28 Genetics in the Genomics Era

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Objectives

- **Define important genomics concepts and terms.**
- **Discuss the proper use of genomic information for genetic selection.**
- **Discuss the implementation of genomics in National Cattle Evaluation.**
- **Identify genomic technologies impacting the future of beef cattle selection.**

Terminology

Bases - The building blocks of deoxyribonucleic acid (DNA) sequence. Can be either A, T, G or C. Because mammalian genomes are diploid (two copies of each chromosome, one from each parent), bases are typically talked about in terms of base pairs.

Chromosome - A linear (in mammals) structure comprised of packaged DNA. Bovines have 29 pairs of autosomal chromosomes plus the sex chromosomes.

DNA Markers - Mutations at a specific place in the genome, which can be utilized for genomic analyses.

Genome - The sum total of all genetic material in an animal ,including DNA found in the nucleus of a cell. Also may include DNA from the mitochondria (mitochondrial genome).

Heterozygous - The organism possesses two different alleles at the same locus (for example, CT or AB). Heterozygous animals can be referred to as carriers because they "carry" two different forms of the allele.

Homozygous - The organism possesses the same allele (two copies of the same allele) on each chromosome at a locus (for example, CC or TT, AA or BB).

Microsatellite - Repeats of certain pieces of the DNA sequence that can be used as genetic markers. Genotypes are typically expressed as the number of repeats (for example, 123/126). These markers are sometimes used in parentage panels, but their use is declining due to expense and assay difficulty.

Scored - Refers to the act of recording a genotype on an animal.

SNP - Stands for Single Nucleotide Polymorphism. Pronounced "snip." A single base mutation in the DNA sequence that can be used as a genetic marker. For example, one chromosome exhibits an A and another a G at the same position (which would be called an A/G SNP). These types of markers are gaining in popularity due to their ease and decreasing expense to genotype.

SNP chip - Panel of SNP markers used to obtain genotypes on an animal. Common SNP chips used in the beef industry include the 50K, 80K and 150K. Lower density chips also are used by some breed associations.

What is DNA?

Use of genomic technologies in the beef industry is ever-increasing. To understand the value and use of DNA markers, it helps to begin with basics about the genome and DNA. DNA provides the code needed to perform processes within an organism's body both to keep it alive and to perform in an expected way. DNA is simply a sequence of nitrogen bases (coded A, C, T and G) bound together in long strings that are packaged into chromosomes within each cell. All of these chromosomes make up an organism's genome.

There are enough bases (letters) within the cattle genome to fill up approximately 2,000 New York City phone books. Given that, think of the genome as a giant stack of large phone books. Each book is like a chromosome and the entire stack of books is the genome. Inside each of the books, the names and numbers represent the DNA sequence.

Cattle have 29 pairs of autosomal (non-sex determining) chromosomes and one pair of sex chromosomes (either XX for females or XY for males). Cattle inherit one chromosome from each of their parents, so for every place in the genome, there are two different alleles (alternative forms of a gene). This is why genotypes (genetic make-up of an individual) are recorded in pairs (for example, AA, BB or AB). Some regions of the genome are the same in all animals of a species. Regions of the genome that vary between animals

All Web addresses given in this chapter are subject to change. The links to these websites will be updated regularly at the Master Cattleman website at extension.okstate.edu/programs/master-cattleman.html

are mutations, and these mutations can be used as DNA markers. If an animal has two copies of the same mutation (PP or pp), they are homozygous (think homozygous polled or horned). If an animal has two different copies of the mutation (Pp), they are heterozygous (think heterozygous polled).

What are DNA Markers?

There are several different types of DNA markers used in the beef industry today. The oldest type are called microsatellites, and these are small pieces of DNA (a few bases) that are repeated over and over again. These markers would be scored and the genotype recorded as the number of repeats. For example, a heterozygous animal might have a genotype of 123/142, meaning one chromosome has 123 repeats, and the other had 142 repeats. Microsatellite markers often are used in parentage testing, although their use has declined in recent years. Because microsatellites are numbers of repeats, there often are more than two possible alleles at any given location. This makes microsatellites a very powerful tool, but also harder and more expensive to assay. A newer type of marker, SNPs, have increased in popularity because they are simpler and less expensive to genotype than microsatellites. SNPs are a single base change in the DNA sequence and typically only have two possible alleles at any given location. A SNP genotype would look like A/C, or G/T if calling the bases directly, or AA, AB or BB using genetic coding.

