Tom Reed: Genomic selection is a very promising tool for sport horse breeding. Please explain the process.

Johan Knaap, director of the KWPN: The use of DNA information as a selection tool in breeding is nowadays common in farm animals like chicken, pigs and dairy cattle. In breeding sport horses it is a fully new tool for selection. In Friesian horses, tests are available to detect carriers of dwarfism and hydrocephalus, both birth defects caused by only one gene. However, most of the traits selected on in sport horse breeding are influenced by many genes, such as performance, behaviour and health criteria. The influence of many genes makes it far more complex to select horses. In that case it is beneficial to use DNA information of the whole genome or as much information as available. This is known as ‘genomic selection’.

Tom: A fundamental requirement for success of a genomic selection program is the development of a ‘reference population’, a large and representative sample of horses and their characteristics that accurately reflects the entire population of horses from which the tested horses are drawn. The KWPN and the members have invested a lot of money to develop a large and representative reference population, and I believe this is what sets this effort apart from some others.

Johan: First condition for genomic selection is a large and well balanced reference population. A reference population is a representative group of horses from which both phenotypes – our traits of interest – and genotypes – the DNA – are known. Comparing both delivers an algorithm that can be used to predict a trait in selection candidates. This is exactly what the Royal Dutch Warmblood Studbook, KWPN, did for osteochondrosis. From 2009 onwards all approved KWPN stallions had to undergo progeny testing for osteochondrosis.

From each stallion twenty randomly selected yearlings were X-rayed and

“The ultimate goal is genomic selection for jumping and dressage ability and behaviour.”
- Johan Knaap
classified for osteochondrosis in a scientifically-based scoring system by a KWPN X-ray commission. All financed by the KWPN, so the breeders were happy to collaborate. Based on this data, conservative breeding values were calculated and published for the breeders. However, the two disadvantages of progeny testing were firstly the costs of X-raying and secondly the extended time before selection. It took at least two to three years after approval of the stallion before breeding values could be published and before breeders could use this information.

Once established, genomic selection is cheaper and selection decisions can be made quicker. However, genomic selection for osteochondrosis was impossible without progeny testing. Progeny testing enabled the KWPN to set-up a reference population, as it rendered a beautifully balanced and large database with detailed and well-defined information on osteochondrosis from 3,000 yearlings. All 3,000 yearlings were genotyped. Subsequently, through research these genotypes and osteochondrosis classifications were compared. The outcome of this comparison was a DNA test that is, from a genetic point of view, three times more reliable than the observations from X-rays.

The results of this DNA test are used in breeding value estimation together with all the phenotypic data that is available. Nowadays, breeding values for osteochondrosis can be published as soon as stallions are approved without the need for any X-rays. The introduction of genomic selection was only possible because of the availability of well-defined phenotypes in a randomly selected large group of horses. Without high quality phenotypes from many horses, genomic selection is impossible.

**Tom:** How is the testing process done?

**Johan:** The process of testing selection candidates is simple. It starts with an agreement between the KWPN and the breeder in which the conditions for the DNA test, including the rights, are set. A hair sample is taken from which DNA is extracted in the laboratory and tested with the Illumina® equine HD array containing about 70,000 genetic markers, known as SNPs (single-nucleotide polymorphisms). The result from the laboratory of each selection candidate is a genotype for all the available genetic markers, which is send to the KWPN. To warrant quality, several controls are performed on the genotypes from each candidate, including parentage verification when possible. The KWPN, being the only one knowing the relation between the phenotypes and the genotypes, uses the genotypes of a selection candidate together with all the phenotypic data to estimate a breeding value. Mean of the breeding values is 100 with a standard deviation of 4. Publishing the breeding value for osteochondrosis enables the breeders to make a better match between mare and stallion in a compensation breeding model.

**Tom:** Setting a breeding value score of 96 as the minimum for approved stallions and elite mares means that approximately 14% of the population is excluded from being approved stallions or elite mares. This seems to me to be a reasonable approach: it allows for improvement of the population through selection but it is not so strict that a lot of good potential stallions may be excluded because their breeding value is less than 96. How does this exclusion rate compare to the percentage of stallions that were excluded from approval based on their X-rays?

**Johan:** The breeding value for osteochondrosis has to be 96 or higher for approval as a stallion in the KWPN breeding program. As a result, about 14% of the population is excluded, which is comparable to the selection intensity obtained using the former X-ray criteria.

**Tom:** In the past, KWPN has taken a ‘compensating differential’ approach to osteochondrosis in the stallion approval process: A truly outstanding stallion would be approved even if he had a chip in the hock, for example. And this would be disclosed to breeders. Did KWPN decide to adopt a strict selection criterion or is the compensating differential with disclosure approach still being used?

**Johan:** With a value of 95 or lower, stallions can only be approved in exceptional cases when extraordinary talents seem present. The stallion selection committee decides whether a stallion seems to have extraordinary talents to compensate for the low breeding value. For mares, the same selection criteria are used for the Elite predicate. When their breeding value for osteochondrosis is 96 or higher, they obtain the D-OC predicate. During a transitional period of three years, mare owners can choose to perform the DNA test or X-raying of their mare to obtain the Elite predicate.

**Tom:** Are more tests using genomic profiles likely to be offered and is there a projected schedule for release dates?

**Johan:** The DNA test for osteochondrosis was launched in April 2016 and until end of December of that year almost 2,500 were sold.

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