

Moving from Microsatellites to SNPs

Introduction

Advances in molecular biology, especially genomics, provide a new set of information to be used in the animal industry. On one hand, the use of molecular information may contribute to the enhancement of consumers' trust in the ability to monitor and control the animal production chain. On the other hand, molecular information will greatly contribute to the achievement of genetic improvement for animal traits through the use of marker assisted selection (MAS), gene introgression, heterosis prediction, and advanced pedigree validation.

In most cases, advantages of using molecular information through genomic evaluations, comes from improved accuracy of animal breeding values, shortened generation intervals and increased intensity of selection. Nevertheless, there is still a need for research and development in the search for associations between genetic markers and traits of interest.

Background & important Terms

Microsatellites

A Microsatellite is a DNA sequence with repeatable base pairs. The DNA segments can range in length from 1 to 6 or more base pairs, which typically repeat 5 – 50 times. In cows 12 (as recommended by ICAR) to 18 (additional loci used to aid verification) loci are used to verify parentage.

SNPs

SNPs are single nucleotide polymorphisms located throughout the genome – ie variations in a single nucleotide base occurring at a specific point in the genome. ICAR have set a minimum requirement to use at least 95 SNPs from the ISAG defined 200 parentage SNPs to be used. In addition if parentage discovery is likely to be needed – for example when the sire needs to be established, 400 SNPs are required.

MS imputation

Some SNP chips contain an extra 1000 to 2500 SNPs; using these it is possible to impute microsatellites from SNP haplotypes. This has been trialed in Ireland at ICBF for some time and has been shown to have 91% accuracy in Slovenia Brown Swiss.

A quote from Matt McClure who discovered these SNPs 'MS imputation is not a perfect process but it will help you transition from MS to SNP in a cost effective manner. Once you are using SNP you are then able to access all the extra things a SNP based panel brings—more accurate parentage, parent prediction, genomic breeding values, genetic defect/trait status,....'

Parentage verification and parental assignment

Prior to the emergence of SNP genotyping, parentage verification was one of the commercial uses of genetic markers. Traditionally, parentage testing was based on the exclusion of relationship (i.e.: sire or dam) when an animal has a genotype inconsistent to an expected relationship.

As the cost of the analysis decreases and the number of genetic markers available increases, breed societies are now able to build up pedigree records using genetic markers to track the pedigree of calves born in a herd at a given time. This requires a prior knowledge of candidate sires and dams for a set of identified markers. The probability of assignment to a correct pair of animals will depend on the number of alleles per loci, allelic frequencies in the population, the number of parents and number of possible matings.

The International Society of Animal Genetics (www.isag.us) has recommended a panel of 200 SNP markers for this purpose.

Sire Verification

Sire verification can occur where you have the genetics of a calf and its sire. This must be done with care, as several sires may be possible. It should be combined with farmer records to ensure the correct sire is attributed to the calf. It is NOT parentage verification as the dam is not taken into consideration.

Sire Discovery

Sire Discovery is where the calf's sire is unknown or may be one of several. The DNA of the calf is compared to all known sires and a probability is assigned to each sire that it is the parent of the calf.

SNP offers more for your money!

The major reason to change from MS to SNPs, is that you get far more for your money. If you have an animal run on a SNP panel, you can get parentage verified, your animal can be checked for genetic traits, and you can get a genomic evaluation all from the same sample!
SNPs are more accurate.

Microsatellite data will tell you who is **not** the sire, but cannot tell you exactly who **is** the sire. The primary reason is because each microsatellite provides limited information and so few of them are used for parentage analysis. For example if we say a calf has allele A for microsatellite MS1. The sire we're comparing the calf to does not have allele A for MS1 so he cannot be the sire. The problem arises when multiple sires have all have allele A at MS1, the calf could belong to any of them. Using more microsatellites helps but you can only do so much with 12 of them.

SNPs test anywhere from 100 (not very accurate) to 800 (very accurate) base pairs on the animals' genotype to verify parentage. Some laboratories are doing a full check on all 55,000 SNPs so if they say a sire is the sire of your calf you can be 99%+ sure that it is correct. This is also useful if you can get enough sires genotyped in your database you can then predict a sire.

The main points on why to go to SNPs:

- a. Imputation allows you to move forward with the technology without needing to re-test historical animals
- b. More accurate parentage
- c. Options for more information (Major Genes, Genetic Traits)
- d. Genomic breeding values can be calculated from the same sample

Issues

Moving to SNPs presents two main issues; price and the availability of genotypes.

Cost of SNP Genotyping

In some countries testing a DNA sample for SNPs is more expensive than testing for microsatellites. Cost depends on the size of the chip and on the number and tests. If only a parentage test is required, it could be difficult to justify the additional expense. However the additional information obtained far out ways the additional cost. Also the more samples analysed, the better price that can be obtained.

How to get access to the SNP genotypes

Today Bull SNP genotypes are more readily available than in the past. Dam SNP genotypes will be few, however usually a sample of dam DNA is required at time of verification. If Organisations began to verify parentage using SNPs, in the future the female calf genotypes would be available for SNP testing their calves. The main genotypes that Organisations could get quick access to are for bulls and possibly bull mothers and ET donor dams. Also more and more farmers are starting to test all the female calves.

How to move



Organisations need to carefully plan the move to using SNPs for parentage verification.

Suggestions for making this transition happen smoothly include

- a. Mandating that all calves should have DNA taken at birth and stored
- b. Requesting SNP genotypes for high-risk animals such as AI sires, male registrations and ET donor dams, ET calves and high genetic animals including some imported animals.
- c. Move to SNP parentage verification for all possible parentage checks including male registrations, ET donor dams, random parentage checks and farmer requests.
- d. Microsatellite imputation can be used where there is a mix of MS and SNP parentage, so long as the organisation uses a DNA lab that is able to impute microsatellites from SNPs.
- e. Use a lab that is accredited with the ICAR accreditation for DNA interpretation centres. The lab can be then accredited to access GenoEX-PSE and use the SNPs available on GENOEX for parentage verification on behalf of the organisation

More information on ICAR genomic services can be found at <https://www.icar.org/index.php/technical-bodies/working-groups/dna-working-group/>

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ICAR Guidelines

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