ests A ertain Question

DNA testing of horses has become increasingly common in recent years, but some degree of mystery and misunderstanding still surrounds the process. A better understanding of DNA testing illustrates how invaluable the testing can be, while at the same time it still has its limitations.

The History

UC Davis Veterinary Genetics Laboratory (VGL) in Northern California was a pioneer in the use of DNA to verify the parentage of horses. They were the first to offer DNA testing of horses and cattle in the mid-1990s. Today they offer DNA testing for 17 animal species. Equine DNA testing has become increasingly common and tests have been added to cover a variety of things from color testing to the heritability of certain diseases.

Horse registries have also begun to increasingly utilize DNA testing as a requirement for registration or for the approval of breeding stock. Most internationally recognized Warmblood and sport horse registries now require at least some degree of DNA testing as a part of the registration and/or breeding approval process.

Testing Your Horse

DNA testing is done using a sample of hairs from the mane or tail of the horse. Usually 20-30 hairs are required. The hairs must be pulled (and not cut) so that the root bulb is attached. Generally, the hair sample is pulled from the mane, but for foals it may be necessary to pull the hair sample from the tail.

The person collecting the hair sample should have clean hands and should be careful not to touch the roots of the hair, as this could contaminate the sample. Care should also be taken to keep all samples completely separate from each other. The hair sample is then submitted either to a

horse registry or directly to the lab, usually taped to a piece of paper. It is usually recommended not to submit hair samples in plastic bags as bacteria is more likely to grow on the sample, which could make testing more difficult or impossible.

The Basics

The laboratory will use the hair sample to extract DNA from the hair follicle cells. This DNA will then be used to identify "microsatellites" or pairs of DNA markers. A unique "DNA Marker Report" is then generated for the horse. The two most prominent labs in the United States for equine DNA testing are UC Davis Veterinary Genetics Laboratory (VGL) and the University of Kentucky (UK). VGL uses fifteen microsatellite markers to generate an equine DNA marker report, and UK uses thirteen.

An example of a DNA Marker Report provided by the author.

UNIVERSITY OF CALIFORNIA, DAVIS BERKELEY . DAVIS . IRVINE . LOS ANGELES . MY

EQUINE PARENTAGE AND GENETIC MARKER REPORT

FRIESIAN SPORTHORSE ASSOCIATION P.O. BOX 283 SEVILLE, FL 32190

Case: FSA18 Date Received: 24-Jul-2015

Name: LIBERTY ROF DOB: 07/04/2015 Sex: Reg: FSA 2015701

Sire: Lexinaton

Reg: ESA 20037001 / 840039020037001 Reg: KWPN 97.04525

PARENTAGE ANALYSIS

Liberty ROF FSA 20157011 qualifies as an offspring of Pandora-Tettie KWPN 97.04525 and Lexington FSA 20037001 / 840039020037001.

GENETIC MARKERS

LOCÚS	TYPE	LOCUS	туре	LOCUS	TYPE
AHT4	0	AHT5	JO	AME	YX
ASB17	NR	ASB2	IQ	ASB23	K
HMS2	HK	HMS3	PR	HMS6	LP
HMS7	LO	HTG10	IO	HTG4	KM
IFY3	Ti.	1 5733	0	17177 20	TD



Friesian Sporthorse Association The official registry of the Friesian Sporthors www.FriesianSporthorseAssociation.com

The Veterinary Genetics Laboratory is an institutional member of ISAG. DNA types are reported according to standardized no

Determining Parentage

DNA testing can be used to match the DNA markers of a horse with one or both of its parents. One or both parents of the horse must be known and they must have a DNA marker report of their own already on file with the registry and/or the lab, in order to perform a DNA parentage verification test.

The results are believed to be more than 99% accurate when a horse's DNA is matched with both of its parents. Matching a horse's DNA with only one parent is less conclusive, but can still exclude or qualify that parent depending on the DNA match. (Note: "Qualifying" a parent doesn't guarantee that horse is the parent, especially if only one parent is tested, but it indicates a high likelihood of that horse being the parent and is generally considered to be a verification of parentage.)

