Testing for Inherited Recessive Genetic Disorders for U.S. Red and Black Wagyu Cattle

A Fact Sheet and Guide for Producers – REVISED 9-13-2022

What is a Recessive Trait?

Many physical traits are associated with variation in the DNA. These variants can be passed from parent to offspring. Because an animal inherits a copy of DNA from each parent, these DNA variants can be transmitted from either the sire or the dam.

A "dominant" trait is an allele that is inherited from a sire or dam that is observable in an animal. Dominance can be complete or partial, with complete dominance appearing as a phenotype that is completely masking the effect of the recessive allele.

A "recessive" trait is one that is only physically observable when a calf inherits a recessive allele from both parents. Note that not all recessive traits are bad or of an economical problem for the animal and some may in fact have a selective advantage. Two copies of the allele are necessary to change the phenotype of that animal. For this reason, an animal with only one copy of the recessive allele will typically not show any sign of the trait. An animal with two copies of the recessive allele will show the trait. A cross between two Carrier parents, between Carrier and Affected parents, or between two Affected parents have the possibility of producing an Affected animal.

Why have these Recessive Traits recently shown up in Wagyu cattle?

The whole genome sequence is expansive, with cattle having about 3 billion basepairs, with each basepair varying by animal to animal and population to population. New variation can be introduced into these populations through mutation or recombination of alleles. Because an animal must inherit two copies of a given recessive allele to be Affected, and with only a few animals typically sharing the same allele in a population, there is rarely a mating cross that has the potential to create Affected offspring under natural selection. Most abnormal recessive traits are never even discovered and disappear in the population through genetic drift.

However, under the selection pressure of most current breeding strategies, and given the relatively small founding population of Wagyu cattle outside of Japan, it is common to utilize a backcross and line breeding with a highly regarded sire line. If the original sire was a Carrier, there is strong likelihood of its generational offspring will also be Carriers. In this case, it creates a higher-than-average frequency of Carriers and potentially Affected cattle in the population. Artificial insemination of a popular sire which is a carrier of a recessive allele can spread this

allele throughout a population much faster than natural breeding conditions. This phenomenon has occurred in the Wagyu breed in Japan, Australia, and the United States

What are the Recessive Disorders?

In the last 10 years, several Recessive Disorders have been shown to be caused by recessive traits in the Wagyu breed:

- Erythrocyte Membrane Protein Band III Deficiency (Spherocytosis): Affected cattle (cattle with two copies of the causative allele) are morbidly anemic. The mutations affect a protein necessary for proper shape and function of red blood cells. Calves are typically born weak and small (40-55 lbs birth weight) with severe anemia, labored breathing, palpitations, and not able to stand or suckle at birth. This disorder is often lethal, but some affected cattle survive to adulthood, although with severely retarded growth.
- Claudin 16 Deficiency (CL16) This recessive disorder causes a buildup of fibrous tissue in the kidneys as well as other tissues. Affected cattle suffer from a severe risk of kidney failure throughout their lives. Other symptoms include growth retardation, increased blood urea nitrogen and creatinine values, diarrhea and overgrowth of hooves. It may or may not be lethal, but affected cattle tend to have atypically short lives.
- Chediak-Higashi Syndrome (CHS) Affected cattle have a deficiency in cells that make up a functional immune system. As a result, these calves are often more susceptible to disease and infection. These cattle may also have a light coat color, and slight coagulation problems (hemorrhaging). This disorder is usually not lethal.
- Bovine Blood Coagulation Factor XIII Deficiency (F13) This disorder is where one of the proteins needed to form blood clots is missing or reduced. Symptoms include severely prolonged bleeding time, bruising from castration/branding, and severe anemia. Death occurs in most cases.
- Factor XI Deficiency (F11): This disorder affects the efficiency of the clotting factor F11. Affected cattle suffer from mild hemophilia-like bleeding tendencies, either spontaneously or following trauma and surgical procedures. It is also possible that Carrier x Carrier mating have increased difficulty producing viable fertilized embryos and full-term pregnancies and are often Repeat Breeders[1]. Normal repeat breeding may be considered 40% with 60% conception being an industry average. It has been reported that factor 11 increased rebreeding by 50% in the Canadian Holstein breed, so now instead of 60% conception we will get 40% conception with 60% of the animals open to be rebred.
- IARS (isoleucyl-tRNA synthetase) is a protein coding gene that has been linked to Weak Calf Syndrome in Wagyu. The disease is responsible for embryonic and perinatal death in Affected calves. Japanese and Australian studies show that it has an estimated incidence rate of 17% in a population of Wagyu cattle, and 0.7% of the population are calculated to be at risk of becoming Affected. Since there is an estimated 56.5% -59.8% mortality rate

for Affected animals, the calculated death rate in a population is 0.4%. The surviving 0.3% generally do not mature to full potential.

[1]Repeat Breeders are Cows that are cycling normally, with no clinical abnormalities, which have failed to conceive after at least two successive inseminations or embryo transfers. From a clinical perspective, there are two types of repeat breeders:

1. Early repeats - Cows that come into heat within 17-24 days after insemination or embryo transfer. In these animals the luteal function has been shorter than normal or typical for the physiological estrus cycle in non-bred cows. In these cows the most probable event is either failure of fertilization (delayed ovulation, poor semen quality etc.) or early embryonic death (delayed ovulation, poor embryo quality, unfavorable uterine environment, precocious luteolysis).
2. Late repeats - Cows that come into heat later than 25 days after insemination or embryo transfer. In these animals the luteal function was maintained for longer than the physiological luteal phase in non-bred cows. Fertilization and initial recognition of pregnancy probably took place but for some reason (inadequate luteal function, inadequate embryo signaling, infectious diseases, induced luteolysis) luteolysis was induced and pregnancy lost.