Returning to the phone book analogy, think of microsatellites as common last names in the phone book. If examining a phone book, likely the last name Smith or Jones is repeated many times, just as combinations of DNA bases repeated in a microsatellite. SNP markers are similar to finding small typos in the phone books. Only one letter has changed, but it can be identified and tracked within the phone book. When performing a genetic marker test, it is similar to testing each phone book for the errors or repeated names at known positions. Unlike receiving the entire phone book, only some of the "errors" are received. Not every typo or repeat is surveyed, but given those that have been obtained, tests can be performed for characteristics of important regions of the genome.

DNA markers are used in genomic applications just like one would use a marker on the side of a road while taking a long trip. Consider planning a trip across the U.S. from Boston, MA to San Diego, CA, which is about 3,000 miles (Figure 28.1). Since the bovine genome is about 3 billion base pairs, it makes an easy comparison. If there are only 300 road markers (300 SNP markers) for your road trip, There is a marker every 10 miles (10 million bases). If you have 50,000 road markers (like a 50K SNP chip), there is a marker every 316.8 feet (60,000 bases). If given the task of finding an interesting landmark to visit on the trip, it would be much easier to find one with markers every 316.8 feet rather than every 10 miles. SNP markers within the genome work similarly: the more markers you have, generally the easier it is to locate interesting regions of the genome that affect economically important traits in livestock.

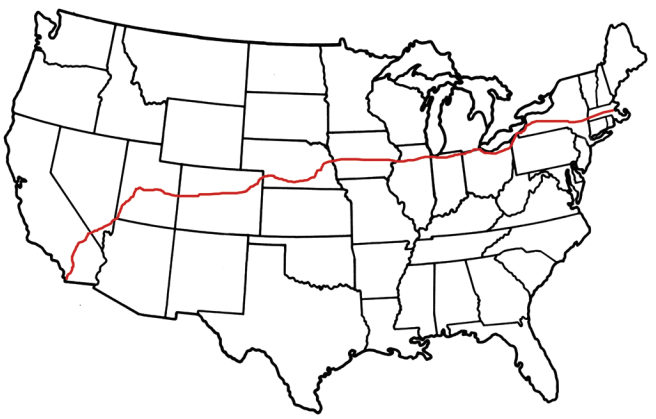

Figure 28.1. Map depicting a trip from Boston, MA to San Diego, CA.

As genotyping prices continue to decrease, these types of tests will become even more affordable and have greater industry acceptance.

Uses of DNA Technologies

There are a variety of options for utilization of genetic testing, including parentage testing; management of genetic abnormalities; testing for simply inherited traits, such as coat color; and panels dedicated to production traits.

Parentage Testing

Parentage testing involves using DNA markers to establish the most probable sire (or dam) for a particular calf. Parentage testing is typically used to exclude potential sires, as dams are typically assumed known. In the past, parentage testing was performed exclusively with a small number of micro-satellite markers. Most organizations are now, or have already transitioned to a larger number of SNP markers for parentage testing because they are abundant, can be automated, have low error rates for genotyping and are easy to standardize between different testing laboratories and organizations.

To more closely examine how parentage testing works, consider this example using SNP genotypes at one locus being used to identify the most likely parent of the calf:

Sire 1	Sire 2	Dam	Calf
A1 A2 A G	A1 A2 G G	A1 A2 G G	A1 A2 G A

Because one allele is inherited from each parent, one can start by examining the dam genotypes. She is homozygous for G, so it is known she passed on a G to the calf. Upon examination of the calf's genotype, it can be seen that the G must have come from the dam, which means the A was inherited from the sire. Sire 1 could have passed on an A allele, but sire 2 could not have, because he is homozygous G. In this case, where dam genotypes are known, and there are a small number of potential sires,

sire 2 can be excluded by using only one locus. However, if the dam genotypes were unknown, exclusion of either sire is not possible. While parents can be excluded using this process, results from parentage testing cannot be used to conclusively prove parentage. Additionally, paternity testing is complicated when close genetic relationships exist between bulls, because they are more likely to share the same marker alleles. Because of this limitation, it is very helpful to avoid using related sires in multi-sire pastures and obtain dam genotypes whenever it is cost effective to do so. Additionally, because this approach only excludes sires, it is vital all potential sires are submitted for testing to avoid an incorrect result. To make this process easier, it may be helpful to collect DNA samples from all sires before turning them out at the beginning of the breeding season.

Parentage testing often is utilized to verify the sires and dams of embryo transfer calves and used frequently to discern between an artificial insemination sire and a herd sire when the calf's birth date is somewhat ambiguous. It also can be utilized in multi-sire pastures to evaluate the efficacy of a sire and manage genetic potential within the herd. A bull's worth in a commercial cattle operation is relative to his ability to sire calves with additional value captured when he sires calves with superior performance. The second objective can be evaluated and selected for using EPDs. However, in multi-sire pastures, the bull's ability to sire calves can be hard to pinpoint without additional information. Genetic testing to establish parentage in multi-sire pastures can be a valuable tool. Knowledge of which bulls are making money, and which are a drain on limited resources is an effective, if not essential, risk management tool.

