most applications have used the additive infinitesimal model, whereby a trait is assumed to be influenced by an infinite number of genes each with an infinitely small, non-interacting effect. While this has been and remains a powerful tool for industry improvement, the limited breadth of focus has constrained the potential overall benefits.

The end of the last century saw the advent of molecular marker maps for livestock which