One of the common misconceptions about DNA parentage verification is that it can be used to learn the parents of a horse even if they are unknown to the owner. Although it is possible, it's unfortunately not usually that simple. First, the parent must have been DNA tested previously, and many horses registered before DNA was a common requirement were never DNA tested. Second, if the parent has been DNA tested it is necessary to know which registry and which lab did the test, because results are generally not shared between labs and registries. For example, if the parent was DNA tested at VGL the results won't be in the database of UK and vice versa. Likewise, if the parent was tested through one breed registry the results are not automatically shared with all breed registries. In other words, there is not one huge universal database containing the DNA results of all horses ever tested. Sometimes, if a person is lucky, a horse of unknown parentage can be tested with a registry and a match will be made, but in most cases it's like searching for the proverbial needle in a haystack.

Color Testing

DNA testing is also popular to test for a horse's color or to help predict what color foal may be produced from certain pairings. Color testing can be especially helpful in establishing the homozygosity of colors, since this can't be determined simply by looking at a horse and it can play a crucial role in predicting what color foals a horse may produce. Testing for the homozygosity of tobiano (pinto) and black are especially popular. A homozygous pinto horse will always produce a pinto-colored offspring, and a homozygous black horse cannot produce a chestnut-colored offspring and is more likely to produce a black-colored offspring.

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Disease Testing

DNA testing can also be used to screen for fatal and/or life-threatening inherited diseases such as HERDA (Hereditary Equine Regional Dermal Asthenia), GBED (Glycogen Branching Enzyme Deficiency), MH (Malignant Hyperthermia), HYPP (Hyperkalemic Periodic Paralysis) and LFS (Lavender Foal Syndrome). Although HERDA, GBED, MH and HYPP are most common in the stock horse breeds (Quarter Horses and Paints) and LFS is most common in Arabians, the advances in DNA testing for diseases is still noteworthy to all horse enthusiasts, regardless of breed preference. As people introduce stock horse breeds and/or Arabians into their Warmblood breeding programs, these diseases could also begin to infiltrate the Warmblood breeding population.

HERDA is a great example of how priceless DNA testing can be. HERDA is a genetic skin disease which usually won't show symptoms until the horse is around the age of two, and will usually require the horse to be euthanized. By testing both parents before breeding, HERDA can be avoided entirely, because as long as one parent is free of the disease the foal will be free of the disease as well. If both parents are carriers of HERDA, however, there is a 25 percent likelihood the foal will inherit the disease, so this heartbreak can be avoided by DNA testing the parents for the disease prior to breeding in order to not breed two HERDA carriers together.

Sezuan's Surprise

n 2012 the three-year-old stallion Sezuan, who at the time was believed to be by Blue Hors Romanov, was presented at the Danish 10-day stallion performance test. Owned and presented by international rider Andreas Helgstrand, the young stallion was a star at the testing and was granted his breeding license.



As one of the requirements for the Danish breeding approval, Sezuan's DNA had to be tested. The spectacular young stallion turned out to not be sired by Blue Hors Romanov at all, but instead was by the stallion Blue Hors Zack. Although it didn't affect his licensing or approval, the revelation sent the Warmblood breeding world buzzing and helped to bring the discussion of DNA testing to the forefront.

These tests are also surprisingly affordable. DNA testing for each of these diseases listed can be done thru VGL for only \$40 per test.

Breed Verification

Perhaps the biggest misconception is that DNA testing will determine a horse's breed. Although some labs are working on tests to identify DNA markers and associate them with certain breeds, these tests are still very much in the developmental stages and are highly unreliable.

Can I Have My Horse Tested Myself?

Most people have their horse's DNA tested through a registry, especially if it is a requirement for registration, but DNA testing can also be done directly through the lab. VGL and UK both offer step-by-step instructions for setting up an account as an individual, selecting and paying for tests, and submitting samples to the lab.

Moving Forward

In all likelihood it's only a matter of time before all legitimate registries require DNA testing, at least of their breeding stock. And as more and more horses are DNA tested, parentage verification will also become easier and it is also probably only a matter of time before all legitimate registries will require parentage verifications as a part of the registration and/or breeding approval process. Luckily the ease and affordability of DNA testing shouldn't cause much of a hardship for horse owners.