Genetic Abnormality Testing

In recent years an abundance of genetic defects in a variety of breeds have resulted in substantial associated economic losses. In this section, how genetic defects are inherited, how to determine the probability of producing an affected calf and strategies to decrease issues related to genetic defects in the herd will be discussed.

Inheritance

The cattle genome is made up of 29 pairs of autosomal chromosomes plus the sex chromosomes. Genetic defects are a result of mutations resulting in alleles that cause a lethal condition or severely handicap the performance of an individual. Simply speaking, at any location in the genome where there is a mutation, there are two alleles, which can be thought of as alternate forms of a gene. One allele comes from the sire and the other from the dam. Many mutations within the genome cause no known effects. Alleles of interest are either those which cause small effects on phenotype (the animals' performance or how it looks), typically for performance traits, those which cause large detrimental effects on phenotype, or those that are lethal. Most genetic defects in beef cattle are a result of autosomal recessive mutations. The term autosomal recessive reflects the mode of inheritance for these defects, meaning the mutation is on one of the 29 autosomal chromosomes (not a sex chromosome). These conditions are not expressed as a phenotype unless the animal receives two copies of the damaged allele (one on each of the two chromosomes inherited from its parents). The inheritance of these genetic defects works exactly like the inheritance of horns or red coat color, which also are autosomal recessive conditions and require two copies of the red (or horned) allele before a difference is seen in phenotype. Because these traits are recessive, possessing only one copy of each of these alleles (a defect, red color or horns) is not enough to change phenotype, because it is masked by a dominant allele (normal condition, black color or polled). These animals often are called carriers, because they carry a recessive condition, but do not express it.

In any mammalian genome (the size of most mammalian genomes is 3 billion base pairs), it is almost assured there will be multiple recessive lethal genetic defects present within the DNA sequence. However, because they often are recessive, there must be two copies in the genome (one from each parent) to see affected progeny. In practice, the pairing of two disease alleles rarely occurs when animals are not inbred, because each animal likely carries mutations that cause different conditions. One of the best strategies to avoid incidence of genetic defects is to avoid mating animals to their relatives, or to employ a planned crossbreeding system (but keeping carrier status in mind when mating animals having a common breed composition).

Determination of Risk

When the mode of inheritance is known, the probability of producing an affected calf can be calculated by knowing the carrier status of the parents. Because the normal allele is dominant, there is no way to visually determine which animals are carriers and which are not. The only way to make that determination is through genetic testing or through known parentage of an affected calf.

If the animal tests as normal for a particular genetic defect, it has two normal alleles (represented by NN) and if the animal is tested and determined a carrier, it has one normal allele and one disease allele (represented by Nn). If an animal has ever produced an affected progeny (where the defect can be observed in the phenotype, represented by nn), they can automatically be determined a carrier (Nn) and do not need to undergo genetic testing. If an inherited disease/defect is lethal, all adult animals are either normal or carriers (NN or Nn). They cannot have two copies of the disease allele (nn) and live.

When the carrier status of the parents is known, the probability of producing normal, carrier or affected progeny can be calculated using what is called a Punnett square. It is simply a square with four quadrants and works like a multiplication table. The alleles for each possible parental gamete (sperm and egg) are placed around the outside. The letters are matched on the interior squares and the percentages are multiplied together to determine the probabilities of each status in the progeny. To illustrate this point, consider the following examples:

Example 1: Punnett square for the mating of two normal (NN) animals.

		Male	
	Potential Gametes	*N* 50%	*N* 50%
Female	N 50%	NN 25%	NN 25%
	N 50%	NN 25%	NN 25%

In the example shown above, the two parents tested as normal, and do not carry any disease alleles. Because each gamete will contain one of the two parental gametes, each one will be seen approximately 50% of the time (outside column and row). Both parents are normal, so there are N gametes 100% of the time and all progeny (inside the bolded square) will have all normal alleles. This result illustrates why animals can be declared "clean" through pedigree. If the sire and dam have both been tested and determined carrier-free, any progeny of the mating will be free of disease alleles and do not need to undergo genetic testing.

Example 2: Punnett square for the mating of one normal (NN) and one carrier animal (Nn).