Genetic Testing Status and Offspring Distribution Predictions

The genetic status of each tested animal will be reported as one of the four following results. Each mating type can be found in Table 1.

Free (F): animal has 2 copies of the dominant allele

Carrier (C): Animal has one copy of the recessive allele

Affected (A): animal has 2 copies of the recessive allele

No Result (NR): DNA sample was rejected and yieled no result

Mating	Offspring Outcome		
	Free	Carrier	Affected
Free x Free	100%	0%	0%
Free x Carrier	50%	50%	0%
Carrier x Carrier	25%	50%	25%
Free x Affected	0%	100%	0%
Carrier x Affected	0%	50%	50%
Affected x Affected	0%	0%	100%

Table 1

Managing Recessive Genetic Disorders in Fullblood, Purebred, and Percentage U.S. Red and Black Wagyu Cattle

For proper herd management it is important for breeders to have an accurate understanding of the status of their cattle with respect to the genetic disorders. Without knowing the Free, Carrier, and/or Affected cattle in a breeder's herd, it is impossible to eliminate or reduce the risk of propagating the disorders in future generations of cattle. Actively addressing these genetic disorders today will pay dividends to both the breeder and collectively for the Wagyu breed.

Methods for managing recessive genetic disorders are breeder specific and depend on the type of cattle operation, i.e., registered, commercial, fullblood, purebred, percentage, seedstock, beef production, etc. Below are some suggested methods for breeders to consider. It should be noted that this list is not all inclusive:

1. Test all animals and remove Affected animals from the herd. Always use Free animals to mate with any Carrier animals remaining in the herd. A commitment must be made to test all offspring from Carrier animals that will remain in the breeding herd. The Carrier rate will be reduced over time in future generations
2. Test all animals and remove all Affected animals and Carrier sires. Use only Free sires in the breeding program going forward. A commitment must be made to test all offspring from Carrier dams that will remain in the breeding herd. This will reduce the Carrier rate the same as in Method 1 above.
3. Test and remove Carrier and Affected animals from the herd. Only use Free animals in the breeding program going forward. No further testing will be required. This Method will ensure a totally Free herd going forward.

4. Test all animals and use Carrier and/or Affected[2] animals ONLY in a terminal breeding program.
5. Test all animals and use Carrier and/or Affected[2] animals as recipients. If a cleanup bull is used, it should be Free. Offspring DNA verified to the cleanup bull must be tested for any animals that will remain in the breeding herd.
6. These are just five examples of management methods that can be used exclusively or in combination.

[2] According to research in the Holstein breed, Affected F11 animals tend to be repeat breeders. It is anticipated that a study will be conducted to determine if there is a correlation between Affected F11 Wagyu and Repeat Breeders.

As discussed above, proper management has a major impact on reducing the frequency of Carrier and Affected animals in a breeder's herd. Table 2 demonstrates the reduction in the Carrier rate in future generations when each generation is mated to a Free Sire. As you can see the Carrier percentage is basically eliminated by the seventh generation and no Affected cattle are in the offspring distribution.

Generation	Offspring Outcome		
	Free	Carrier	Affected
1	50.00%	50.00%	0%
2	75.00%	25.00%	0%
3	87.50%	12.50%	0%
4	93.75%	6.25%	0%
5	96.88%	3.12%	0%
6	98.44%	1.56%	0%
7	99.22%	0.78%	0%

Table 2

However, if a Carrier sire were reintroduced to fourth-generation dams, the fifth-generation offspring would be 48.44% Free 50% Carrier and 1.56% Affected. The inclusion of Carrier parents in a breeding program can offset the hard work of breeding out a genetic disorder.

Testing Policy - Required Testing

All live cattle and genetics sold at Association sanctioned production sales must be tested as well and the results must be published in the sale catalog for all potential buyers to review prior to the sale.

As an exception to Required Testing, if both the sire and dam of the offspring are Free from all of the genetic disorders set out above, the offspring will be exempt from testing and will be given "Free" status by the Association for all genetic disorders.

Publication of Test Results

Test results that were submitted to the AWA will be posted on the DigitalBeef website. Once the implementation is complete the information will be tracked and updated in the database and be available to the public.

Authorized Lab

The following US Based lab is authorized by the American Wagyu Association to conduct testing for the genetic disorders set out in this document:

GeneSeek, a Neogen company
4665 Innovation Drive, (Suite 120)
LINCOLN, NE 68521
Phone: (402) 435-0665 Fax: (402) 435-0664 Website: www.neogen.com

The AWA website has all the information and documents you will need from ordering your sample collector cards to receiving your results.

Important Information

All Wagyu sample orders, and testing is done through GeneSeek. The current storage of Wagyu samples is located at Noegen. The primary purpose of this storage is for extracting parent information (if viable and sufficient quantity) for SNP profiling. The preferred sample type is HAIR and DNA sample collector cards are available through the AWA office and 'How to Collect' instructions are available via the AWA website. Blood, semen, and tissue is also acceptable for DNA testing but maybe subject to surcharge. Please contact the AWA if you have any questions.

PLEASE NOTE that all DNA ordering and payments are handled through the AWA Office. All samples are to be sent directly to: American Wagyu Association, PO Box 1115, Post Falls, ID 83877 and clearly labeled.

Contact Information

Should you have any questions, please contact the AWA Office on (208) 262-8100 or office@wagyu.org

DISCLAIMER - Genetic test results are based on samples provided by breeders. The American Wagyu Association AND the Board of Directors make no statements, representations or warranties about the accuracy or completeness of, any information relating to the status of a particular animal; and, disclaims all responsibility for information and all liability (including without limitation, liability in negligence) for all expenses, losses, damages, and costs you may incur as a result of information being inaccurate or incomplete in any way for any reason