		Male	
	Potential Gametes	N 50%	n 50%
Female	N 50%	NN 25%	Nn 25%
	N 50%	NN 25%	Nn 25%

In Example 2, the mating of a carrier bull to a normal female. The female's gametes will assort the same as in Example 1, and she will always contribute N (normal) alleles to her progeny. The sire, however, can contribute either a normal (N) or disease (n) allele to his progeny. Each allele will occur in approximately 50% of his gametes, meaning one half will be normal and one half will contain the disease allele. When the possible gamete combinations (seen inside the bolded square) are matched, = half of the calves (50%) will have two normal alleles (left side of the bolded box) while the other half of calves will be carriers (Nn - right side of the bolded box). In this scenario, affected calves are not seen, but they do have the potential to transmit the disease alleles to progeny in the next generation.

The only time affected calves are generated is when two carrier animals are mated together, such as illustrated in Example 3. Each sire and dam produces 50% gametes with normal alleles and 50% with disease alleles. When combined together in the progeny (bolded black box) all three genotypic classes can be present. This can be interpreted in terms of each progeny or in terms of a group of progeny produced from a group of carrier sires and dams. In the former, each progeny from the mating would have a 25% chance of having two normal alleles, a 50% chance of being a carrier and a 25% chance of being affected (yellow box). In a group scenario (for example, a large number of full-sib flushmates), 25% of progeny is expected to have two normal alleles, 50% to be carriers and 25% to be affected. Knowing the carrier status of the herd through genetic testing makes effective risk management possible.

Example 3: Punnett square for the mating of two carrier animals (Nn).

		Male	
	Potential Gametes	N 50%	n 50%
Female	N 50%	NN 25%	Nn 25%
	n 50%	Nn 25%	nn 25%

Strategies to Manage Risk of Genetic Defects

Whether or not there is a genetic test for the defect of interest, the incidence of genetic defects will dictate how it can be managed. If there is a test available for the genetic defect, it is advantageous to test those animals which have never had an affected calf, placing special emphasis on testing individuals which have carrier animals in their pedigrees. There is no need to test an animal that has produced an affected progeny, since they are known carriers or one out of two animals that are tested "clean." If an animal is genetically superior in many other ways, but is a carrier of a genetic defect, they do not need to be culled immediately. As observed in the Punnett square examples above, as long as a carrier is always mated to an animal that has two normal alleles (homozygous normal), they will never have affected progeny. However, those progeny will then need to be tested to determine if they are carriers. It is a good risk management practice to test any animals to be retained, such as replacement females, if there is a chance they may be carriers. Alternatively, females do not need to be tested if only non-carrier bulls are utilized. As a general rule, animals scheduled for culling do not need to be tested.

Seedstock producers are encouraged to be cognizant of any breed registry requirements relevant to an animal's carrier status. Some breed associations do not allow carrier animals to be registered, and many will require testing to determine carrier status if there are carriers in the animal's pedigree, provided that a genetic test exists. Often, progeny of animals that are carriers can be registered if the genetic test shows the animal itself is not a carrier. It is much more effective to know these requirements before making decisions regarding replacement females or culling, rather than suffering the consequences later. Breed associations do a very good job of identifying and clearly displaying the

status of carrier animals on the breed association webpages. If there are questions about whether a carrier animal can be registered, contact the breed association for more information.

Commercial cattlemen may find it easier to manage the genetic defect, rather than indiscriminately cull animals. For example, if a known normal (NN) bull is purchased, the carrier status of females may not matter because there will be 100% normal calves with an autosomal recessive inheritance. Alternatively, if using a bull in one breed on cows of another breed in a crossbreeding program, the likelihood of encountering the same defect in two different breeds is extremely low. Because of this, crossbreeding can be an effective risk management strategy for genetic defects. However, it should be noted that care must be taken when keeping replacement females if utilizing bulls of the same breed in subsequent years so no affected calves are produced.

Testing Procedure and Availability

If ordering DNA tests for genetic defects in beef cattle, there are several providers. Contact your provider to obtain information on pricing, sample submission procedures and preferred sample types. If test results are desired to be utilized by a breed association, make sure to follow the association's procedures for sample submission. Often, the sample should be submitted to the breed association office first, rather than submitting samples to the company directly.

Knowing how genetic defects are inherited can assist in making good management decisions, both to preserve genetic progress and reduce or eliminate economic losses associated with genetic defects. In addition, awareness of genetic defect testing and how to calculate the risk of producing affected progeny will help analyze risk and manage the incidence of genetic defects and their associated economic losses in the herd.

DNA Testing for Qualitative Traits

Qualitative traits, also called simply-inherited or Mendelian traits, are those controlled by one or a few genes in the genome. Knowing the genotype of an animal at these loci will often provide the information needed to determine the phenotype. Examples of these types of traits include coat color or horned/polled status. These types of traits are excellent candidates for marker assisted selection. The Punnett squares outlined before will help determine the odds of obtaining a desired phenotype in the same way they can be utilized for risk management of genetic abnormalities. For example, breeding two heterozygous polled animals will yield a 25% chance for either horned or homozygous polled offspring and a 50% chance for producing a heterozygous polled offspring.

DNA Testing for Quantitative Traits

Availability and adoption of genomic technologies has been expanding rapidly in the last few years. A variety of products are now available for many different types of cattle. Modern genomic profiles result from a panel of DNA markers. This genomic information, in tandem with phenotypic performance data collected from within contemporary groups and pedigree information provides values that can be used in the calculation of genomic-enhanced EPDs. Most major U.S. breed associations are utilizing these technologies in their National Cattle Evaluation programs. The Beef Improvement Federation has outlined the position that the value of genomic testing can only be realized when the results are incorporated into National Cattle Evaluation and provided to producers in the form of an EPD and its associated accuracy. These are most commonly called genomic-enhanced EPDs. When both a genomic-enhanced EPD and separate, individual results from a genomic test both exist, selection should ALWAYS be practiced on the genomic-enhanced EPD alone, as it already incorporates performance data, pedigree data and the genomic test. The result of all of this information being incorporated into the genomic-enhanced EPD is an increased accuracy of the EPD, which means increased in confidence in the EPD estimate. These accuracy increases often are expressed in terms of progeny equivalents, which can range widely (some estimates have included anywhere from two to 20 progeny equivalents, although they vary from breed to breed and from one test to another), depending on the heritability of the trait and the amount of genetic variation the test explains. This expression equates the information gained from the genomic test to a specified number of progeny out of the sire or dam which would provide an equal increase in accuracy. To find out the current progeny equivalents for a particular test, consult the specific breed association. It is important to invest in this technology only if a robust test exists for your breed of cattle.

One of the main advantages of genomic testing is the increase in accuracy observed on young animals, which receive genomic-enhanced EPDs. In effect, this shortens the generation interval because more accurate culling decisions can be made earlier in the animal's life. Depending on the trait, genomic-enhanced EPDs on unproven bulls have the same amount of accuracy as if they had already sired 10 to 36 calves, depending on the trait. Examples are shown in Table 28.1.

Best Use of DNA Technology for Production Traits

Potential benefits from genomic selection, which is using high-density genomic testing data for selection decisions (like using genomic-enhanced EPDs), are greatest for traits that:

1. Are lowly heritable (such as fertility and longevity).
2. Are difficult or expensive to measure (such as feed efficiency or disease resistance).
3. Are measured late in life (longevity or stayability).
4. Are not typically measured in breeding animals (carcass traits).
5. Are sex-limited and cannot be measured on that sex of animal (for example, milk production in bulls).

It is important to remember these tips for success:

1. Only use a test designed for the breed of cattle owned (for example, do not use a test designed for Angus cattle on Hereford animals).

Table 28.1. Progeny equivalents (PE) - Carcass trait PE equate to actual carcass harvest data not ultrasound scan equivalents.

Trait	*PE*
Calving ease direct	26
Birth weight	23
Weaning weight	27
Yearling weight	23
Scrotal circumference	15
Dry matter intake	12
Docility	12
Claw angle	10
Foot angle	10
Calving ease maternal	20
Heifer pregnancy	17
Milk	36
Mature weight	15
Carcass weight	15
Carcass marbling	11
Carcass ribeye	17
Carcass fat	14

Source: angus.org Sire Evaluation Report. *Genomics*, 2020

2. Verify the test being considered explains a sufficient portion of genetic variation to provide useful information (check with the breed association to get current information on progeny equivalents to better evaluate potential return on investment).
3. Reward producers who provide the additional information (whether it is genomic data, ultrasound data or whatever data deemed most important) needed to make appropriate selection decisions.
4. Keep in mind the expected additional value from being able to buy animals with increased accuracy at a young age.
5. Whenever confronted with two types of information (for example, genomic enhanced EPDs vs a genomic test result), always choose to select on the metric providing the most comprehensive data on an animal's genetic merit (in the previous examples, the genomic-enhanced EPD should be the primary selection tool.)
6. Carefully consider return on investment in this technology and invest in those products that provide ample return on investment, and pass on using those that do not.

Conclusion

Genomic-enhanced EPDs are the best estimate of an animal's genetic value as a parent, combining all sources of information. Genomics permit higher prediction accuracies for younger animals and characterizes genetics for traits where it is difficult to measure the phenotype.

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